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## Nutricereals Role in Indian Agriculture, Food and Nutritional Security : A Review

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### ABSTRACT

Millets are very important plant genetic resources for agriculture sector that extends food security to poor farmers having arid, marginal and poor lands especially in Asia and Africa. Millets are a group of small grain cereal food crops which are highly nutritious and are grown under marginal/low fertile soils with very low inputs such as fertilizers and pesticides. Indian agriculture is highly dependent on monsoon. Millets are also gaining popularity among farmers as climate-friendly, drought-resistant crops which can thrive even on barren soil. These crops are preferable choice of farmers for cultivation under various adverse environments - prone to climatic extremes. Millets are gluten-free and have a low glycemic index, making them a balanced and healthy diet for people suffering from diabetes. An intensive attempt to include millet crops in cropping systems, especially in vulnerable environments, is a positive step towards long-term sustainability. An effort has been made to expand global and regional scope and target for millet production, usage and in this regard, India's proposal to observe an International Year of Millets in 2023 was approved by the Food and Agriculture Organization (FAO) during 2018 and the United Nations General Assembly has declared the year 2023 as the International Year of Millets. In this esteem, the status of millets and their importance is compiled in this paper.

*Keywords* : Nutricereals, Nutrition, Areas, Production or Millet, Cultivation, Duration, Marketing

MILLETS are being referred as Nutri-cereals are important crops in the country with higher area coverage as compared to wheat and rice before green revolution period. After launching green revolution, the area of nutri-cereals drastically reduced due to shifting of irrigated area from nutri-cereals to more remunerative crops like rice, wheat and sugarcane. At present, Nutri-cereals are grown in resource poor

agro-climatic regions, hilly & tribal areas of the country in rainfed conditions. Nutri-cereals are known for nutri-rich content (Gupta *et al.*, 2017) and having characteristics like drought tolerance, photo-insensitivity and resilient to climate change etc. The millets role can never be overlooked for attaining justifiable means for nutritional safety (Kumar *et al.*, 2018). Nutri-cereals are grown in arid

and semi-arid tracts under low rainfall (300-600 mm) conditions, where cereals like wheat and rice cannot be grown profitably. The abiotic stresses such as drought, salinity and nutrient deficiencies (N, P, K, B and Zn) seems to have lesser impact on the performance of finger millet (Maharajan *et al.*, 2018; Ramakrishnan *et al.*, 2017; Yamunarani *et al.*, 2016). Millets are grown from mean sea level to 2300 m above mean sea level showing their ability under diverse soil and climatic conditions. However, these ignored crops are important by virtue of their role in biodiversity and the means of livelihood of the poor in various parts of the world (Belton & Taylor, 2004). In India, they are seen from Tamil Nadu in the south to Uttarakhand in the North and Gujarat in the West to Arunachal Pradesh in the Northeast (Sukanya *et al.*, 2022). Millets are a group of relatively small cereal grasses that are classed as major millets and minor millets based on grain size. Millets have excellent nutritional value and grow well under diverse situations, but they aren't utilized to their full potential. Sorghum and pearl millet are regarded as major millets while, finger millet, foxtail millet, kodo millet, proso millet, barnyard millet, little millet and browntop millet are named as minor millets. Millets are richer in minerals and vitamins than rice and wheat and have a significant potential to supply food, nutrition, fodder, fiber, health, livelihood and environmental security. Millets have been the first cereal grain to be cultivated for domestic use. Millets are hardy and grow well in rain-fed situation under marginal soil fertility and low moisture. Millets are versatile, climate-resilient crops and can be grown under diverse soil and climatic situations. India is the world's largest producer of millets. In India, millets are cultivated over a total area of 35.71 million hectares, yielding 62.49 million tonnes in 2020 (Anonymous 2020). They are significant in densely populated countries like India and millets can be kept in good condition for many years and hence they are called as famine reserves. Millets are also being used in the animal feed industry and distilleries.

### Global Scenario of Millets

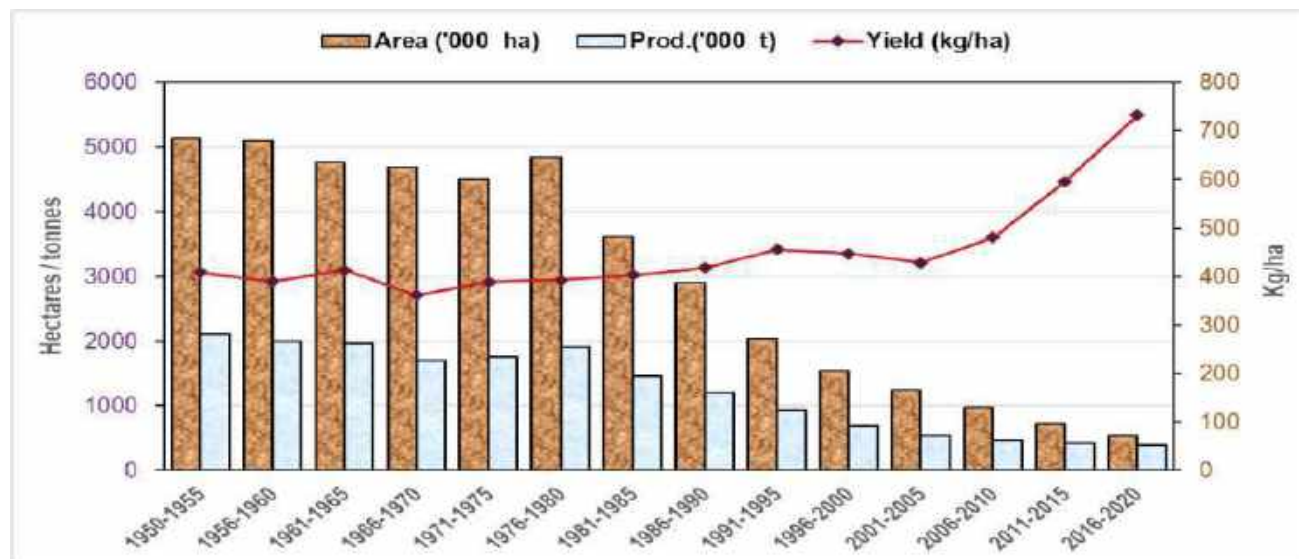
India is one of the important consumers and producers of Nutri-cereals in the world. Group of

crops comprising sorghum (Jowar), pearl millet (Bajra) and small millets *viz.*, finger millet (Ragi/Mandua), little millet (Kutki), kodo millet (Kodo), barnyard millet (Sawa/Jhangora), foxtail millet (Kangni/Kakun), proso millet (Cheena), browntop millet (Makra/murath) all together comes under Millets which are now called as 'Nutri-Cereals' due to their higher nutritive value. The spatial distribution of millets either as a primary crop or as allied crops, is generally determined by the growing habitat and the amount of rainfall received in the region. Small millets are produced in 31.01 million tonnes around the world, according to the FAO, from an area of 33.56 million hectares (Anonymous 2018). The most common millet crops are sorghum and pearl millet, which account for more than 90 per cent of global millets production, followed by finger millet, foxtail millet, proso millet, barnyard, little millet, kodo millet and browntop millet which account for less than 10 per cent of all millets (Anonymous 2020a). In terms of productivity, foxtail millet outstands all the other millets by accounting 2166 kg/ha followed by finger millet (1623 kg ha<sup>-1</sup>), proso millet (1535 kg ha<sup>-1</sup>), sorghum (1426 kg ha<sup>-1</sup>), barnyard millet (1034 kg ha<sup>-1</sup>), pearl millet (850 kg ha<sup>-1</sup>) and little millet (469 kg ha<sup>-1</sup>) (Assocham, 2021).

### Status of Millets in India

India is the world's largest producer of millets. Millets are grown in almost 21 states across the country. Maharashtra, Karnataka and Rajasthan in Jowar; Rajasthan, Uttar Pradesh and Gujarat in Pearl millet; Karnataka, Uttarakhand and Maharashtra in Finger millet; Madhya Pradesh, Chhattisgarh and Uttarakhand in other small millets are three major states where these nutriceals are grown and the area is showing declining trend from previous years although the productivity is towards promising inclination (Anonymous 2020).

In India, Finger millet and other small millets are cultivated in an area of 10.48 and 5.45 lakh hectares (2016-20) with a production of 16.37 and 3.95 lakh tonnes, respectively. In spite of drastic decline in the area in the last six decades, the total production is maintained same to some extent due to the enhanced productivity of millets over the years (Fig.1).



**Fig.1** Area, production and yield of Small millets in India (1950-2020)  
(Source:Ministry of Agriculture and Farmers Welfare, Govt. of India, 2020)

### Nutritional Benefits

Millets are vital in Africa, Asia, China and are extremely nutritious and in some ways, they outperform rice and wheat in terms of presence of key nutrients like phosphorus, potassium, magnesium, manganese, iron and niacin. Protein, fibre, important amino acids like methionine, lecithin and vitamin E are also found in them. Millets have a low glycemic index, suiting them ideal for diabetics (Dayakar Rao *et al.*, 2017). The calcium content of finger millet is nearly ten times that of rice or wheat and proso millet contains about 12 per cent protein. Every millet is far superior to rice and wheat in terms of nutrients and so is the answer to the malnutrition that is affecting bulk of the Indian population. Small seeded grains are produced by these grasses, which are commonly used as cereals. Millets are being demonstrated in recent exploration to provide therapeutic effects, such as controlling asthma, migraine, blood pressure, diabetic heart disease, atherosclerosis and heart attacks, due to their high level of these nutrients. Gallstones are prevented by the fibre in millet. Whole grains of millets, contain health-promoting properties comparable to, if not superior to, fruits. To overcome malnutrition, systematic eating can help to solve the major issue which is prevalent in our Indian population. Millets provide more calcium, iron,

beta-carotene and other nutrients than rice and wheat. Jowar has eight times the fibre of rice, ragi has forty times the calcium of rice and bajra has eight times the iron and five times the riboflavin and folic acid of rice

### Agronomic Advantages

The agronomic practices are critical for accomplishing an assured harvest (Hegde and Krishna Gowda, 2003). Millets are probably the best alternative for farmers who would like to achieve the triple objectives of farming versatility, sustainability and profitability. The advantages of millets-based farming techniques are many as millets are awfully resistant to harsh temperatures, drought and floods. Millets can be grown successfully in dry zones/rain-fed locations with limited soil fertility and moisture. Because of their excellent root system, water requirement of these crops is less in comparison to other crops and have climate resilient traits (Table 1 & 2). Millets need lesser moisture for production and cultivated under rainfed situations or under regimes of lesser rainfall (200-500 mm) (Sukanya *et al.*, 2022; Laxmi Rawat *et al.*, 2022 and Vinod Kumar Singh, 2022). The storage life of grains is relatively long (two years or beyond). Millets growing is a low-cost practice. The majority of the added ingredients are organic and respond very well to integrated nutrient management

TABLE 1  
The climate resilient traits of small millets

Crop	Duration (days)	Climate resilient traits
Finger millet	90-130	Adapted to wide altitude range, moderately resistant to drought, heat stress and humidity
Foxtail millet	70-120	Adapted to high altitude and low rainfall conditions
Proso millet	60-90	Short duration crops, adapted to high altitude and low rainfall conditions
Little millet	70-110	Famine food, adapted to poor soils, low rainfall and can also with stand water logging to some extent
Kodo millet	100-140	Very hard crop with long duration, adapted to low rainfall, poor soils and shows good response to improved agronomic practices
Barnyard millet	75-90	Short duration crop, well adapted to high altitudes and low rainfall conditions

(Source: Assocham, 2021)

TABLE 2  
List of improved varieties released for cultivation in India

State	Varieties
<i>Finger millet</i>	
Andhra Pradesh	VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), VR 847, PR 202, VR 708, VR 762, VR 900, VR 936, Vakula (PPR2700), VR 929, PPR 1012, PR 10-45
Bihar	VL Mandua 379 (VL 379), Arjuna (OEB-526), VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), RAU 8, VL379, OEB 526, OEB 532
Chhattisgarh	Chhattisgarh Ragi-2 (BR-36), Arjuna (OEB-526), VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), VL 324, VL 315, VL 149, Indira Ragi1, Chhattisgarh 2, BR7, GPU 28, PR 202, VR 708 and OEB-526, OEB-532
Gujarat	VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), GNN7, GNN 6, GN 5, GN 4
Jharkhand	VL Mandua 379 (VL 379), VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), A 404, BM 2
Karnataka	DHFM-78-3, Vakula (PPR 2700), Arjuna (OEB-526), VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), GPU 67, GPU 66, GPU 48, GPU 45, GPU 28, PR 202, MR 1, MR 6, Indaf 7, ML365, KMR 340, KMR 301, KMR 204, KMR 360
Madhya Pradesh	VL Mandua 379 (VL 379), VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), GPU 28, PR 202
Maharashtra	VL Mandua 376 (VL 376), Phule Nachani 1 (KOPN 235), KOPLM 83, Dapoli 1, Dapoli 2
Orissa	VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), Arjuna (OEB-526), OEB 10, OUAT 2, BM 9-1, OEB 526, OEB532
Tamil Nadu	VL Mandua 376 (VL 376), Arjuna (OEB-526), GPU 28, CO 15, TNAU 946 (CO 14), CO 13, CO 12, CO 9
Uttarakhand	VL 379, VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), VL 348, VL 324, VL 315, VL 149, VL 146, PES 400, PRM 1, PRM 2, VL 382
<i>Finger millet</i>	
Andhra Pradesh	VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), VR 847, PR 202, VR 708, VR 762, VR 900, VR 936, Vakula (PPR2700), VR 929, PPR 1012, PR 10-45

Table 1 Continued....

State	Varieties
Bihar	VL Mandua 379 (VL 379), Arjuna (OEB-526), VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), RAU 8, VL379, OEB 526, OEB 532
Chhattisgarh	Chhattisgarh Ragi-2 (BR-36), Arjuna (OEB-526), VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), VL 324, VL 315, VL 149, Indira Ragi1, Chhattisgarh 2, BR7, GPU 28, PR 202, VR 708 and OEB-526, OEB-532
Gujarat	VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), GNN7, GNN 6, GN 5, GN 4
Jharkhand	VL Mandua 379 (VL 379), VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), A 404, BM 2
Karnataka	DHFM-78-3, Vakula (PPR 2700), Arjuna (OEB-526), VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), GPU 67, GPU 66, GPU 48, GPU 45, GPU 28, PR 202, MR 1, MR 6, Indaf 7, ML365, KMR 340, KMR 301, KMR 204, KMR 360
Madhya Pradesh	VL Mandua 379 (VL 379), VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), GPU 28, PR 202
Maharashtra	VL Mandua 376 (VL 376), Phule Nachani 1 (KOPN 235), KOPLM 83, Dapoli 1, Dapoli 2
Orissa	VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), Arjuna (OEB-526), OEB 10, OUAT 2, BM 9-1, OEB 526, OEB532
Tamil Nadu	VL Mandua 376 (VL 376), Arjuna (OEB-526), GPU 28, CO 15, TNAU 946 (CO 14), CO 13, CO 12, CO 9
Uttarakhand	VL 379, VL Mandua 376 (VL 376), VL Mandua 352 (VL 352), VL 348, VL 324, VL 315, VL 149, VL 146, PES 400, PRM 1, PRM 2, VL 382
<i>Foxtail millet</i>	
Andhra Pradesh & Telangana	SiA 3088, SiA3156, SiA 3085, Lepakshi, SiA 326, Narasimharaya, Krishnadevaraya, PS 4, SiA 3223
Bihar	RAU-2, SiA 3088, SiA 3156, SiA 3085, PS 4
Karnataka	DHFt 109-3, HMT 100-1, SiA 3156, SiA 3088, SiA 3085, SiA 326, PS 4, Narasimharaya, HN-46
Uttar Pradesh	PRK 1, PS 4, SiA 3088, SiA 3085, Sreelaxmi, Narasimharaya, SiA 326, S-114
Uttarakhand	PS 4, PRK 1, Sreelaxmi, SiA 326, SiA 3156, SiA 3085
Tamilnadu	TNAU 43, TNAU-186, TNAU 196, CO 1, CO 2, CO 4, CO 5, CO (Ten) 7, K2, K3, SiA 3088, SiA 3156, SiA 3085, PS 4, ATL-1
Rajasthan	Prathap Kangani-1 (SR 51), SR 11, SR 16 (Meera), SiA 3085, SiA 3156, PS 4
<i>Little millet</i>	
Andhra Pradesh & Telangana	Chhattisgarh Kutki 1 (BL-6), DHLM 36-3, OLM 203, JK 8, LMV518
Chhattisgarh	Chhattisgarh Kutki 1 (BL-6), Chhattisgarh Kutki 2 (BL-4), JK 8, JK 137, JK 36, DHLM 36-3
Gujarat	GNV-3, Chhattisgarh Kutki 1 (BL-6) GV 2, GV 1, OLM 203, JK 8, DHLM 36-3, DHLM 14-1, LMV 518
Jharkhand	Chhattisgarh Kutki 1 (BL-6), DHLM 36-3, LMV 518
Karnataka	DHLM 36-3, DHLM 14-1, Chhattisgarh Kutki 1 (BL-6), OLM 203, JK 8, LMV 518, BL-41-3
MadhyaPradesh	Chhattisgarh Kutki 1 (BL-6), Jawahar Kutki 4 (JK 4), JK 8, JK 36, JK 137, DHLM 36-3, LMV 518
Maharashtra	Chhattisgarh Kutki 1 (BL-6), Phule Ekadashi (KOPLM 83), JK 8, OLM 203, DHLM 36-3, DHLM 14-1, LMV 518

Table 1 Continued....

State	Varieties
Orissa	Chhattisgarh Kutki 1 (BL-6), OLM 203, OLM 208, OLM 217, DHLM 36-3, DHLM 14-1, LMV 518, OLM 217
Tamil Nadu	Chhattisgarh Kutki 1 (BL-6), DHLM 14-1, DHLM 36-3, Paiyur 2, TNAU 63, CO 3, CO 4, K1, OLM 203, OLM 20, TNPSu177, LMV 518
<i>Proso millet</i>	
Andhra Pradesh & Telangana	TNAU 202, TNAU 164, TNAU 151, Sagar, Nagarjuna, CO 4, CO 3, ATL 1(TNPm 230), GPUP 25
Bihar	ATL 1(TNPm 230), BR 7, TNAU 164, 145, PR 18, TNAU 202, GPUP 25
Chhattisgarh	TNAU 202, GPUP 25
Gujarat	TNAU 202, GPUP 25
Karnataka	ATL 1 (TNPm 230), DHPM-2769, GPUP 8, GPUP 21, TNAU 145, TNAU 151, TNAU 164, TNAU 202
Madhya Pradesh	TNAU 202, GPUP 25
Tamil Nadu	ATL 1 (TNPm 230), Co 5, TNAU 151, TNAU 164, TNAU 145, TNAU 202, CO 4, K 2, CO 3, CO 2, GPUP 21, GPUP 8, GPUP 25
Uttarakhand	PRC 1, TNAU 145, TNAU 164, TNAU 151, GPUP 25
Uttar Pradesh	Bhawna, PRC 1, TNAU 145, TNAU 164, TNAU 151, GPUP 25
<i>Kodo millet</i>	
Andhra Pradesh & Telangana	RK 390-25, TNAU 86, ATL-2, BK-36
Chhattisgarh	Chhattisgarh Kodo-2, Jawahar Kodo 137, RBK 155, Indira Kodo 48, Indira Kodo 1, GPUK 3, JK 439, JK 98, JK 65, Chhattisgarh-2, RK 390-25, TNAU 86, ATL-2, BK36
Gujarat	GK 2, GK 1, GPUK 3, JK 65, JK 13, RK 390-25, GAK-3, ATL-2, BK-36
Karnataka	GPUK 3, RBK 155, RK 390-25, TNAU 86, ATL-2, BK-36
Madhya Pradesh	JK 439, JK 137, JK 106, JK 98, JK 65, JK 48, JK 13, RBK 155, RK 390-25, GPUK 3, DSP 9-1, TNAU 86, ATL-2, BK-36
Tamil Nadu	KMV 20 (Bamban), CO 3, TNAU 86, GPUK 3, RK 390-25, ATL-1, ATL-2, BK-36
<i>Kodo millet</i>	
Andhra Pradesh & Telangana	RK 390-25, TNAU 86, ATL-2, BK-36
Chhattisgarh	Chhattisgarh Kodo-2, Jawahar Kodo 137, RBK 155, Indira Kodo 48, Indira Kodo 1, GPUK 3, JK 439, JK 98, JK 65, Chhattisgarh-2, RK 390-25, TNAU 86, ATL-2, B36
Gujarat	GK 2, GK 1, GPUK 3, JK 65, JK 13, RK 390-25, GAK-3, ATL-2, BK-36
Karnataka	GPUK 3, RBK 155, RK 390-25, TNAU 86, ATL-2, BK-36
Madhya Pradesh	JK 439, JK 137, JK 106, JK 98, JK 65, JK 48, JK 13, RBK 155, RK 390-25, GPUK 3, DSP 9-1, TNAU 86, ATL-2, BK-36
Tamil Nadu	KMV 20 (Bamban), CO 3, TNAU 86, GPUK 3, RK 390-25, ATL-1, ATL-2, BK-36

(Source: Anonymous, 1986 - 2020)

practices (Basavaraja Patil *et al.*, 2022), (Deepti *et al.*, 2022 and Kumar *et al.*, 2003). Millets have a higher number of tillers than other crops. They serve as both food and forage for the animals.

These millets have agronomic advantages *viz.*, highly adapted to low rainfall conditions, able to withstand

fairly long dry spells, recover fast after delayed rain, make them good contingent crops. Millets are highly resilient in adapting to different ecological conditions; ideal crops for climate change and contingency planting. Being C4 plants these are more environment friendly with high water use efficiency and low input

requirement, but equally responsive to high input management

### As Ecofriendly Crops

These have lower requirement of water, chemicals and management interventions for raising. Besides, millets can come up in marginal lands and harsh weather conditions where no other crop can be grown. In India, finger millet farmers realize good yields even with reduced rains and minimum inputs. As these crops are resilient to climate change and provide yield assurance despite environmental risks, they have sustained the onslaught of rice and wheat all these years, despite drastic reduction in cultivation. Another important byproduct of millet cultivation is fodder which is a main source of roughage for cattle in dryland ecosystem. In times of climate change millets are often the last crop standing and thus, are a good risk management strategy for resource-poor marginal farmers. Relatively these crops are less affected by pests and this is a characteristic that comes in very handy when planning a mixed crop cultivated using non-pesticide management techniques. A few rows of millets separating rows of more susceptible leguminous crops are a common practice in farms in different parts of the world.

### Nutricereals as Fodder

Millet crops are critical to the country's overall agricultural development. Because majority of the produce is eaten at the farm/village level, the true worth of their crops and their importance have been overlooked. Food, feed, fodder and nutritional security for a substantial portion of the rural community have

not been recognized so far. Around 33.5 million hectares of nutricereals are cultivated. For finger millet, the dry weight ratio of grain crop residue is about 30:70, while for other small millets, it's about 25:75. Other small millets (Other than Finger millet) contribute 1.2 million tonnes grain and 3.5 million tonnes straw while finger millet only contributes 2.5 million tonnes grain and 5.9 million tonnes straw. The fodder or stover of nutricereals is the most valued fodder source in crop or livestock system where millets are cultivated, regardless of the fact that millet grain is not usually used as animal feed. Millets are appropriate for fragile and vulnerable agro ecosystems despite of environmental friendly nature and are the crops for sustainable and green agriculture. The promotion of millets can lead to much more efficient natural resource management and as a result, to a more holistic approach in preserving precious agro-biodiversity.

### Cultivation of Millets

The millets are often rain-fed crops grown in dryland farming conditions even though they respond well to irrigation. Because they grow well in warm weather and are dependent on rain, cropping is often associated with summer moisture systems like the South Asian monsoons. Fertilizers will increase yield, yet this is often not practiced (Deepthi *et al.*, 2022). Field pests and diseases are not of much concern when compared to other cereal crops but there is a need for weeding. Yet grain yield can be significant with minimal energy relative to more traditional crops (Table 3). Maximum millet cultivation happens in the *kharif* period, *i.e.*, during the monsoon season. In areas that receive more

TABLE 3  
The yield potential of small millets under optimum management practices

Small millet	Harvesting and Yield
Finger millet ( <i>Eleusine coracana</i> L.)	25-35 q/ha of grain and 60-70 q/ha of fodder
Foxtail millet ( <i>Setaria italica</i> L.)	20-25 q/ha of grain and 30-40 q/ha of straw is expected
Little millet ( <i>Panicum sumatrense</i> L.)	18 - 20 q/ha of grain and 25-30 q/ha of straw is expected under well managed agronomic practices

Table 3 Continued....

Small millet	Harvesting and Yield
Proso millet ( <i>Panicum miliaceum L.</i> )	Crop comes to harvest in 65-80 days of sowing in most of the varieties. Harvesting is to be done at physiological maturity and with the adoption of improved package of practices, it is possible to harvest 18-20 q/ha grain and 25-30 q/ha straw under rainfed situation. Under irrigated situation, 20-25 q/ha grain and 50-60 q/ha straw is expected.
Kodo Millet ( <i>Paspalum scrobiculatum L.</i> )	With the adoption of package of practices, kodo millet can yield upto 20-25 q/ha grain and 30-40 q/ha straw
Barnyard millet	The crop should be harvested when it attains the physiological maturity. Generally, it is cut from the ground level with the help of sickles and stacked in the field for about a week before threshing is done by trampling under the feet of bullocks. 15-20 q/ha grain and 25-30 q/ha can be realized by following the improved production practices
Browntop millet ( <i>Brachiaria ramosa L.</i> )	The crop should be harvested as soon as it attains the physiological maturity. 15-20 q/ha grain and 20-25 q/ha straw can be harvested by following the improved production practices.

(Source: Anonymous, 1986 - 2020)

than 800 mm of rains, many of the millets can be cultivated in the second season, *i.e.*, as a *rabi* crop (during the post monsoon, early winter months). And in some places with the right soil and geography, a few millets can even be grown in the third season, during the dark days of winter, utilizing residual moisture in the soil and the dew that precipitates (Sukanya *et al.*, 2022).

### Constraints in Millet Production

Beside its numerous advantages, there are a few drawbacks in millet production that must be addressed. Low productivity of millets, non-availability of good quality seeds, lesser shelf life of millet value-added products, lack of technologies and machinery for primary and secondary processing, dearth of continued demand, lack of awareness on the nutritive value of millets, lesser established market linkages and lack of uniform standards and grades for exporting are the major problems. Further, the expansion of area under millets in non-traditional millet cultivating markets, more value-added products, lack of more ready to cook products are also the issues to be addressed.

### Way Forward and Scope for Small Millets in the Coming Years

Due to growing nutritional awareness and processing technology, millets can be consumed more often as a

food. People are becoming more cognizant of their healthy living habits in order to combat metabolic illnesses and lifestyle diseases, which have resulted in increased demand for various millets. Despite the fact that millet food products are known for their nutritional value, public awareness of their nutritional and therapeutic benefits is gaining. In spite of the fact that millets are recognized to have a rich composition of nutrients and minerals, health branding has not been used effectively to promote millet foods.

Millets have been gaining huge popularity due to their nutritional advantages and these are gluten free. These crops are also eco-friendly, low cost and input consuming crops and found with lower incidence of pests and diseases. More importantly these are crops of dryland where uncertain rainfall, shorter length of growing period (LGP), limited soil moisture, low soil fertility and poor socio-economic conditions are exceedingly observed.

Rich diversity of millet crops has made them well suited for contingency crop planning and also to address the issues of climate change. The farmers who had shifted from millets to other crops are keen to go back to millets in view of the stable harvests ensured, easy crop production, drought resistance and eco-friendly production, provided the assured market is in place.



To develop a millet-based climate smart cropping system, the role of researchers, farmers, policy makers, and rural agro-service providers are essential to achieve success.

Farmers can be motivated through the supply of high-yielding varieties, range of tools such as hand-held crop sensors, rain gauges, decision support tools, leaf color charts, zero-till machinery and improved production options in order to study the crop status in field.

To boost millet production and productivity, expanding the area under millets in non-traditional millet cultivating regions, using good quality seeds, cultivating high yielding varieties of millets, implementing good agricultural practices and encouraging millet cultivation are all important.

To bring consumer-specific millet value-added goods of export grade, extensive research is required. Millets must be mainstreamed in public-sector programmes such as Mid-Day Meals and Integrated Child Development Services to reach the general population and address issues such as lifestyle diseases and malnutrition. Millet promotion must be spread across several states and districts with policy support.

They are also the potential feed stalks for biofuel, a source of renewable and environment friendly raw material for fuel, circumventing the food versus fuel debate. This new demand for millets, leading to higher prices, can make their cultivation profitable, ensuring the legitimate place for millets in the national food basket.

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## Nutrigenomics - A Novel Way of Disease Diagnosis and Prevention

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### ABSTRACT

Nutrigenomics is a study that combines genetic science and nutrition science. The emergence of nutrigenomics as a science opens up new avenues for better identifying individual dietary needs and the effect of food and its components on gene expression, as well as understanding the nutrigenomics - disease interrelationship. Currently, nutrigenomics concentrate on Single Nucleotide Polymorphisms (SNPs) in genes. A SNP is a germline substitution of a single nucleotide at a specific position in the genome. SNPs are the most common type of human genetic variation and a valuable resource for mapping complicated genetic characteristics. The revelation of SNPs enabled us to diagnose numerous health conditions well before symptoms appeared. This can also help dieticians and health workers educate their patients with personalised nutrition and efficiently manage a healthier physique throughout their lifespan. However, because the tests are primarily conducted at the laboratory level, the efficacy and feasibility of this are still in question. As a result, greater research on dietary components using tissue/cell model systems may aid in a better understanding of the interactions between nutrigenetics, nutritional epigenomics, nutritional transcriptomics, proteomics and metabolomics.

**Keywords :** Nutrigenomics, Personalised nutrition, Diabetes, Cancer, Disease diagnosis

EVER since the beginning of time, scientists have been working diligently to understand the etiology of human diseases. In recent decades, scientists could find that not only the nature and nurture of the human beings are the reason for disease conditions. The fact that even the presence of a gene or the mutation of the gene can be a cause of the diseases was recently unveiled. This has led to the development of various researches to understand the relation between genes and health. One such approach led way to the development of a branch of science named 'Nutrigenomics'.

Nutrigenomics is the study of integrating the genomic science and nutrition science. Initially, it was referred to the study of the effect of nutrients in the expression of a genetic makeup. Later on, the study was expanded to understand the nutritional factors which protected the genome from damage.

It is more often termed as personalised nutrition, precision health care as it is a type of customised nutrition therapy that not only helps to combat diseases but also to maintain a healthy life (Mutch *et al.*, 2005).

The advent of nutrigenomics as a science, provides opportunities to better identify individual dietary demands and the effect of food and food components on gene expression, as well as to comprehend the nutrigenomics-disease interrelationship. It studies genetic differences, as well as the effects of epigenetic changes and transcriptome homeostasis on the response to individual bioactive dietary components. This concept is based on the notion that bioactive dietary components can alter the human genome, either directly or indirectly and hence alter gene and gene product expression. As a result of this influence, dietary patterns and specific dietary components may alter many biological

processes, including ageing, as well as the origin, incidence, progression and severity of a variety of diseases. The health impacts of a diet are determined by the balance of health and disease in an individual's genetic makeup.

### History of Nutrigenomics

The history of nutrigenomics begins with the isolation of DNA. In 1869, the Swiss physician Friedrich Miescher, isolated DNA. He was researching the proteins in leucocytes when he discovered a material that was not a protein yet had special properties. Later in 1944, Avery, MacLeod and McCarty revealed that DNA is the hereditary material. Eventually, in 1953, Watson and Crick published the molecular structure of DNA. It was subsequently commercialised by 1980's. The first nutrigenomics company was launched in 1997. Nancy Fogg-Johnson and Alex Merolli renamed nutritional genomics as nutrigenomics in 1999 as it provides a potent technique for uncovering genetic variables in disease. In 2003, the Human Genome Project was launched and the project contained the complete sequencing of the human genome which paved the way for the 'omics revolution' (Priyadarshini *et al.*, 2016).

### Nutrigenomics - the Food Genome Interface

The balance of human health is maintained by the adequate intake of all nutrients in a balanced way and the proper utilisation of these is required by each human being. The deficiencies can be reduced by slightly increasing the quantities and improving the quality of food taken. Eventhough a person's intake of nutrients is sufficient, he may have nutrient deficiencies (Winkels *et al.*, 2008). Hence, it is understood that beyond the intake of food and the metabolism of the food, there are even more factors affecting the occurrence of a disease. Studies have concluded that the problem of deficiency is not only due to over poverty, food choices, conditions or medications which alter nutrient utilisation and malabsorption disorders but also because of the genetic polymorphisms which modify the individual needs (Refsum and Smith, 2008).

Researchers have evidently shown that the macronutrients (fatty acid and proteins), micro nutrients (vitamins), bioactive compounds (phyto chemicals) and certain zoo-chemicals (Eicosapentanoic acid and Decosahexanoic acid) regulate gene expressions. Most of these nutrients are also involved in the metabolic reactions which maintain things from hormonal balance, detoxification and immunity to even the utilization of macro nutrients for energy and growth. Some bioactive compounds in food are ligands for transcription factors and some alter transduction pathways and chromatin structure. These can alter gene expression directly and indirectly. Studies have also pointed out that the deficiency or excess of nutrients plays a major role in determining genome health.

At present, nutrigenomics focuses much on Single Nucleotide polymorphisms (SNPs) of the genes. A Single Nucleotide Polymorphism (SNP) is a germline substitution of a single nucleotide at a particular point in a genome. SNPs are the most abundant form of human genetic variation and a resource for mapping complex genetic traits (Collins *et al.*, 2003). Some of the essential nutrient gene interactions and their clinical manifestations are furnished below (Table 1).

In addition to the essential nutrients, there are several other factors dependent on the genetic expression and disease conditions. Other factors include diet composition, fibre, food structure and antioxidant capacity (Domínguez-Reyes *et al.*, 2015, O'Sullivan *et al.* 2010, Papa athanasopoulos and Camilleri 2009, Puchau *et al.*, 2009), environmental and metabolic regulation, including gut microbiota composition (Vijay-Kumar *et al.*, 2010), prebiotics (Cani *et al.*, 2009), metabolic phenotype (Peppas *et al.*, 2010) and physical activity (Ilanne-Parikka *et al.*, 2010).

Nutrients can affect gene expression *via* different mechanisms: (i) directly; (ii) through their metabolites and (iii) through signal transduction molecules. Nutrients present in food and diet can affect gene expression in a number of ways. They may directly act as ligands for transcription factors and change

TABLE 1  
Essential Nutrient- Gene interaction and their clinical manifestations

Nutrient	Gene polymorphism	Effects on nutrient status	Clinical manifestations	References
Carbohydrates	Beta-2-adrenergic receptors Q27E	Unknown	Higher risk of obesity in female carriers with carbohydrate intake >49% of energy	(Martinez <i>et al.</i> , 2003)
Omega 3 and 6 fatty acids	Fatty acid desaturase, FADS SNP rs174537	Lower plasma arachidonic and eicosapentaenoic acids and higher plasma alpha linolenic and linoleic acids in carriers of the minor allele versus non carriers	The minor allele homozygotes (TT) have lower plasma total cholesterol and LDL-C compared with non carriers	(Tanaka <i>et al.</i> , 2009)
Vitamin A	$\beta$ -carotene 15, 15'-monooxygenase (BCMO1) R267S (rs12934922) and A379V (rs7501331)	Carriers of 267S or 267S + 379V have reduced activity in converting B-carotene to retinal	Increased risk for vitamin A deficiency, when $\beta$ carotene is the major dietary source	(Leung <i>et al.</i> , 2009)
Vitamin D	Vitamin D binding protein DBP-1 (rs7041, exon 11 T>G) and DBP-2 (rs4588, exon 11C>A)	SNPs for DBP-1 and DBP-2 are inversely related to levels of circulating 25 (OH) vit D3 in premenopausal women	Unclear whether carriers would benefit from dietary supplementation or sun exposure	(Sinotte <i>et al.</i> , 2009)
Vitamin K	Vitamin K epoxide reductase complex subunit 1 (VKORC1)j - +2255T>C	Associated with vitamin K recycling, vitamin K-dependent clotting factors and Warfarin resistance	Increased risk of arterial vascular disease such as stroke, coronary heart disease and aortic dissection	(Suh <i>et al.</i> , 2009)
Vitamin B12 (cobalamin)	Methionine synthase TCN2 776C>G and 67A>G	Causes hyperhomocysteinemia	Associated with birth defects	(Brouns <i>et al.</i> , 2008)
Folate	5,10-methylenetetrahydrofolate reductase (MTHFR) 677C>T	Causes a 70% reduction in MTHFR activity, hyperhomocysteinemia and reduced plasma folate concentration	Hyperhomocysteinemia is associated with increased risk of coronary heart disease, neural tube defects, occlusive vascular disease and breast cancer. In carriers, sufficient folate dietary intake decreases risk of colorectal cancer and deficiencies increase risk of colorectal cancer	(Ericson <i>et al.</i> , 2009, Friso and Choi, 2002, Hustad <i>et al.</i> , 2004, Messika <i>et al.</i> , 2010, Simopoulos 2010)
Calcium	Calcium sensing receptor (CASR) A986S	Loss of function for calcium, associated with higher serum calcium and higher urinary calcium excretion	Association with bone mineral density	(Laaksonen <i>et al.</i> , 2009)

Nutrient	Gene polymorphism	Effects on nutrient status	Clinical manifestations	References
Selenium	Missense mutation in selenium binding protein 2 (SBP2)	Causes defective selenocysteine insertion sequence (SECIS)-driven selenocysteine incorporation, downregulates expression of selenoproteins	Defective thyroid function	(Hesketh 2008)
Iron	Human hemochromatosis protein (HFE) 187C>G or 845G>A	Both 187C>G or 845G>A associated with iron overload (hemochromatosis)	Iron overload, liver cirrhosis and cardiomyopathy, especially in diets high in iron	(Hulgan <i>et al.</i> , 2008)
Sodium	Angiotensin gene (AGT) nucleotide -6 G>A	The A substitution in AGT affects the interaction between at least one <i>trans</i> -acting nuclear factor and its promoter, resulting in increased gene transcription and increased angiotensin protein levels	Carriers of the A allele respond to low sodium diets with reductions in blood pressure; GG genotype is not salt-sensitive	(Simopoulos 2010)

gene expression. Nutrients may be metabolized by different pathways, thereby modifying the concentration of substrates or intermediates that affect gene expression. Alternatively, the substrates or intermediates may act on or alter cell signaling pathways involved in gene expression. Moreover, nutrients may directly alter signal transduction pathways responsible for modifications in gene expression. Finally, the modifications in the signaling pathways, caused by nutrients, may modulate the metabolism of nutrients affecting gene expression. The modifications in gene expression may affect muscle, liver, pancreatic  $\beta$  cells, hypothalamus and adipose tissue, thereby regulating glucose homeostasis.

In short, Nutrigenomics involves the characterisation of gene products, their physiological function and interactions. By focusing on the effects of nutrients on genome, proteome, metabolome it explains the relationship between the nutrients and nutrient-regimes on human health.

At the onset, nutrigenomics was concentrating on the genetics of the disease conditions such as obesity, diabetes mellitus and cardio vascular diseases. But advancements in researches, SNPs related to various

other disease conditions like cancer, allergies, periodontitis, non alcoholic liver diseases and many more were identified.

### Nutrigenomics – Role in the Prevention of Diseases

From several studies, it is understood that by examining the interaction of nutrients and their functions in the human body, the genetic changes occurring in them can be identified. This in turn helps to diagnose a disease in advance and thus helps to maintain good health. It is found that if there is a nutrient deficiency persisting in the body; it can lead to certain gene alterations. These gene alterations and the deficiency condition can together lead to certain disease conditions in the future. Some of such nutrient deficiencies, the gene alterations and the disease conditions are furnished below (Table 2).

Nutrigenomics and its involvement in the diagnosis and prevention of various diseases are discussed further.

### Diabetes Mellitus

Diabetes mellitus (DM) is a group of metabolic diseases characterised by hyperglycemia, which results from defects in insulin secretion, insulin

TABLE 2  
Nutrient deficiencies, gene alterations and the disease conditions

Nutrients	Gene Alterations caused by deficiency	Deficiency diet and disease potential	Preventive foods
Protein	Alters gene expression	Kwashiorkor, Marasmus	Egg, milk, soya milk, tofu, yoghurt, cheese, broccoli, almonds, peanuts, cashew, poultry
Fatty acids	Alters gene expression	Obesity, CVD, Diabetes	Salmon, sardines, herring, mackerel, soy oil, sunflower oil, palm oil, flaxseeds, rapeseeds, peanuts, walnuts, almonds, mustard seeds, cloves, oregano, cauliflower, broccoli
Vitamin A (Retinol)	Repression of PEPCK gene	Termination of pregnancy and fetal death	Carrots, spinach, turnip, kale, apricots, Cantaloupe, bell pepper, Papaya, mango, peach, beef liver, chicken liver
Vitamin D (Calciferol)	Prevent gene variation	Colon, breast, prostate cancer	Beef liver, cod liver oils, salmon, mackerel, tuna, egg orange juice, cow milk, yogurt, cheese
Vitamin E (Tocopherols)	Mimics radiation damage	Colon cancer, heart disease, immune dysfunction	Tomato, spinach, broccoli, blueberries, mangoes, kiwi, papaya, almonds, hazelnuts, peanuts, wholegrain cereals & vegetable oils
Vitamin B6 (Pyridoxine)		Cancer, heart disease, brain dysfunction, male infertility, leukemia	Spinach, potato, bell peppers, turnip, mushroom, garlic, cauliflower, banana, chicken, pork, beef, salmon, tuna, turkey
Vitamin B3 (Niacin)	Hampers DNA repair	Nerve problem, memory loss	Pork, tuna, prawns, kidney, liver, poultry, carrots, turnips and celery, mushrooms, beans, almonds, wheat products, rice bran, as well as milk and other dairy products
Vitamin B6 (Pyridoxine)		Cancer, heart disease, brain dysfunction, male infertility, leukemia	Spinach, potato, bell peppers, turnip, mushroom, garlic, cauliflower, banana, chicken, pork, beef, salmon, tuna, turkey
Vitamin B12 (Cobalamin)	Chromosome break and hampers DNA repair/methylation	Cancer, heart disease, brain dysfunction, male infertility, leukemia, memory loss	Liver, sardines, salmon, clam, beef, milk, cheese, yoghurt
Folic acid	Chromosome break and hampers DNA repair/methylation	Cancer, heart disease, brain dysfunction, male infertility, leukemia	Liver, kidney, egg yolk, asparagus, pea, cowpeas, lentils, peanuts, spinach, beetroot, broccoli, orange
Zinc	Chromosome breaks	Brain and immune dysfunction	Oysters, beef, crab, pork, lobster, chicken, spinach, broccoli, cashew nuts, almond, milk, cheese, yogurt
Flavonoids	Alters gene expression	Cancer	Onion, green bean, broccoli, curly kale, endive, celery, cranberry, orange juice, grape fruits, lemons, red, blue and purple berries, peppers, tomatoes and eggplants (Neeha and Kinth, 2013)

activity or both. It is associated with the dysfunction and failure of different organs, such as the blood vessels, heart and kidneys (Georgoulis *et al.*, 2014) and this disease is considered as a global burden (IDF, 2013). Food intake is a key component that affects the incidence of DM. Thus, the identification and analysis of nutrient/gene interactions can assist in understanding the DM etiopathogenesis.

There are several factors which help to identify the incidence of DM. The intake of excess fat and calories can increase the prevalence of diabetes. It was also reported that adiponectin gene polymorphism can contribute to insulin resistance and can cause DM. This is said to aggravate in the persons consuming foods with a high glycemic index (Mohan *et al.*, 2007). Likewise, the sufficient or insufficient intake of many bioactive compounds, amino acids, vitamins and other major or minor nutrients predispose to the exposition of genetic variations related to DM.

The intake of dietary fibre in the diet is interrelated to the microbiota in the body. The activity of this microbiota is highly influential in the absorption of various nutrients. The deficiency caused because of the improper absorption or utilisation of the essential nutrients may cause gene polymorphism. This gene polymorphism may in turn increase the prevalence of diseases like DM (Wu *et al.*, 2011). It is found that apart from the absorption and utilisation of nutrients in the microbiota, the change in the microbial activities may affect the gut immunity. The alterations in gut immunity can precipitate diabetes in DM prone persons.

Studies revealed that not only dietary factors but other environmental factors also influence DM incidence and development (Cornelis *et al.*, 2012 and Lee *et al.*, 2011). These factors primarily include the use of breast milk vs. infant formula (Virtanen and Knip, 2003), highly hydrolyzed infant formula vs. conventional infant formula (Knip *et al.*, 2011), early/late exposure to gluten (Norris *et al.*, 2003) and vitamin D (Hypponen *et al.*, 2001).

Identifying the SNPs related directly and indirectly to these changes in the body can thus diagnose the incidence of DM in people. Scientists working in the field of nutrigenomics have already pointed out several SNPs associated with both Type 1 and Type 2 Diabetes Mellitus.

### Cardiovascular Diseases

Cardiovascular diseases include heart attacks, stroke, hypertension, rheumatic heart disease, congenital heart disease and heart failure (Mozaffarian *et al.*, 2016). Diet and exercise play a major role in controlling these conditions to a great extent. In spite of the advances in the diagnosis and treatments of cardiovascular diseases, there were some lacunae (Nishi *et al.*, 2014 and Wang *et al.*, 2014). These lacunae were addressed to some extent when genetics was employed to identify the gene-disease relationship.

There are several food related factors linked with cardiovascular diseases starting with the increased intake of saturated fatty acids (SFA) and lower intake of dietary fibre. The potential molecular mechanisms for nutrigenomic interactions in CVD risk include (1) Differential intestinal metabolism and uptake of nutrients depending on the gut microbiota, (2) Differential absorption and nutrient binding, depending of the genotype and phenotype, (3) Modulation of gene expression through specific transcription factor binding, (4) Effects on methylation and epigenetic modification and (5) Modulation of metabolic signalling through lipids, metabolites and proteins. The interaction of genetic variants with the environment and specific dietary consumption can alter the overall risk of CVD. There are many genes and SNPs identified which is associated with cardiovascular diseases.

With sufficient knowledge on the connection between certain genetic variants, diet and CVD risk, it may be capable of giving individuals dietary counselling suited to their genotype, hence extending life expectancy and maintaining health (Juma *et al.*, 2014).



## Obesity

The fraction of the global population that is overweight or obese is reaching epidemic proportions, with all of the associated health, social and economic consequences. Although there are several causes for the rise in obesity, the most plausible reason is the modern lifestyle's increased calorie consumption and decreasing exercise. Most people who overeat and are sedentary gain weight but being overweight or obese is a gradual process that takes years of even little excess calorie consumption (Loos and Bouchard, 2003). Although weight reduction is challenging, sustaining weight loss is even more challenging. In fact, just a few non-surgical therapies for obesity succeed in long-term weight loss. Long-term weight loss maintenance necessitates permanent lifestyle changes in exercise and food habits. One such approach paved the way for the inclusion of nutrigenetically personalised diets to such persons. A study conducted by Arkadianos *et al.* (2007) on the patients who strived to reduce their weight revealed that the adoption of nutrigenetically personalized diets resulted in improved long-term BMI reduction. The inclusion of such a diet also helped in maintaining the blood glucose levels in the persons.

Obesity susceptibility is influenced in part by hereditary factors, but 'obesogenic' environment is often required for its phenotypic expression (Martinez, 2014). As a result, while new evidence of genetic influence and neuroendocrine imbalance emerges on a daily basis, it is crucial to analyse a holistic model in which biological and psychological factors interact in a complex manner. As a result of several studies, several genes and SNPs associated with obese phenotypes were discovered (Qi *et al.*, 2014 and Miae and Yangha, 2015). The important genes associated with obesity are the FTO gene, INSIG2 Gene, MC4R gene and APO-A gene. Several SNPs connected with these genes were uncovered and they had an impact on weight loss in a variety of ways (Pena-Romero *et al.*, 2018).

## Cancer

According to WHO, cancer is a vast category of disease that can begin in practically any organ or tissue of the body when abnormal cells develop uncontrollably, invade neighbouring tissues and spread to other organs. The latter phase is known as metastasizing and it is a primary cause of cancer death. Cancer is also known as a neoplasm and a malignant tumour. Globally, cancer is the second most important reason for death. As the incidence of cancer is rising day by day and the mortality increases because of the late diagnosis, an effective preventive strategy has to be developed. In such scenario, cancer is a disease which can be more benefitted with the advent of nutrigenomics. Dietary changes have the potential to be an effective approach of lowering cancer risk. Many studies have pointed out the relationship of nutrients and cancer (Davis, 2007 & Davis and Milner, 2004).

Dietary components are believed to be important predictors of cancer risk in humans. Genetic variations influence absorption and metabolism, resulting in altered response to dietary components. Epigenetic processes can alter DNA methylation patterns, affecting overall gene expression, which can be modified in response to diet components. Many dietary constituents influence post-translational events and may contribute for some of the variances in response to dietary components. Bioactive food components have the potential to influence cellular and molecular activities that are crucial in cancer prevention.

Covalent adducts with individual nucleic acids of DNA or RNA is produced as a result of carcinogen activation. It has also been discovered that reactive oxygen species (ROS) such as superoxide anions, hydrogen peroxide and hydroxyl radicals damage DNA bases, potentially resulting in DNA sequence mistranscription (Bartsch, 1996). Such interruptions can affect DNA replication, resulting in alterations in oncogenes and tumour suppressor genes. ROS can also cause DNA strand breaks, resulting in mutations or deletions of genetic material (Chao and Lipkin, 2006). There are certain DNA repair

mechanisms in the body such as base excision repair, direct repair, nucleotide excision repair and double strand break repair, which helps in maintaining the genome stability which will help in cancer prevention (Cooke *et al.*, 2005 and Sancar *et al.*, 2004). Deficiency in the dietary components such as flavonoids, vitamin E and C, isothiocyanates can disrupt the mechanisms of DNA repair (Frisco and Choi, 2005). The interactions of food with these actions can be clearly estimated with the help of nutrigenomics.

### Other Disease Conditions

Non Alcoholic Fatty liver disease (NAFLD) is a liver disease where the fat accumulation exceeds 5 per cent of the liver weight which is not attributed to alcohol intake (Angulo, 2002). NAFLD develops with a complex interaction between genetic susceptibility and other environmental factors such high calorie diet and physical inactivity (Dongiovanni, 2013). Epigenetic factors such as liver specific DNA methylation and microRNAs, which regulate liver transcriptome also contribute to the NAFLD development and progression. Esterification in the form triglycerides, excess hepatocellular triglycerides, oxidative stress, inflammation triggered by endotoxin, activation of hepatic stellate cells, insulin resistant and altered profile of adipokines are some of the factors which aggravates the disease incidence. There are various genes which are related to these and identifying the genetic variations can thus help in the early diagnosis of the disease condition.

Periodontics is an area where nutrigenomics have intervened very recently. Periodontics is the study of supporting structures of teeth and the disease conditions related to it. Requirand *et al.* (2000) have found out that nutrient factors such as PUFA levels may contribute to bone loss. Zinc deficiency is said to have an increased susceptibility to periodontal disease progression. Disease like DM, insulin resistance can also contribute to the periodontal diseases. The SNPs related to zinc absorption and utilisation is also associated with periodontal

diseases. There are even other factors like this, to be estimated to completely identify the condition.

### Nutrigenomics and Personalised Diet

The identification of certain genes in the body which may be the potent reason for the cause of a specific disease can be a great advent in managing the disease condition as well as in reducing the mortality rates (Sachidanandam *et al.*, 2001 and Potter, 2001). The ultimate aim of personalised diet is to provide a diet by identifying those differences that are due to the heritable genetic sequence variation which can be evidently portrayed by nutrigenomics. The goals of nutrigenomics in determining a personalised diet can be summarized as: 1. Identification of transcription factors (as nutrient targets) and the genes they target; 2. Identification of signaling pathways involved at the cellular level and characterisation of the main dietary signals; 3. Measurement of specific micronutrients and macronutrients inducing cell and organ specific gene expression signatures; 4. Identification of interactions between nutrient related regulatory pathways and pro-inflammatory stress pathways for a better understanding of diet related diseases; 5. Identification of genotypes which can be risk factors for the development of diet related human diseases (such as diabetes, hypertension or atherosclerosis); 6. Use of nutritional systems biology to discover biomarkers for early detection of disease and susceptibility (stress signatures) that are changed in response to diet (Elliott and Ong, 2002; Daniel, 2002 and Ommen and Stierum, 2002).

Nutrigenomics is an upcoming branch of science which reveals the relationship between genes and nutrients. The identification of Single Nucleotide Polymorphism (SNPs) helps us to diagnose the incidence of various disease conditions far before the symptoms are portrayed. This can also help the dieticians and health workers to guide their patients with personalised nutrition and helps to maintain a healthier body throughout the lifetime effectively. But since, the studies are only done at laboratory

levels, the efficiency and practicality of this is yet in vain. Hence, more studies of dietary components employing tissue/cell model systems may aid in a better understanding of the interrelationships between nutrigenetics, nutritional epigenomics, nutritional transcriptomics, proteomics and metabolomics.

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## Tree Diversity and Carbon Sequestration Potential Assessment of Urban Landscapes : A Case Study in Bengaluru City

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### ABSTRACT

Utilizing available land efficiently in the cities helps in climate stabilization through carbon sequestration and conserves biodiversity, apart from providing many other ecosystem benefits. This study investigates the diversity and carbon sequestration in trees that are grown in different landscapes of cities such as residential area, avenue trees, parks, industrial area and around lakes in one of the wards of Bengaluru city. In the present study, 44 tree species belonging to 23 families were recorded among the different landscapes mentioned. Among the five landscapes studied, the highest tree population is noticed among the avenue trees. *Pongamia pinnata* is the most dominant tree species. The residential area had the highest tree biomass and Parks had the lowest biomass assessed from the top ten dominant trees that constituted about 82 per cent of the population. Total carbon sequestered by the trees from the study area is 505 tons and total CO<sub>2</sub> sequestered is 1852 tons. The total amount of carbon stocked in five landscapes varied from 155 tons km<sup>-1</sup> in the industrial area, followed by avenue trees (143 tons km<sup>-1</sup>) and least was found around the lakes (26 tons km<sup>-1</sup>). Thus, maintaining trees in urban areas help in conserving biodiversity and ameliorating climate.

*Keywords:* Tree diversity, Landscape, Biomass, Carbon stock

CITIES are the hubs of economic growth and development. Urban areas contribute close to half of India's gross domestic product today, but rapid urbanization is a major driver of global change, driving land use change, habitat loss, biodiversity loss and epicentres of climate change and pollution. Deteriorating quality of urban ecosystems is a major concern of urban planners. Environmental problems such as air and water contamination and pollution are widespread in urban areas which currently account for 78 per cent of global carbon emissions and 60 per cent of water for domestic use (Shivanand *et al.* 2010). It is therefore essential to take steps to redesign the urban ecosystems to address these environmental problems and to sustain clean air, water and other ecosystem services needed for healthy urban living. In the urbanization process, built-up areas replace the vegetation cover and also increase vehicle movement. These activities are likely

to increase the release of pollutants and greenhouse gases resulting in increased atmospheric temperature, decreased air quality and increased levels of stress for trees and humans (Saini, 2017). Trees absorb carbon from the atmosphere through photosynthesis (Francesco Ferrini, 2011), extracting carbon dioxide from the air, separating the carbon atom from the oxygen atoms and returning oxygen to the atmosphere. In doing so, trees retain a tremendous amount of carbon in their structures which increases periodically with time. Conservation and restoration of urban green spaces comprising of 'urban trees' is therefore an important approach to improve the environmental quality of urban areas.

Urban tree includes trees in gardens, parks and along the streets, roads, canal, residential area *etc.*, which contribute to green space in the city. These spaces provide a variety of ecosystem services such

as improving air quality, buffering noise pollution, biodiversity conservation, mitigating the Urban Heat Island effect, microclimate regulation, stabilization of soil, ground water recharge, prevention of soil erosion and carbon sequestration. Tree canopies provide a cooling effect directly by shading the ground surface and indirectly through transpiration (Scott *et al.* 1999). Studies conducted by several scientists have claimed that urban green spaces can play a very important role in limiting the city's carbon footprint. The vegetation and soil of a green space cannot only sequester carbon, directly contributing to a reduction in atmospheric CO<sub>2</sub> concentration, but also affect the carbon balance indirectly, through their effects on the urban energy balance and thus on CO<sub>2</sub> emissions related to energy use. Urban trees perform important ecological functions in cities by sequestering carbon and reducing automobile pollution. The net carbon emissions that can be reduced by urban tree planting can be up to 18 kg CO<sub>2</sub> per year per tree, equal to the benefits provided by 3 to 5 forest trees of similar size and health (Francesco Ferrini, 2011).

Urban trees face an array of man-made and natural stresses that may lead to the degradation of urban forests and reduce their life spans compared to trees in rural areas or natural stands. Although estimates vary, life spans of trees in downtown areas are often less. One of the important stressors of urban trees is air pollution, which has a negative impact on tree health. Air pollution reduces plant growth and the extent of growth reduction depends on the plant species, concentration and distribution of pollutants and climatic factors. In this background present study is an attempt to assess the diversity of tree species among the different landscapes the urban areas and their contribution towards ameliorating the urban environment and conservation of biodiversity.

#### MATERIAL AND METHODS

In order to study the above factors on a pilot basis the study was conducted in five different landscapes of Ward number four (Yelahanka New town),

Bengaluru, Karnataka. In this region of Bengaluru city five different landscapes namely; Avenue Trees, Residential Area, Industrial area, Lake and Parks were studied. An area of two kilometre stretch of each of the landscapes were sampled while in case of lake, the entire lake was assessed and three parks were enumerated which covers a length of two kilometres. Trees present in these landscapes were identified to their species level and carbon stocks were assessed.

#### 1. Calculation of Alpha Diversity Index

Shannon Alpha Diversity index  $H = -\sum (P_i * \ln P_i)$

Where,

$P_i$  is the proportion of individuals found in species  $i$

$$(P_i = n_i / N)$$

$N$  is the total number of individuals in the community

#### 2. Calculation of Beta Diversity Index

Jaccard  $\beta$  diversity  $C_j = j / (a+b-j)$

Where,

$j$  is the number of species found in both the sites.

$a$  is the number of species in site A.

$b$  is the numbers of species found in site B.

#### 3. Biomass of the Tree (kg)

Standing tree biomass is estimated using the height and girth of the tree to derive trunk volume.

Above ground biomass (dry biomass) = Volume X Specific wood density (Bandana and Sanjay, 2014).

$$\text{Volume} = \pi r^2 h$$

For getting the value of radius from the girth of the tree

$$C = 2\pi r \text{ or } r = C / 2\pi$$

Where,

$C$  = circumference of the tree trunk

$h$  = The height of tree is measured with the help of Blume-Leiss Altimeter.



The estimated volume of the trunk is converted into biomass of individual tree by multiplying with the wood density of tree species.

#### 4. Carbon content in the biomass (kg)

Standing Biomass  $\times$  0.45 (Pearson *et al.*, 2005).

#### 5. CO<sub>2</sub> content of the Biomass

CO<sub>2</sub> = (12 + (16 $\times$ 2)) / 12 = 44 / 12 = 3.67.

One molecule of carbon = 3.67 molecules of CO<sub>2</sub> or  
1 ton of carbon = 3.67 ton of CO<sub>2</sub>

6. Single factor ANOVA is used to know the significant differences of Carbon stock in each species at different landscapes.

### RESULTS AND DISCUSSION

The present study is conducted in the urban landscapes of north Bengaluru. Tree diversity in Industrial area, Avenue Trees, Residential Area, Lake and Parks is presented in Table 1. There were 44 tree species found in the study area belonging to 23 different families. Out of the 44 species highest numbers of species were found in Parks (40) followed by industrial area (27), road sides (25) and along Lakes (18) and the least number of species (16) were found in residential area (Fig. 1). The numbers of trees found in different landscapes are 206, 336, 378, 143 and 291 km<sup>-1</sup> in Industrial area, lake, avenue trees, Residential area and Parks respectively (Table 1). Trees planted as avenue trees recorded the highest number of trees.

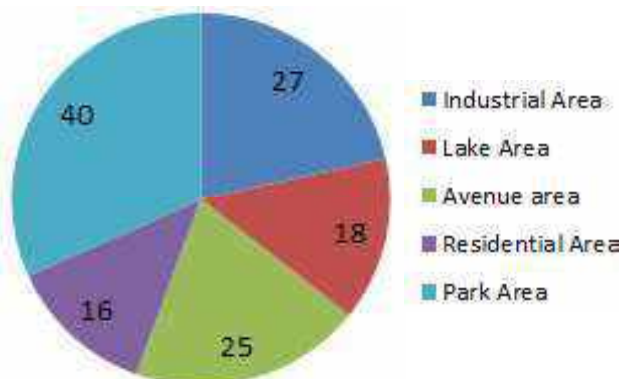


Fig. 1 : Distribution of total tree species (S= 44) in five landscapes studied

Out of 206 trees in Industrial area, the highest number of trees belonged to *S. companulata* (51) followed by *P. pinnata* (47), *P. pterocarpum* (16) and *G. robusta* (10) and one each from *A. scholaris*, *C. lanceolatus*, *D. sissoo* and *F. benghalensis* species were noticed. In case of lakes, a total of 336 trees came from only 18 different trees species among which 114 trees were of *H. Lagenicaulis* and 110 trees belonged to *P. pinnata* while least number is found that of *F. benghalensis*. Similarly in road side planted trees, the highest numbers of trees found were that of *P. pinnata* (163) followed by *P. pterocarpum* (48), *B. purpurea* (40), *S. mahagoni* (28) and *M. champaca* (24). Residential area had least number of trees compared to all other landscapes, in which the highest number of trees found were of *B. purpurea* (24) and *T. rosea* (24) followed by *P. pinnata* (23), *P. pterocarpum* (17) and *C. guianensis* (16) and least number were seen in the species *A. indica* (1), *M. calabura* (1), *T. argentea* (1) and *T. catappa* (1). Similarly in the parks, out of the total 291 trees recorded the highest number of trees were that of *P. pinnata* (27) followed by *G. robusta* (26), *D. Regia* (21), *A. indica* (19) and *L. flosreginae* (19). The least number of trees found were *A. columnaris* (1), *F. religiosa* (1), *A. lebbeck* (2), *F. benghalensis* (2) and *S. saman* (2). Among the five landscapes studied, 30 per cent of the tree population was found among the avenue trees, 25 per cent were found in the lake ecosystem, 21 per cent in the parks, 15 per cent in industrial area and 10 per cent in residential area. The type of species as well as the tree density varied across the landscapes. *P. pinnata* is found to be most dominant species which constitutes about 27 per cent of the total population from all the five landscapes studied. This could be due to the following reasons; *P. pinnata* is leguminous in nature that has ability to assimilate atmospheric nitrogen and thus it helps in not only reducing the nutrient deficiency but also help in reducing the NO<sub>2</sub> emission (which is a greenhouse gas) that comes from inorganic fertilizer application (Usharani *et al.* 2019). Higher population is indicative of higher survival rate and therefore suggests that it has higher stress tolerance (Arjunan *et al.* 1994). The

TABLE 1  
List of tree species and their distribution in five urban landscapes

Name of the species	Family	Number of Individuals per kilometre					Total
		Industrial Area	Lake	Avenue Trees	Residential Area	Parks	
<i>Albizia lebbek</i>	Fabaceae	3	-	4	-	2	9
<i>Alstonia scholaris</i>	Apocynaceae	1	-	-	-	7	8
<i>Anthocephalus cadamba</i>	Rubiaceae	-	-	1	-	-	1
<i>Araucaria columnaris</i>	Araucariaceae	-	-	-	-	1	1
<i>Artocarpus heterophyllus</i>	Moraceae	-	5	3	-	6	14
<i>Azadirachta indica</i>	Meliaceae	3	-	3	1	19	26
<i>Bauhinia purpurea</i>	Fabaceae	7	5	40	24	15	91
<i>Bixa orellana</i>	Bixaceae	-	-	-	-	6	6
<i>Callistemon lanceolatus</i>	Myrtaceae	1	-	2	-	5	8
<i>Cassia spectabilis</i>	Fabaceae	5	-	-	2	-	7
<i>Cocos nucifera</i>	Aracaceae	0	6	6	-	4	16
<i>Couroupita guianensis</i>	Lecythydaceae	1	-	-	16	5	22
<i>Dalbergia sissoo</i>	Fabaceae	1	-	-	-	5	6
<i>Delonix regia</i>	Fabaceae	2	-	1	-	21	24
<i>Eucalyptus globulus</i>	Myrtaceae	-	12	-	-	-	12
<i>Ficus benghalensis</i>	Moraceae	1	1	-	-	2	4
<i>Ficus racemosa</i>	Moraceae	-	5	2	-	3	10
<i>Ficus religiosa</i>	Moraceae	-	2	-	-	1	3
<i>Grevillea robusta</i>	Proteaceae	10	-	2	-	26	38
<i>Hyophorbe lagenicaulis</i>	Aracaceae	-	114	-	-	3	117
<i>Jacaranda mimosifolia</i>	Bignoniaceae	-	-	1	2	9	12
<i>Kigelia pinnata</i>	Bignoniaceae	1	2	-	-	12	15
<i>Lagerstroemia flosreginae</i>	Lythraceae	9	-	-	-	19	28
<i>Mangifera indica</i>	Anacardiaceae	-	-	3	-	4	7
<i>Michelia champaca</i>	Magnoliaceae	3	3	24	13	7	50
<i>Millingtonia hortensis</i>	Bignoniaceae	1	-	2	-	5	8
<i>Muntingia calabura</i>	Muntingiaceae	7	3	0	1	5	16
<i>Peltophorum pterocarpum</i>	Caesalpiniaceae	16	3	48	17	7	91
<i>Phyllanthus emblica</i>	Phyllanthaceae	-	-	-	-	3	3
<i>Plumeria alba</i>	Apocynaceae	3	48	-	-	4	55
<i>Polyalthia longifolia</i>	Annonaceae	-	-	3	-	11	14
<i>Pongamia pinnata</i>	Fabaceae	47	110	163	23	27	370
<i>Samanea saman</i>	Fabaceae	1	-	3	2	2	8
<i>Santalum album</i>	Santalaceae	-	-	-	-	3	3
<i>Saraca asoca</i>	Fabaceae	6	-	5	-	5	16
<i>Schefflera actinophylla</i>	Araliaceae	-	-	-	-	5	5
<i>Spathodea campanulata</i>	Bignoniaceae	51	-	2	4	7	64
<i>Swietenia mahagoni</i>	Meliaceae	5	3	28	5	5	46
<i>Syzygium cumini</i>	Myrtaceae	5	6	-	-	4	15
<i>Tabebuia argentea</i>	Bignoniaceae	-	-	1	1	-	2
<i>Tabebuia rosea</i>	Bignoniaceae	4	2	11	24	5	46
<i>Tectona grandis</i>	Lamiaceae	4	-	1	-	-	5
<i>Terminalia catappa</i>	Combretaceae	-	6	-	1	3	10
<i>Thespesia populnea</i>	Malvaceae	8	-	19	7	8	42
Total		206	336	378	143	291	1354

morphological features of *P. pinnata* such as moderate height, medium size leaflet, lush green canopy (Bohre *et al.* 2014), make them suitable for growing in the cities across the landscapes and also it is easy to manage. Availability of planting material is also another factor. *P. pinnate* is the preferred tree species to increase the production of tree born oil seeds to promote the bio-fuel manufacturing.

For assessing the diversity of tree species present in different land use systems of the city, Shannon alpha diversity index was used to know the richness and evenness of the trees. Typical values of Shannon index vary between 1.5 and 3.5 in most ecological studies. The index values in the present study varied from 1.81 in lake to 3.35 in parks (Table 2). The diversity values depend on both the number of individuals present as well as the number of species. Higher the index, more diverse is the species in the habitats. Hence tree planting in the available lands in the city has helped in conserving biodiversity. From the alpha diversity index, it is observed that the diversity of trees in roadside as well as Residential area did not differ much, while the diversity of the trees in parks was found to be highest.

TABLE 2  
Shannon alpha diversity index

Land Use System	Diversity Index
Industrial Area	2.54
Lake	1.81
Avenue Trees	2.06
Residential Area	2.29
Parks	3.35

Jaccard beta diversity index between Industrial Area and Lake were found to be 0.28 which suggest that there is about 28 per cent similarity between these two landscapes while a value of 0.48 between Industrial Area and avenue landscapes indicates about 48 per cent similar species or 52 per cent of the species are unique to these two landscapes. In between Avenue Trees and Lake about 26 per cent similarity is seen, likewise in Residential Area and Industrial area 54 per cent of the species were unique.

Lake and Residential area have a similarity of up to 29 per cent whereas Residential and Road side trees have a similarity up to 35 per cent. Diversity index between Industrial Area and Park were found to be 0.58 which suggests 58 per cent similarity. Similarly between the lake and park landscapes 57 per cent species were unique. In between Park and Avenue Trees, 53 per cent similar species are seen and in between Park and Residential area 67 per cent species were unique to these two ecosystems (Table 3). This is quite obvious because trees planted in these landscapes are deliberately planted with specific purposes. The trees on the road side are planted mainly with the intention of providing shade and to absorb the pollutants, while in the parks and on the lake bunds are for improving aesthetic value. Higher diversity could help in higher convectional rains in turn contribute in maintaining microclimatic conditions and reduce temperature and the pollution caused due to vehicular movement (Doddabasawa, 2017 and Vailshery *et al.* 2013).

Tree biomass of the ten dominant species in the Industrial area is 309.22 t km<sup>-1</sup> out of 378.85 t km<sup>-1</sup> total biomass of trees present in this landscape per kilometre length. The contribution of top ten tree species to total tree biomass in this landscape was 82 per cent. In case of Lake, 72 per cent (35.92 t km<sup>-1</sup>) of tree biomass is from top ten species present in the ecosystem. Similarly, the ten dominant species in Avenues contain 290.58 t km<sup>-1</sup> biomass out of the total 320.99 t km<sup>-1</sup> and in Residential area, 99 per cent (133.01 t km<sup>-1</sup>) of tree biomass is from top

TABLE 3  
Jaccard Beta Diversity Index in different urban landscapes

Land Use System	Industrial Area	Lake	Avenue Trees	Residential Area	Parks
Industrial Area	–	–	–	–	–
Lake	0.28	–	–	–	–
Avenue Trees	0.48	0.26	–	–	–
Residential Area	0.46	0.29	0.35	–	–
Parks	0.58	0.43	0.53	0.33	–

TABLE 4  
Biomass contribution of dominant ten tree species in five landscapes

Industrial Area		Allalasaandra Lake		Avenue Trees		Residential Area		Park	
Species	Biomass (tons)	Species	Biomass (tons)	Species	Biomass (tons)	Species	Biomass (tons)	Species	Biomass (tons)
<i>Spathodea campanulata</i>	232.94 (75.3%)	<i>Hyophorbe lagenicaulis</i>	6.08 (16.93%)	<i>Pongamia pinnata</i>	71.62 (24.65%)	<i>Bauhinia purpurea</i>	15.54 (11.69%)	<i>Grevillea robusta</i>	25.89 (16.47%)
<i>Pongamia pinnata</i>	15.61 (5.05%)	<i>Pongamia pinnata</i>	1.65 (4.58%)	<i>Peltophorum pterocarpum</i>	101.84 (35.05%)	<i>Tabebuia rosea</i>	32.65 (24.55%)	<i>Delonix regia</i>	25.74 (16.37%)
<i>Peltophorum pterocarpum</i>	31.97 (10.34%)	<i>Plumeria alba</i>	2.60 (7.23%)	<i>Bauhinia purpurea</i>	6.43 (2.21%)	<i>Pongamia pinnata</i>	6.22 (4.67%)	<i>Pongamia pinnata</i>	20.53 (13.06%)
<i>Grevillea robusta</i>	4.90 (1.58%)	<i>Eucalyptus globulus</i>	14.82 (41.29%)	<i>Swietenia mahagoni</i>	43.31 (14.91%)	<i>Peltophorum pterocarpum</i>	19.76 (14.86%)	<i>Lagerstroemia flosreginae</i>	3.00 (1.91%)
<i>Lagerstroemia flosreginae</i>	0.46 (0.15%)	<i>Terminalia catappa</i>	1.76 (4.91%)	<i>Michelia champaca</i>	6.21 (2.14%)	<i>Couroupita guianensis</i>	31.38 (23.59%)	<i>Bauhinia purpurea</i>	3.65 (2.32%)
<i>Thespesia populnea</i>	3.53 (1.14%)	<i>Michelia champaca</i>	2.11 (5.88%)	<i>Thespesia populnea</i>	2.69 (0.93%)	<i>Michelia champaca</i>	2.71 (2.04%)	<i>Azadirachta indica</i>	11.67 (7.42%)
<i>Bauhinia purpurea</i>	15.74 (5.09%)	<i>Cocos nucifera</i>	3.40 (9.47%)	<i>Tabebuia rosea</i>	32.49 (11.18%)	<i>Thespesia populnea</i>	1.43 (1.07%)	<i>Spathodea campanulata</i>	2.76 (1.75%)
<i>Muntingia calabura</i>	0.44 (0.14%)	<i>Bauhinia purpurea</i>	0.90 (2.50%)	<i>Cocos nucifera</i>	4.97 (1.71%)	<i>Swietenia mahagoni</i>	6.00 (4.51%)	<i>Swietenia mahagoni</i>	12.59 (8.01%)
<i>Saraca asoca</i>	0.51 (0.17%)	<i>Michelia champaca</i>	1.24 (3.46%)	<i>Saraca asoca</i>	0.31 (0.11%)	<i>Spathodea campanulata</i>	17.13 (12.88%)	<i>Jacaranda mimosifolia</i>	18.46 (11.74%)
<i>Syzygium cumini</i>	3.12 (1.01%)	<i>Ficus racemosa</i>	1.37 (3.81%)	<i>Albizia lebbbeck</i>	20.70 (7.12%)	<i>Samanea saman</i>	0.19 (0.15%)	<i>Peltophorum pterocarpum</i>	32.95 (20.96%)
Total	309.22 (82%)		35.919 (72%)		290.585 (90%)		133.013 (99%)		157.208 (57%)
Total biomass in each landscape from all species (kg)	378.857		50.385		320.999		134.273		271.525

ten species present in the ecosystem. Similarly in the park 157.21 t km<sup>-1</sup> of biomass found in top ten tree species out of (271.52 t km<sup>-1</sup>) accounting for 57 per cent of biomass from top ten species (Table 4).

Residential area had the highest tree biomass from top ten dominant trees upto 99 per cent. This could be because of presence of greater wood size, higher girth class and older trees. Higher biomass will be contributed by the higher girth class individuals (Hareesh and Nagarajaiah, 2019). Park showed the lowest percent of biomass may be because parks are man-made ecosystem and it is mainly focused on growing trees for ornamental, recreational and aesthetic value. Park had lesser old trees when compared to other landscapes as most of the trees are newly planted and had smaller girth class. The tree size (DBH and Height) in urban parks established in more recent years is less as compared to old parks in Bangalore (Harini and Diya, 2011). Assessment of biomass provides information on the structure and functional attributes of trees. Bigger the size and structure greater will be the biomass. With approximately 50 per cent of dry biomass comprises of carbon (Montagu *et al.* 2005), biomass assessments illustrate the amount of carbon that may be sequestered by trees. Biomass is an important indicator in carbon sequestration therefore estimating the biomass in trees is the first step in carbon accounting.

The mean carbon accumulated in industrial area is highest in *S. campanulata* (104.82 tons) followed by *P. pterocarpum* (14.38 tons), *B. purpurea* (7.08 tons) and the least values were found in *C. lanceolatus* (0.009 tons) followed by *P. alba* (0.11 tons), *D. regia* (0.12 tons). In case of Lake landscape, the mean carbon stock was found to be highest *E. globulus* (6.67 tons) followed by *F. benghalensis* (4.41 tons), *F. religiosa* (2.73 tons) and the least was found in *M. calabura* (0.022 tons) followed by *T. rosea* (0.11 tons), *S. mahagoni* (0.12 tons). The mean carbon accumulated in road side trees were found highest in *P. pterocarpum* (45.82 tons) followed by *P. pinnata* (32.22 tons), *S. mahagoni* (19.49 tons) and the least mean carbon stock found in *T. argentea*

(0.029 tons) and *C. lanceolatus* (0.033 tons). In case of residential area, the mean carbon stock was found highest in *T. rosea* (14.69 tons) followed by *C. guianensis* (14.12 tons), *P. pterocarpum* (8.89 tons) and least was found in *T. argentea* (0.0062 tons) and *C. spectabilis* (0.032 tons). Similarly in case of park ecosystem, the mean carbon stock was found to be highest in *S. campanulata* (15.67 tons) followed by *G. robusta* (11.65 tons), *D. regia* (11.58 tons) and least was in *H. lagenicaulis* (0.0093 tons), *P. alba* (0.11 tons) and *S. asoca* (0.11 tons) (Table 5).

In order to quantify the carbon stocks among the tree species, the total amount of carbon sequestered from each species was computed which varied as follows. Out of the total amount of 505 tons, *S. campanulata* contributed 132 tons; this could be due to higher Diameter at Breast Height and height and higher density of trees, followed by *P. pterocarpum* (73 tons), *P. pinnata* (52 tons). Among the species, the least carbon accumulated species was found to be *T. argentea* (0.03 tons) followed by *P. emblica* (1.29 tons), *S. album* (0.08 tons).

The total amount of carbon stocked in five landscapes varied from 155 tons km<sup>-1</sup> in industrial area, followed by 143 tons km<sup>-1</sup> in avenue trees, 121 tons km<sup>-1</sup> in parks, 61 tons km<sup>-1</sup> in residential area and the least of 26 tons km<sup>-1</sup> was found in the trees around the lake. From these five ecosystems, a total of 505 tons of carbon is stored in the standing biomass of trees which is equal to 1852 tons of carbon dioxide sequestered from atmosphere over years (Fig. 2).

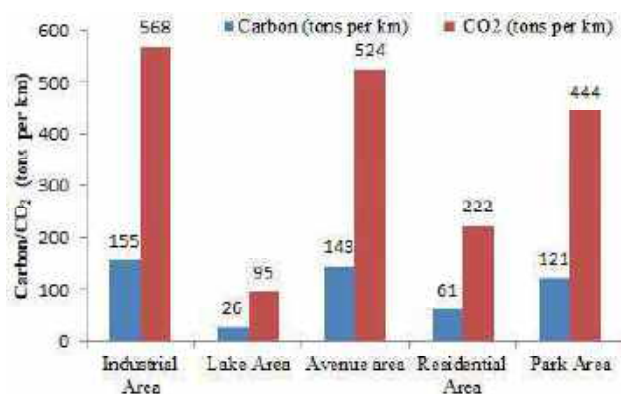


Fig. 2 : Total Carbon stocks and CO<sub>2</sub> in the biomass of standing trees in five landscapes

TABLE 5  
Carbon stock present in each species present in different landscapes

Name of the species	Carbon stock (tons km <sup>-1</sup> )					Total	Mean
	Industrial Area	Allalassandra Lake	Avenue Trees	Residential Area	Parks		
<i>Albizia lebbek</i>	0.40	-	9.31	-	1.66	11.38	2.274 <sup>c</sup>
<i>Alstonia scholaris</i>	0.49	-	-	-	2.98	3.48	0.694 <sup>c</sup>
<i>Anthocephalus cadamba</i>	-	-	1.91	-	0.22	2.13	0.426 <sup>c</sup>
<i>Araucaria columnaris</i>	-	-	-	-	1.44	1.44	0.288 <sup>c</sup>
<i>Artocarpus heterophyllus</i>	-	0.48	0.24	-	0.64	1.37	0.272 <sup>c</sup>
<i>Azadirachta indica</i>	0.14	-	0.17	0.011	0.39	0.72	0.142 <sup>c</sup>
<i>Bauhinia purpurea</i>	7.08	0.41	2.89	6.99	1.62	18.99	3.798 <sup>bc</sup>
<i>Bixa orellana</i>	-	-	-	-	0.15	0.15	0.03 <sup>c</sup>
<i>Callistemon lanceolatus</i>	0.0097	-	0.033	-	0.27	0.32	0.063 <sup>c</sup>
<i>Cassia spectabilis</i>	0.24	-	-	0.032	-	0.27	0.054 <sup>c</sup>
<i>Cocos nucifera</i>	-	1.52	2.23	-	2.53	6.31	1.256 <sup>c</sup>
<i>Couroupita guianensis</i>	1.12	-	-	14.12	7.34	22.59	4.516 <sup>bc</sup>
<i>Dalbergia sissoo</i>	0.96	-	-	-	4.18	5.14	1.028 <sup>c</sup>
<i>Delonix regia</i>	0.12	-	0.24	-	11.58	11.94	2.388 <sup>c</sup>
<i>Eucalyptus globulus</i>	-	6.67	-	-	-	6.67	1.334 <sup>c</sup>
<i>Ficus benghalensis</i>	2.35	4.41	-	-	6.43	13.19	2.638 <sup>c</sup>
<i>Ficus racemosa</i>	-	0.61	1.82	-	3.89	6.33	1.264 <sup>c</sup>
<i>Ficus religiosa</i>	-	2.73	-	-	2.32	5.05	1.01 <sup>c</sup>
<i>Grevillea robusta</i>	2.21	-	0.65	-	11.65	14.50	2.902 <sup>c</sup>
<i>Hyophorbe lagenicaulis</i>	-	2.73	-	-	0.0093	2.74	0.548 <sup>c</sup>
<i>Jacaranda mimosifolia</i>	-	-	0.13	0.32	8.31	8.76	1.752 <sup>c</sup>
<i>Kigelia pinnata</i>	1.51	0.41	-	-	4.31	6.21	1.246 <sup>c</sup>
<i>Lagerstroemia flosreginae</i>	0.21	-	-	-	1.35	1.55	0.312 <sup>c</sup>
<i>Mangifera indica</i>	-	-	0.12	-	0.44	0.56	0.112 <sup>c</sup>
<i>Michelia champaca</i>	0.37	0.59	2.79	1.21	0.88	5.83	1.168 <sup>c</sup>
<i>Millingtonia hortensis</i>	0.59	-	0.55	-	1.15	2.31	0.458 <sup>c</sup>
<i>Muntingia calabura</i>	0.19	0.022	-	0.01	0.12	0.33	0.064 <sup>c</sup>
<i>Peltophorum pterocarpum</i>	14.38	1.35	45.82	8.89	3.07	73.54	14.702 <sup>b</sup>
<i>Phyllanthus emblica</i>	-	-	-	-	0.05	0.05	0.01 <sup>c</sup>
<i>Plumeria alba</i>	0.11	1.16	-	-	0.11	1.39	0.276 <sup>c</sup>
<i>Polyalthia longifolia</i>	-	-	0.33	-	1.08	1.429	0.282 <sup>c</sup>
<i>Pongamia pinnata</i>	7.02	0.74	32.22	2.79	9.24	52.03	10.402 <sup>bc</sup>
<i>Samanea saman</i>	3.69	-	1.31	0.086	6.58	11.67	2.333 <sup>c</sup>
<i>Santalum album</i>	-	-	-	-	0.08	0.08	0.016 <sup>c</sup>
<i>Saraca asoca</i>	0.23	-	0.13	-	0.11	0.48	0.094 <sup>c</sup>
<i>Schefflera actinophylla</i>	-	-	-	-	0.41	0.41	0.082 <sup>c</sup>
<i>Spathodea campanulata</i>	104.82	-	4.25	7.71	15.67	132.45	26.49 <sup>a</sup>
<i>Swietenia mahagoni</i>	1.65	0.12	19.49	2.71	5.66	29.63	5.926 <sup>bc</sup>
<i>Syzygium cumini</i>	1.41	0.94	-	-	1.54	3.89	0.778 <sup>c</sup>
<i>Tabebuia argentea</i>	-	-	0.029	0.0062	-	0.035	0.007 <sup>c</sup>
<i>Tabebuia rosea</i>	0.47	0.11	14.62	14.69	0.32	30.22	6.042 <sup>bc</sup>
<i>Tectona grandis</i>	1.33	-	0.17	-	-	1.509	0.3 <sup>c</sup>
<i>Terminalia catappa</i>	-	0.79	0	0.35	0.24	1.39	0.276 <sup>c</sup>
<i>Thespesia populnea</i>	1.58	-	1.21	0.64	0.68	4.12	0.822 <sup>c</sup>
Total	154.77	25.78	142.75	60.59	120.85	504.74	

SEM = 3.52

CD = 9.83

Note: Alphabets in superscript indicate the significant differences

Industrial area contributed highest towards carbon sequestration compared to all other landscapes because of better growth noticed. The tree species *S. campanulata* contributed more biomass which resulted in more sequestration of carbon and also the presence of more number of trees. *S. campanulata* is often grown as ornamental and a fast-growing tree, even in unfavourable conditions makes the tree survive. Hence, the tree has the ability to survive in Industrial area. Similarly road side planted trees contributed the highest carbon sequestration next to Industrial area, in which *P. pterocarpum* contributed more among other species. It is a large tree that has a dense spreading crown and the flower retains the bright yellow colour even when fallen and also as they dry. So this tree species may be used as a shade tree in addition to its use as an ornamental avenue tree. Therefore, the *P. pterocarpum* trees in road side had bigger size and was good in number which helped in removing of carbon from the atmosphere in the avenue landscape.

The least amount of carbon sequestered from the Lake landscape could be due to low tree diversity and also the presence of less number of larger sized trees. Trees that can be planted in this environment should have ability to tolerate high soil moisture content and even flooding and such characters are generally present in all trees. This could be the reason for less diversity of tree species here (Stoffberg *et al.* 2010).

Urban regions characterized with high human and vehicular density emanate higher GHG's creating heat islands. A city where land is the most constraining input for tree planting should be utilized very diligently. Hence, the landscapes such as roadsides, parks, lake bunds, residential areas and industrial areas should be effectively utilized to grow plants. This approach is one of the most economical way of ameliorating climate as well as conserving biodiversity apart from deriving many other ecosystem benefits.

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## Trait Specific Recombinant Inbred Lines for High Temperature Tolerance in Finger Millet

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### ABSTRACT

Agriculture production and productivity are adversely affected due to increasing episodes of high temperature and drought in the changing climate scenario. Finger millet is relatively a drought-tolerant crop but is sensitive to high temperature because it originated in the cool climate of the highlands in Africa. Identification of varieties for high-temperature adaptation could serve against the rising temperature and considering its nutritional importance, the crop can be extended to the non-traditional high-temperature regions. One of the approaches for identifying a genotype tolerant to high temperature could be by exploiting available genetic resources like germplasm lines. However, the use of a stable mapping population is better in identifying better lines over the better parent. Therefore, a study was conducted using 222 recombinant inbred lines (RIL's;  $F_6$ ) developed for high-temperature tolerance. Differences in temperatures during the crop growth period were achieved by adopting two dates of sowing. The study showed a 1.4 °C rise in temperature during the vegetative to the flowering stage that resulted in a significant decrease in days to flowering, leaf chlorophyll content, the number of productive tillers, mean ear weight, threshing percentage, and grain yield. Correlations showed a significant positive relationship between SCMR, leaf thickness, productive tillers, mean ear weight and threshing percentage with the grain yield under high-temperature condition. Whereas, a significant negative relationship was observed for grain yield with days to flowering and leaf temperature under HT conditions. The principal component analysis (PCA) also showed that grain yield and the number of productive tillers were the most contributing factors to variance. Based on these two traits, significantly HT tolerant RILs over the better parent, PR-202 (PT,  $111.2 \pm 1.22/m^2$  and grain yield,  $263.2 \pm 3.44 g/m^2$ ) are 6.2.5, 6.5.7, 6.3.9, 6.2.25 and 6.19.15, which can be used in crop improvement or for direct cultivation.

**Keywords :** Finger millet, High temperature, Recombinant inbred lines, Principal component analysis

GLOBAL mean surface air temperatures are increased by 0.5 °C during the twentieth century and are expected to rise by 1.4 to 3.1 °C by the end of the twenty-first century (Stocker *et al.*, 2013). Hence, the high temperature (HT) stress could be one of the major limiting factors for crop production in arid and semi-arid regions. For such regions, millets are better suited; particularly the finger millet (*Eleusine coracana* L. Gaertn.) and is an important crop that is

predominantly cultivated in India and Africa. It stands superior amongst cereals and millets, owing to its higher nutritional quality, wider adaptability and subsistence farming. However, high-temperature stress can lead to changes in the plant morphology, physiology and biochemical processes that reduce the plant's growth and development, leading to a loss in grain yield (Sato *et al.*, 2002; Vinay Kumar, 2015 and Yogeesh *et al.*, 2016). In the case of finger millet,

the optimal day and night temperatures for growth and development were reported as 27 to 32 °C and 22 °C, respectively, any increase in temperatures beyond 32 °C affects the flowering and grain filling (Directorate of Millets Development, 2014).

Developing a variety that is tolerant to high temperatures could be of immense help to farmers in high-temperature stress regions. One of the approaches for identifying such lines would be the exploitation of available genetic resources, which is quite challenging and time-consuming (Opole *et al.*, 2018). In this direction, although identification of tolerant lines could be achieved by use of germplasm lines, their availability and time could be of impediment. Therefore, the use of a stable population derived specifically for high-temperature tolerance could be more appropriate. Keeping in mind the future prospects and lacunae in the current scenario, an attempt was made to study the physiology-based field performance within a RIL population of finger millet to identify traits and lines tolerant to HT stress over the better parent.

#### MATERIAL AND METHODS

A field study was carried out at the University of Agricultural Sciences, GKVK, Bengaluru, Karnataka, India using 222 finger millet RILs ( $F_6$ ) developed for high-temperature tolerance. The experiment was conducted in an augmented block design with single replication in 6 blocks with three check varieties (GPU-28, PR-202 and KJNS-46) in each block. The parental lines of this population were PR-202 (high temperature tolerant) and KJNS-46 (high temperature sensitive). Two different dates of sowing were undertaken, the first and second sowing during January 2019 ( $D_1$ /Normal) and February, 2019 ( $D_2$ /high-temperature stress), such that the vegetative to flowering phase of the second sown crop coincided with the months of April and May to have high day temperatures. All the recommended practices for finger millet cultivation were followed to raise a good crop. The leaf temperature, leaf thickness and SCMR were measured at the time of 50 per cent flowering. At the crop maturity, yield attributes like a number of productive tillers (No./m<sup>2</sup>), mean ear head

weight (g/ear) and grain yield (g/m<sup>2</sup>) were measured. Principal component analysis was performed to analyze genetic variability among the RILs and to identify the most important traits contributing to the variability. For the identified traits, the RILs performed superior over the better parent were identified. Data were analyzed in R package, MS excel and Past4 software.

#### RESULTS AND DISCUSSION

In view of achieving differences in temperature during the crop growth period, sowing dates were managed as  $D_1$  (normal) and  $D_2$  (high temperature, HT). In the present study, the mean maximum and minimum ambient temperatures (from sowing to 50 Per cent flowering) were 32.3 °C and 18.6 °C, respectively, for  $D_1$ , and a higher temperature of 33.7 °C and 19.6 °C, respectively, for the  $D_2$  crop. The  $D_2$  sown crop experienced a higher day temperature of 1.4 °C as compared to the  $D_1$  crop (Fig.1), which is expected to reduce the yield attributing traits and grain yield. Plant response for such HT was reported to be highest during flowering and at the early seed filling phase (Djanaguiraman and Prasad, 2014) and genetic variability for HT tolerance exists (Djanaguiraman *et al.*, 2017). In fact, previous research reported that a mean HT of 35.6 °C resulted in a 43.0 per cent decrease in grain yield of finger millet when compared to 32.5°C (Anonymous, 2013). However, finger millet will not be affected considerably up to a temperature of 30 °C during the reproductive phase,

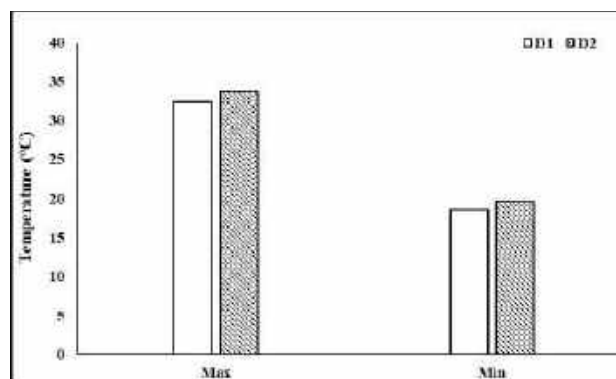


Fig. 1: Mean maximum and minimum temperature from sowing to 50% flowering in January sown ( $D_1$ ) and February sown ( $D_2$ ) crop

above which it significantly decreases the grain yield (Ramya and Nanja Reddy, 2018).

In view of this, it would be pertinent to identify HT tolerant genotypes. The existence of genetic variability is the primary criterion for selection (Opole *et al.*, 2018). Such variability existed in the RIL population for physiological and yield attributes (Table 1) (Djanaguiraman *et al.*, 2017). The RILs differed significantly for all the traits studied both under normal and HT conditions. The variability (mean sum of squares) in the RIL population was higher under HT conditions (D<sub>2</sub>) for days to flowering, leaf temperature, SCMR and productive tillers (Table 1), indicating that individual RILs differ in their response to the HT (Yogeesh *et al.*, 2016; Opole, 2018). The higher variability under HT provides an opportunity for the selection of RILs for a given trait, such as early duration or higher productive tillers per hill, to achieve higher grain yield under HT conditions (Yogeesh *et al.*, 2016). The variability in grain yield of RILs was less under HT than under normal temperatures (Table 1), implying that the potential expression in terms of grain yield did not differ significantly under HT. However, trait selection is possible, which results in a higher grain yield.

There was a considerable variability noticed among RILs for days to flowering, leaf temperature, leaf thickness and SCMR under high-temperature condition which have ultimately resulted in decreased yield attributes and grain yield as compared to the normal conditions (Fig. 2). Research on other cereals, reported that HT affects the photosystem II quantum yield, photosynthetic rate, pigment system and grain yield (Sunoj *et al.*, 2016). The mean leaf temperature and leaf thickness increased under HT conditions as compared to normal conditions (Fig. 2). Under HT conditions, plants might have developed thicker leaves to reduce the loss of water compared to the normal condition.

The extent of the stressful effect of HT on different traits can be studied by the degree of association. Under HT conditions, a significant positive correlation was observed for grain yield with SCMR, leaf

TABLE 1  
Mean sum of squares (ANOVA) for phenology, physiological and yield parameters in finger millet sown during January (D<sub>1</sub>) and February (D<sub>2</sub>), 2019

Source of variation	Df	Day to 50% flowering		Leaf temperature (°C)		SCMR values		Leaf thickness (mm)		Productive tillers (No./ m <sup>2</sup> )		Mean ear head weight (g/ear)		Threshing (%)		Grain yield (g/m <sup>2</sup> )	
		D1	D2	D1	D2	D1	D2	D1	D2	D1	D2	D1	D2	D1	D2	D1	D2
RIL's (Ignoring blocks)	224	2.78**	36.0**	1.69**	2.21**	5.64**	6.3**	0.0006**	0.0004**	360.5**	369.5**	1.73**	1.56**	49.1**	55.7**	34506**	23433**
Checks	2	26.9**	255.2**	5.95**	11.0**	51.0**	6.5**	0.0024**	0.004**	297.1**	365.5**	4.31**	4.82**	758.7**	614.6**	81101**	63144**
RIL's Vs checks	1	6.31**	53.3**	23.4**	18.4**	0.23 ns	2.17**	0.02**	0.02**	10.3 ns	350.5**	0.95**	0.31**	22.6**	20.2 ns	19206**	2954**
RIL's	221	2.4**	34.0**	1.56**	2.05**	5.25**	5.79**	0.0005**	0.0003**	362.6**	369.7**	1.71**	1.54**	246.9**	50.8**	34154**	23166**
Mean		98.8	98.3	27.8	29.3	29.5	27.6	0.30	0.32	117.0	104.4	2.98	2.59	78.4	74.0	294.5	218.6
SE±		0.59	0.6	0.29	0.48	0.44	0.45	0.01	0.01	2.45	4.1	0.08	0.09	1.89	2.73	9.37	8.34
CV		0.48	0.49	0.84	1.32	1.2	1.31	1.63	1.30	1.68	3.14	2.07	2.93	1.93	2.96	2.57	3.07

TABLE 2  
Correlation between phenological, physiological and yield attributes in RIL population of finger millet for heat response in normal (D<sub>1</sub>) and HT (D<sub>2</sub>) conditions

	DFF	L.Temp	SCMR	L.Thick	PT	MEHW	Th%	GY
DFF (D <sub>1</sub> )	1.000							
DFF (D <sub>2</sub> )	1.000							
L.Temp (D <sub>1</sub> )	0.221	1.000						
L.Temp (D <sub>2</sub> )	0.421	1.000						
SCMR (D <sub>1</sub> )	-0.223	-0.533	1.000					
SCMR (D <sub>2</sub> )	-0.496	-0.596	1.000					
L.Thick (D <sub>1</sub> )	-0.135	-0.198	0.362	1.000				
L.Thick (D <sub>2</sub> )	-0.210	-0.259	0.359	1.000				
PT (D <sub>1</sub> )	-0.104	-0.380	0.559	0.345	1.000			
PT (D <sub>2</sub> )	-0.464	-0.453	0.578	0.309	1.000			
MEHW (D <sub>1</sub> )	-0.294	-0.632	0.759	0.415	0.656	1.000		
MEHW (D <sub>2</sub> )	-0.491	-0.667	0.780	0.423	0.617	1.000		
Th% (D <sub>1</sub> )	-0.174	-0.441	0.477	0.253	0.449	0.589	1.000	
Th% (D <sub>2</sub> )	-0.350	-0.437	0.517	0.281	0.454	0.637	1.000	
GY (D <sub>1</sub> )	0.044	-0.006	0.024	-0.007	0.114	-0.017	0.055	1.000
GY (D <sub>2</sub> )	-0.519	-0.639	0.768	0.431	0.751	0.964	0.691	1.000

(DFF: Days to 50% flowering, L.Temp: Leaf temperature (°C), SCMR: SPAD chlorophyll meter reading, L.Thick: leaf thickness, PT: productive tillers (No.m<sup>2</sup>), MEHW: Mean ear head weight (g/ear), Th%: Threshing %, GY: Grain yield (g/m<sup>2</sup>)).

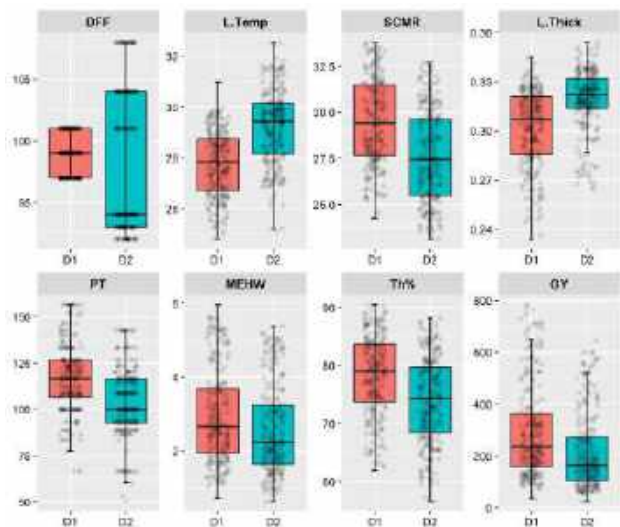
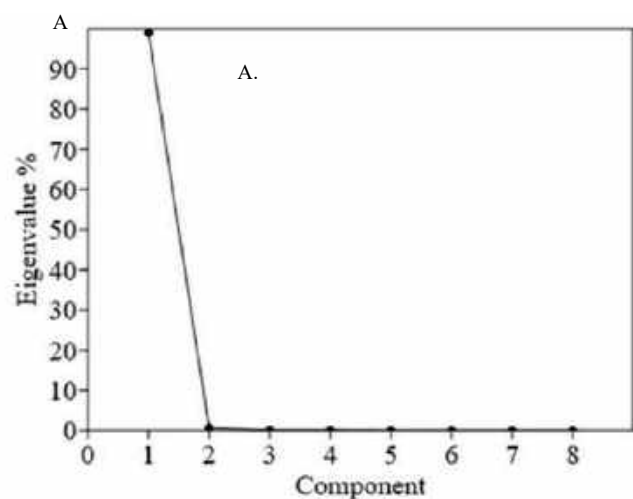


Fig. 2 : Variability in RILs for different traits between January (D<sub>1</sub>) and February (D<sub>2</sub>) sowings (DFF: Days to 50% flowering, L.Temp: Leaf temperature (°C), SCMR: SPAD chlorophyll meter reading, L.Thick: leaf thickness, PT: productive tillers (No.m<sup>2</sup>), MEHW: Mean ear head weight (g/ear), Th%: Threshing %, GY: Grain yield (g/m<sup>2</sup>)). The slate colour dots in boxes indicate the distribution of RIL's, and central line in the given box is the mean for that parameter)

thickness, productive tillers, mean ear-head weight and threshing percentage (Table 2). The relationship between the days to flowering and leaf temperature with grain yield was significantly negative under HT conditions (Table.2; Chaudhari and Acharya, 1969). This suggests that flowering is sensitive to HT (Jukanti



et al., 2017) and short duration lines are preferable for HT conditions.

### Principle Component Analysis

The principal component analysis is one of the best multivariate statistical tools to analyze large populations in order to identify the variability in given traits or genotypes. Principal component-based bi-plots reveals a group of traits, and cluster of genotypes for combining grain yielding potential (Mvuyekure et al., 2018). Scree plot, based on the eigenvalues explains the percentage of variance associated with each principal component (Ladumor et al., 2021). Our results of PCA analysis showed that the contribution for total variance by PC1 (99.09%) was highest followed by PC2 (0.68%; Fig. 3A). Similarly, Ahmad and Mahmood (2015) have reported 99.8 per cent contribution by PC1 and PC2 for total variance. The degree of association of PC1 is strongly associated with grain yield (0.994) and PC2 with productive tiller (0.991). This indicates the number of productive tillers is the most important component of grain yield in finger millet (Chaudhari and Acharya, 1969).

Plotting PC1 against PC2 resulted in the grouping of RILs in 4 classes (quadrant I, II, III and IV) as shown in Fig 3B. RILs in quadrant I recorded positive values of grain yield and negative values of productive tillers.

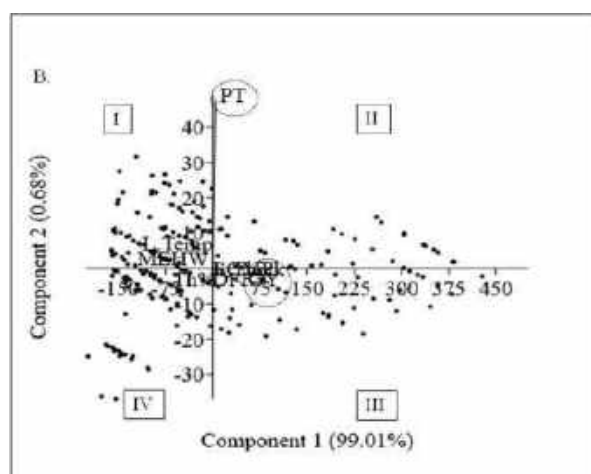


Fig. 3 : Scree plot showing the division of principal components based on eigenvalues (A) and biplot showing the distribution of RILs based on grain yield and productive tillers under high-temperature condition (Component 1 and 2 refers to grain yield and productive tillers respectively, in scree plot)

Similarly, RILs in quadrant II recorded positive values of both grain yield and productive tillers, quadrant III recorded negative values of grain yield and positive values of productive tillers and RILs in quadrant IV recorded negative values of both grain yield and productive tillers. The RILs in quadrants I and IV can be used for grain yield and productive tillers respectively can be utilized in crop improvement by trait specificity. The RILs in quadrant II (Fig 3B) can be considered for direct cultivation. Based on the higher values over the better parent, PR-202 for productive tillers ( $111.2 \pm 1.22/m^2$ ) and grain yield ( $263.2 \pm 3.44 g/m^2$ ), the significantly superior RILs are 6.2.5, 6.5.7, 6.3.9, 6.2.25 and 6.19.25. These identified tolerant RILs can be used for further crop improvement with respect to high-temperature tolerance.

The present investigation on RILs of finger millet revealed a  $1.4^\circ C$  rise in day temperature under HT conditions during the vegetative to flowering stage showed a significant reduction in grain yield and the existence of higher genetic variability among the RILs as compared to the normal condition. The study from principle component analysis showed that the grain yield and productive tiller are the most contributing components to total variance. The RILs performed superior over the better parent, PR-202 for these two traits are 6.2.5, 6.5.7, 6.3.9, 6.2.25 and 6.19.15. These RILs can be used in crop improvement or for direct cultivation under HT conditions.

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## Fungal Endophyte Mediated Salinity Stress Tolerance in Mung Bean (*Vigna radiata* L. Wilczek)

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### ABSTRACT

The soil salinization has led to degradation of agricultural soils that negatively impact crop growth and production. Crops which are sensitive to salinity gets affected severely by soil salinity. The endophyte aid their host to survive in harsh environmental conditions by adapting to environment along with host plant. With this background the study was conducted to evaluate the fungal endophytes isolated from the plants of North western Himalayan region to impart salinity stress tolerance in mung bean. Out of forty-eight fungal endophytes screened four were salt tolerant and they were identified as *Ulocladium* sp., *Fusarium avenaceum*, *Chaetomium* sp. and *F. tricinctum*. The fungal endophytes *Fusarium avenaceum* found superior over other endophytes in imparting salinity stress tolerance in Mung bean. The effect of fungal endophytes on seedling growth was more pronounced under salinity stress condition than in non-stress conditions. Under pot experiment the plants inoculated with *Fusarium avenaceum* recorded higher plant growth, yield and physiological parameters. Thus, the fungus was found as potential endophyte for imparting salinity stress tolerance in mung bean.

**Keywords :** North western Himalaya, Fungal endophytes, Salinity stress, Mung bean, ITS region.

ABIOTIC and biotic stresses substantially reduce the plant growth, vigour, yield and nutritional status. Salt stress is one of the important and widespread abiotic stress, that have visible effects related to various growth parameters of the plants (Gull and Kausar, 2019). It is estimated that by 2050 around 50 per cent of all arable land (~1 billion ha) world wide will be impacted by salinity, which represents about 7 per cent of the earth's continental area. It has been estimated that worldwide 20 per cent of total cultivated and 33 per cent of irrigated agricultural land is affected by high salinity (Shrivastava and Kumar 2014). Salinity affects plants at two levels, initial osmotic effect; increased concentration of salts at soil-root interface enhances osmotic potential that lowers water potential which reduces the ability of the plant to absorb water and

nutrients from the soil, followed by ionic stress when salt accumulation reaches its toxic level (Munns and Tester, 2008).

Endophytes are the organisms, often bacteria or fungi, that invade and thrive in plant tissues without causing any apparent disease symptoms. The shorter generation period of endophytes makes them forerunner in acquiring adaptation to stress under severe selection pressure than their host which are having a lengthy generation period. They also help their host to survive under stress conditions. This is referred to as habitat adapted symbiosis (Rodriguez *et al.*, 2008). Though many efforts are being made to induce plant tolerance to abiotic stress through conventional and molecular approaches, the unconventional approach that is using plant



microbiome is recently gaining attention in research (Ravikanth *et al.*, 2017).

The endophytes have ability to reduce deleterious effect of salt stress on host plant by scavenging reactive oxygen species (ROS) through antioxidant enzymes, ion homeostasis, salt compartmentalisation, accumulation of organic solutes, soluble sugars, proteins, lipids *etc* (Bagheri *et al.*, 2013). Recent studies suggested that, the use of endophytic fungi play a crucial role in plant growth promotion resulting in higher yield and increased resistance to abiotic stresses (Ikram *et al.*, 2019; Manasa *et al.*, 2020; Sampangi-Ramaiah *et al.*, 2020).

Mung bean (*Vigna radiata* L. Wilczek) has been grown during kharif season. Despite the high prices of pulses, large gap between potential and actual yield of pulse crops are lower than the major competing crops. As a result, pulses are mainly grown on marginal land and remote farm areas which are subjected to vagaries of the climate and soils. Growing pulses in such conditions results in large fluctuations in production (Mehandi *et al.*, 2019). Therefore, it necessitates improvement in abiotic stress tolerance in mung bean. In this study, a set of fungal endophytes were examined for their ability to impart salinity stress tolerance in mung bean variety KKM-3 which is sensitive to salt stress.

## MATERIAL AND METHODS

### Collection of Fungal Endophytes

Forty-eight fungal endophytes isolated from North Western Himalaya and conserved in the School of Ecology and Conservation Laboratory, Department of Crop Physiology, University of Agricultural Sciences, GKVK, Bengaluru were collected and rejuvenated by sub-culturing on Potato Dextrose Agar (PDA). These fungi were used for screening against salinity stress.

### Screening of Fungal Endophytes for Salinity Stress Tolerance

The hyphal disc (0.9 mm diameter) of 5 days old fungal cultures was inoculated on PDA supplemented with different concentrations of sodium chloride

(0.5, 1.0, 1.5, 2.0 and 2.5 M) and the control was maintained without addition of sodium chloride. After 5 days of incubation at  $30 \pm 2$  °C, the radial growth of fungal mycelia was recorded by measuring its colony diameter (cm). The Lethal NaCl concentration at which colony growth was reduced to 50 per cent over the respective control (LC50) was calculated by probit analysis (Bekker *et al.*, 2006). The fungal endophytes having 50 per cent growth reduction at higher NaCl concentration and capable to grow at 2.5 M NaCl were selected as salt tolerant.

### Standardization of Salinity Stress Tolerance in Mung Bean

The uniform sized mung bean seeds (KKM-3) were surface sterilized (Arnold *et al.*, 2000) and germinated. The salinity stress was imposed by soaking in the germination papers at different concentration of NaCl solutions (25, 50, 75, 100, 125, 150, 175 and 200 mM) for 30 minutes and then pre-germinated seeds were placed on the germination paper and incubated at 25°C in growth chamber. Sterile water was used for control. Seedling length was recorded after 6 days and LC50 value for NaCl concentration was determined by probit analysis (Bekker *et al.*, 2006).

### In-vitro Evaluation of Selected Fungal Endophytes Against Salt Stress in Mung Bean

The mycelial suspension of 5 days old selected fungal isolates was prepared in sterile distilled water (Dhingra and Sinclair, 1995). The inoculum load (propagules) was adjusted to  $\sim 10^4$  CFU/ml using micrometry by diluting with sterile distilled water. The pre-germinated mung bean seeds were incubated with fungal suspension for 3h for effective colonization followed by washing with sterile water to remove fungal hyphal bits on seed surface. The inoculated pre-germinated seeds were placed on set of germination papers soaked in 112 mM NaCl solution (LC50 value) for 30 min to induce salinity stress. Control was maintained using sterile distilled water. Shoot and root length of seedlings were recorded after 6 days (Sangamesh *et al.*, 2018).

## Confirmation of Fungal Endophytes in Inoculated Seedlings

The leaf, stem and root were cut into small bits and surface sterilized (Arnold *et al.*, 2000). These plant excises were placed on PDA medium and incubated at 30 °C for 3 days. The emerged fungal colonies were sub-cultured and confirmed whether or not they are same by comparing with respective mother culture.

## Identification of Selected Endophytes

The four fungal isolates were identified based on the morphological characters (colony, fruiting body and spore characters) followed by molecular approach using ITS region sequence. The genomic DNA was extracted from the fungal mycelium using the cetyl trimethyl ammonium bromide (CTAB) method. The ITS region was amplified by using universal primers, ITS<sub>1</sub> and ITS<sub>4</sub> (White *et al.*, 1990). The sequences obtained were searched for homology using NCBI BLAST program (<http://blast.ncbi.nlm.nih.gov>; default parameters). The identification of the fungal endophytes was done based on maximum score and query coverage in the BLAST results (Manasa *et al.*, 2015).

## Evaluation of Fungal Endophytes on Mung Bean Against Salinity Stress Under Greenhouse Conditions

The selected fungal endophytes inoculated mung bean seeds were transferred to main pots. The soil physico-chemical properties like soil pH, electrical conductivity (dS/m), available K<sub>2</sub>O (kg /ha) and exchangeable Na<sup>+</sup> (meq/L) were analysed by using standard protocol prior to the experiment. The salts needed to impose salinity was calculated and the plants were subjected to salt stress of 4 dS/m during grand growth stage (*i.e.*, 24 days after sowing) by following Karnal method (Tomar and Minhas, 2004) for 20 days. The Complete Randomized Design (CRD) was used for the pot experiment with five replication and two plants in each replication. Observations for growth, yield and physiological parameters were recorded at different intervals.

## Statistical Analysis

The experiment was conducted using completely randomized design (CRD). The data obtained was statistically analysed using WASP: 2.0 (Web Agri Stat Package 2) statistical tool ([www.icargoa.res.in/wasp2/index.php](http://www.icargoa.res.in/wasp2/index.php)) and means were separated by Duncan Multiple Range Test (DMRT).

## RESULTS AND DISCUSSION

### Screening of Fungal Endophytes Isolates to Salinity Stress Tolerance

Among the 48 fungal endophytes screened for salinity stress tolerance, four isolates (P-82, P-39, P-31 and P-10) were found saline tolerant (Fig 1). However, *Fusarium avenaceum* found superior over other endophytes. The colony growth of fungi reduced as the salt concentration increased. This indicated that the limit of salt tolerance of the different isolates. Shoaib *et al.* (2018) reported that the two primary mechanisms of fungal tolerance to high salt concentrations; first is osmotic effect involves accumulation inorganic (potassium cations) osmolytes and organic (proline and glycine betaine) osmolytes which is high energy demanding process that can result in reduced mycelial growth. There fore, the increase in fungal growth at low solute concentration might be due to selective accumulation of solute to counter the increase in osmotic pressure. Second one is specific ion effect, with increased

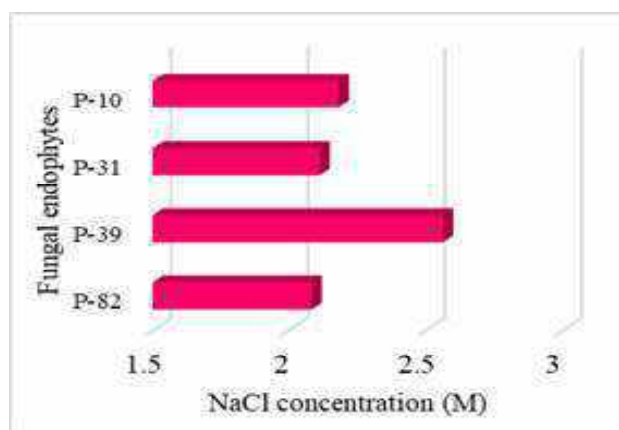


Fig. 1 : LC50 value and radial growth of the salt tolerant fungal isolates

concentration of solute with an increase in osmotic potential, results in exosmosis and can affect fungal growth through plasmolysis, but these salt tolerant fungi can regulate the osmotic stress. Therefore, such four fungi were selected for further evaluation and characterization.

### Identification of the Four-Salt Tolerant Fungal Isolates

Internal transcribed spacer (ITS) region of 18S ribosomal gene has highest probability of successful identification for the broadest range of fungi with the most clearly defined gap between inter and intra specific variations (Manasa *et al.*, 2015) and therefore, in the present study the salt tolerant fungal endophytes were identified using ITS region sequence homology. Based on ITS sequence homology the four isolates were identified as *Ulocladium* sp. (P-82), *Fusarium avenaceum* (P-39), *Chaetomium* sp. (P-31) and *F. tricinctum* (P-10).

### Evaluation of Salinity Stress Tolerance in Mung Bean

The seedling length of mung bean decreased with increased concentration of NaCl (Fig.2). The lethal concentration of NaCl for 50 per cent growth reduction (LC<sub>50</sub>) was found to be 112 mM. The reduced seedling length may be attributed to high salt concentration which increases osmotic potential. In turn lower water potential reduces the ability of plant to absorb water and nutrients that lead to disruption of the cell growth

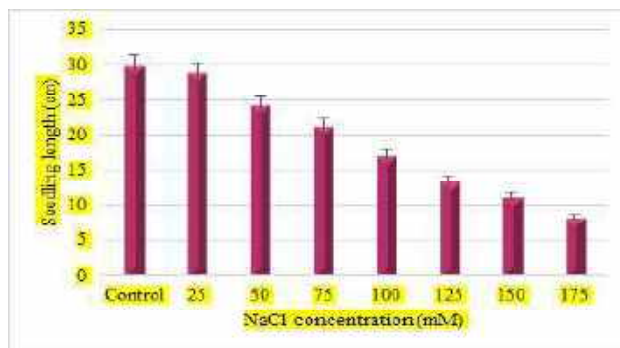


Fig. 2 : Effect of different concentrations of NaCl on seedling growth of mung bean

and development (Munns and Tester, 2008). Similar results were also recorded by Ali *et al.* (2021) in mung bean.

### In-vitro Evaluation of Salt Stress Tolerant Endophytic Fungal Isolates to Impart Salt Stress Tolerance to Mung Bean.

Under salinity stress, the salt tolerant fungal endophyte *Fusarium avenaceum* inoculated plants recorded higher shoot and root length that significantly differ from un-inoculated seedlings (Fig 3). Increased plant growth of endophyte inoculated plants may be attributed to maintaining low Na<sup>+</sup> : K<sup>+</sup> ratio, upregulation of host stress responsive gene and synthesis of host stress responsive proteins and hormones (Manasa *et al.*, 2020). Since the endophytes are isolated from harsh habitat and their effect was pronounced under salinity stress compared to non-stress plants. Ali *et al.* (2021) reported that the

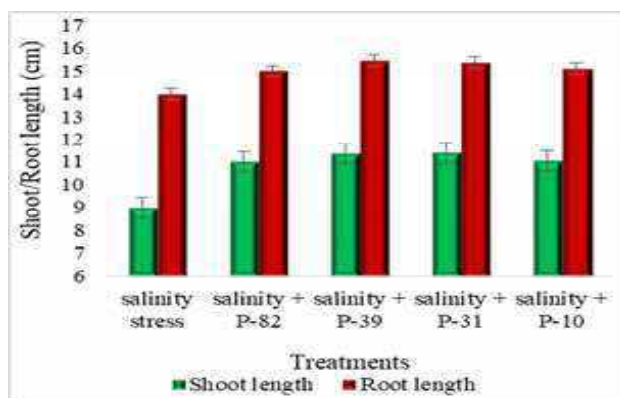
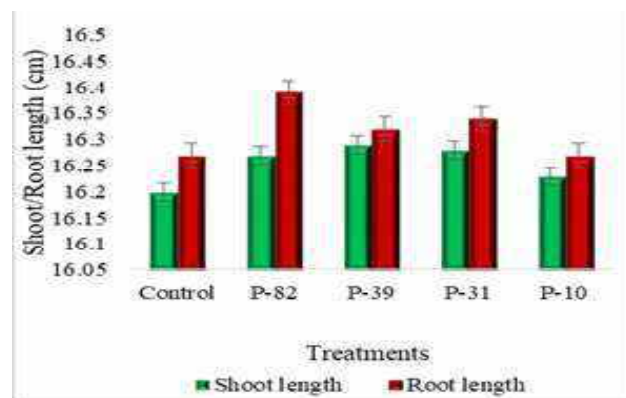


Fig. 3 : Effect of inoculation of endophytic fungal isolates on seedling growth of mung bean without salinity stress (a) and with salinity stress (b)

fungal endophyte (*Aspergillus awamori*) associated with Mung bean seedlings exhibited enhanced seedling growth due to ion homeostasis and antioxidant enzyme activity which resulted in low accumulation of stress markers. Higher concentration of Ca, Mg, K, N, and P also mitigate the salt stress.

### Confirmation of Fungal Endophytes in Inoculated Mung Bean Seedlings

The inoculated fungi were recovered from root, stem and leaf bits of mung bean seedlings and their morphology were compared with their mother culture. These fungi revealed morphologically similar characters of the mother culture (Fig. 4) and therefore they were confirmed as same isolates (Manasa *et al.*, 2020 and Sampangi-Ramaiah *et al.*, 2020).

### Effect of Inoculation of Fungal Endophytes on Growth of Mung Bean Under Greenhouse Conditions

Increased plant height was recorded in endophyte inoculated plants (Table 1). The enhanced plant height in inoculated plants under salinity stress may be due to regulation of ion homeostasis, accumulation of compatible solutes and phytohormone production by endophytes (Ali *et al.*, 2021) this upheld the plant

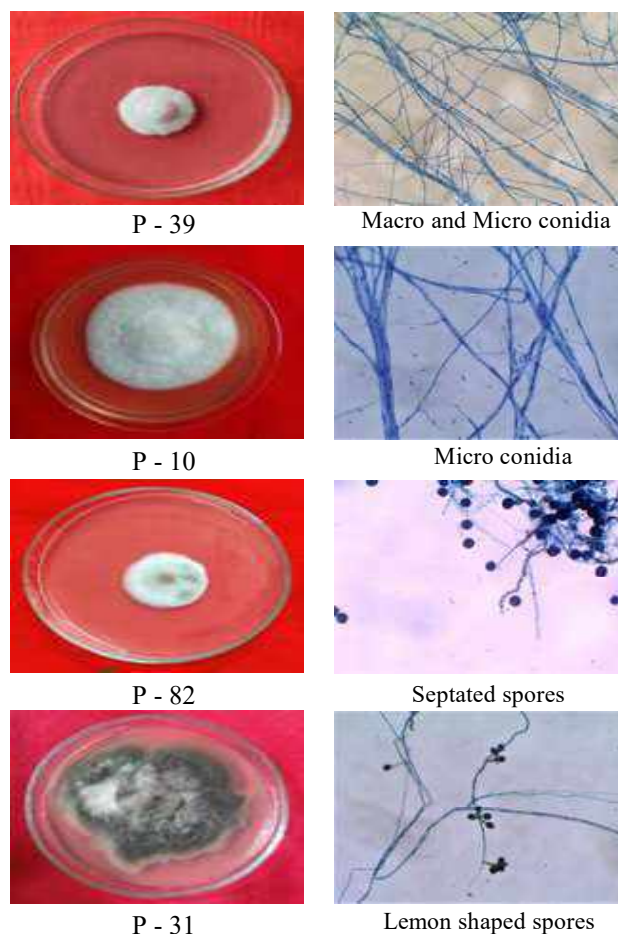


Fig. 4. Colony and microscopic view (40X) of spores of salt stress tolerant endophytic fungal isolates

TABLE 1  
Effect of fungal endophytes on growth parameters of mung bean under salinity stress

Treatments	30 DAS			60 DAS		
	Plant height (cm)	No. of branches	No. of leaves	Plant height (cm)	No. of branches	No. of leaves
Control	22.20 <sup>cd</sup>	2.00	6.00	28.20 <sup>cd</sup>	4.40 <sup>d</sup>	13.20 <sup>d</sup>
Salt stress (4 dS/m)	18.40 <sup>cf</sup>	2.00	6.00	23.20 <sup>g</sup>	3.58 <sup>g</sup>	10.74 <sup>g</sup>
<i>Ulocladium</i> sp.	23.00 <sup>bc</sup>	2.00	6.00	30.70 <sup>b</sup>	4.80 <sup>b</sup>	14.40 <sup>b</sup>
<i>F. avenaceum</i>	24.40 <sup>a</sup>	2.00	6.00	32.30 <sup>a</sup>	5.00 <sup>a</sup>	15.00 <sup>a</sup>
<i>Chaetomium</i> sp.	23.60 <sup>ab</sup>	2.00	6.00	30.40 <sup>b</sup>	5.00 <sup>a</sup>	15.00 <sup>a</sup>
<i>F. tricinctum</i>	23.20 <sup>abc</sup>	2.00	6.00	28.40 <sup>cd</sup>	4.60 <sup>c</sup>	13.80 <sup>c</sup>
Salt stress+ <i>Ulocladium</i> sp.	20.40 <sup>e</sup>	2.20	6.00	27.60 <sup>de</sup>	4.00 <sup>f</sup>	12.00 <sup>f</sup>
Salt stress + <i>F. avenaceum</i>	21.20 <sup>de</sup>	2.00	6.00	29.00 <sup>c</sup>	4.40 <sup>d</sup>	13.20 <sup>d</sup>
Salt stress + <i>Chaetomium</i> sp.	20.60 <sup>e</sup>	2.00	6.00	25.20 <sup>f</sup>	4.40 <sup>d</sup>	13.20 <sup>d</sup>
Salt stress+ <i>F. tricinctum</i>	20.40 <sup>e</sup>	2.00	6.00	26.70 <sup>e</sup>	4.20 <sup>e</sup>	12.60 <sup>e</sup>
CD@5%	1.30	NS	NS	0.91	0.17	0.52

Note : Mean values followed by same super script in each column do not differ significantly at  $p=0.05$  level by DMRT

fitness under salinity stress. Number of branches and leaves did not differ significantly at 30 days of growth but differences were observed in 60 days old plants indicating that the endophytes can influence the growth against salinity stress. However, the *Fusarium avenaceum* was found superior in enhancing the growth at salt stress compared to other three endophytes.

Further, the *Fusarium avenaceum* treated plants showed significant increase in yield parameters (Table 2). The enhanced yield parameter in endophyte inoculated plants may be attributed to sustained photosynthetic process. Inoculation of fungal endophytes reduces the uptake of Na<sup>+</sup> ions due to upregulation of plant response genes, increased phytohormone production, enhanced antioxidant enzymatic activity (Sampangi-Ramaiah *et al.*, 2020). This indicated that the endophyte inoculation resulted in higher photosynthetic efficiency leading to higher yield parameters than uninoculated plants under salinity stress (Munns and Tester, 2008). Ikram *et al.* (2019) reported increased biomass and chlorophyll content in endophyte treated plants.

TABLE 2  
Effect of fungal endophytes on yield parameters of mung bean under salinity stress

Treatments	No. of pods /plant	Weight of pods (g)/ plant	Seed yield (g) /plant
Control	21.60 <sup>b</sup>	15.94 <sup>c</sup>	12.79 <sup>c</sup>
Salt stress (4 dS/m)	20.20 <sup>c</sup>	6.86 <sup>h</sup>	5.14 <sup>g</sup>
<i>Ulocladium</i> sp.	22.40 <sup>a</sup>	16.39 <sup>ab</sup>	14.50 <sup>b</sup>
<i>F. avenaceum</i>	22.20 <sup>a</sup>	16.48 <sup>a</sup>	15.31 <sup>a</sup>
<i>Chaetomium</i> sp.	22.00 <sup>a</sup>	16.40 <sup>ab</sup>	14.07 <sup>b</sup>
<i>F. tricinctum</i>	22.00 <sup>a</sup>	16.27 <sup>b</sup>	13.91 <sup>b</sup>
Salt stress + <i>Ulocladium</i> sp.	22.60 <sup>a</sup>	9.44 <sup>c</sup>	8.90 <sup>dc</sup>
Salt stress + <i>F. avenaceum</i>	22.80 <sup>a</sup>	9.73 <sup>d</sup>	9.55 <sup>d</sup>
Salt stress + <i>Chaetomium</i> sp.	22.20 <sup>a</sup>	9.07 <sup>f</sup>	8.65 <sup>e</sup>
Salt stress + <i>F. tricinctum</i>	22.20 <sup>a</sup>	8.88 <sup>g</sup>	7.89 <sup>f</sup>
CD @ 5 %	1.09	0.16	0.72

Note : Mean values followed by same super script in each column do not differ significantly at  $p=0.05$  level by Duncan Multiple Range Test (DMRT)

TABLE 3  
Effect of fungal endophytes on physiological parameters of Mung bean under salinity stress

Treatments	Chl a (mg/g FW)	Chl b (mg/g FW)	Total chl (mg/g FW)	Carotenoid content (mg/g FW)	RWC (%)	Proline content (μmol/g FW)
Control	0.92 <sup>b</sup>	0.26 <sup>d</sup>	2.11 <sup>d</sup>	0.29 <sup>b</sup>	89.47 <sup>c</sup>	7.03 <sup>g</sup>
Salt stress (4 dS/m)	0.81 <sup>f</sup>	0.21 <sup>f</sup>	1.83 <sup>i</sup>	0.24 <sup>d</sup>	85.90 <sup>f</sup>	12.35 <sup>e</sup>
<i>Ulocladium</i> sp.	0.92 <sup>b</sup>	0.29 <sup>b</sup>	2.14 <sup>b</sup>	0.35 <sup>a</sup>	91.16 <sup>b</sup>	7.05 <sup>f</sup>
<i>F. avenaceum</i>	0.93 <sup>a</sup>	0.30 <sup>a</sup>	2.16 <sup>a</sup>	0.37 <sup>a</sup>	91.84 <sup>a</sup>	7.05 <sup>f</sup>
<i>Chaetomium</i> sp.	0.92 <sup>b</sup>	0.28 <sup>c</sup>	2.13 <sup>c</sup>	0.30 <sup>b</sup>	89.82 <sup>c</sup>	7.04 <sup>fg</sup>
<i>F. tricinctum</i>	0.92 <sup>b</sup>	0.29 <sup>b</sup>	2.14 <sup>b</sup>	0.33 <sup>b</sup>	91.10 <sup>b</sup>	7.04 <sup>fg</sup>
Salt stress+ <i>Ulocladium</i> sp.	0.87 <sup>c</sup>	0.26 <sup>d</sup>	2.00 <sup>f</sup>	0.28 <sup>c</sup>	87.27 <sup>d</sup>	13.35 <sup>b</sup>
Salt stress + <i>F. avenaceum</i>	0.87 <sup>c</sup>	0.28 <sup>c</sup>	2.02 <sup>e</sup>	0.30 <sup>b</sup>	87.62 <sup>d</sup>	13.83 <sup>a</sup>
Salt stress + <i>Chaetomium</i> sp.	0.83 <sup>c</sup>	0.22 <sup>e</sup>	1.89 <sup>h</sup>	0.25 <sup>c</sup>	86.14 <sup>e</sup>	12.75 <sup>d</sup>
Salt stress+ <i>F. tricinctum</i>	0.85 <sup>d</sup>	0.26 <sup>d</sup>	1.96 <sup>g</sup>	0.26 <sup>c</sup>	86.45 <sup>e</sup>	13.14 <sup>c</sup>
CD@1%	0.007	0.008	0.009	0.034	0.60	0.027

Note : Mean values followed by same super script in each column do not differ significantly at  $p = 0.01$  level by DMRT  
FW-Fresh weight, RWC- Relative water content

The physiological parameters such as chl a, chl b, total chl and carotenoid content was significantly higher in *F. avenaceum* inoculated plants (Table 3). This further indicated the superiority of the fungus. Enhanced photosynthetic pigments in endophyte inoculated plants may be due to production of antioxidants by endophytes which could neutralize the ROS and thus protecting photosynthetic pigments. The chl a, chl b, total chl and carotenoid content was reduced in uninoculated plants under salinity stress. This is due to production of reactive oxygen species which oxidizes the photosynthetic pigments (Munns and Tester 2008). Ali *et al.*, (2021) reported the protection of photosynthetic pigments by antioxidants and phytohormones produced by fungal endophytes.

Similarly, the plants inoculated with *Fusarium avenaceum* significantly increased the relative water content (RWC) compared to un-inoculated plants under salinity stress (Table 3). This may be due to ion homeostasis and accumulation of organic solutes, soluble sugars, proteins by fungal endophytes (Bagheri *et al.*, 2013). While, the reduced RWC in uninoculated plants may be due to reduced water uptake by the roots attributed to higher salinity at root surface resulting in higher osmotic potential which leads to the reduced ability of plant to absorb water (Munns and Tester, 2008). Ali *et al.* (2021) reported that the plants under salinity stress without endophytes inoculation lowered RWC compared to inoculated plants. The *Fusarium avenaceum* inoculated plants recorded significantly higher proline content (Table 3) under salt stress. The proline provide protection against ROS produced under salinity stress. Bagheri *et al.* (2013) reported higher proline, proteins and sugars in *P. indica* colonized rice plants under salt stress.

The fungal endophytes isolated from plants of North Western Himalayan region showed salinity stress tolerance. Among them *Fusarium avenaceum* found superior in inducing salinity stress tolerance in mung bean under *in-vitro* as well as in pot experiment. The study revealed that the endophytes can impart salinity stress tolerance in Mung bean.

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## Efficiency of Rice Farms under Different Cultivation Systems : A Stochastic Frontier Cost Approach

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### ABSTRACT

The paper has examined the efficiency of growing rice under different cultivation systems in Mandya district of Karnataka, using the data collected from 30 farmers under each of the cultivation systems namely, conventional, SRI (System of Rice Intensification), aerobic and DSR (Drum Seeded Rice) method. In total, the sample size was 120 rice growing farmers. The data was analysed using stochastic frontier cost function to estimate the cost efficiency of the farmers and to examine the factors influencing cost inefficiency. The results revealed that cost of seeds, fertilizers, human labour and machine labour had significant influence on cost of production. The variables such as age (0.239), landholding (0.022) and experience (0.215) in farming influenced cost inefficiency in conventional rice cultivation. Membership in organization (-0.075) and access to extension services (-0.289) were significant in explaining cost efficiency in SRI method. Education (-0.310) was the major factor which significantly contributed to the cost efficiency of aerobic farmers. Whereas, in DSR method age (-0.200) and access to extension services (0.083) were the significant factors. The mean cost or allocative efficiency score in aerobic was 0.94, which was highest among all the systems followed by SRI, DSR and conventional method with scores of 0.92, 0.88 and 0.55, respectively.

*Keywords* : Rice farms, Cultivation system, Cost efficiency, Stochastic frontier

RICE is a staple food of people in most of the countries of the world and is a very important and essential part of the food system in many countries. India is the second largest producer of rice in the world after China. In India, rice is grown on an area of 45,769 thousand hectares with production of about 124 million tonnes (www.indiastat.com). It is the food cereal which made countries to overcome the problems of hunger and starvation and has certainly played a major role in taking out the people out of food insecurity. Paddy crop holds the key for food security of the country (Bora *et al.*, 2021). Despite its vital role, the rice cultivation system has become one of the major sources for greenhouse gas emission from agriculture and also a cause for higher water consumption. The different rice cultivation

systems like aerobic, SRI (System of Rice Intensification), dry seeded rice cultivation, alternate wetting and drying etc. help in yielding higher returns and reducing costs. Adopting a system which increases yield, reduces water consumption, reduces costs *etc.* will aid in sustainable rice production with increased efficiency.

Efficiency means producing maximum output from given level of inputs with respect to production is concerned. Cost or allocative efficiency is producing output at minimum possible costs and with given input prices. Measuring efficiency will help in knowing what amount of resources can be saved by following a particular system of cultivation, which is reflected in the reduced costs. Efficiency can be measured for



an individual farm or a group of farms or farms practicing different methods of cultivation. A better understanding and measurement of efficiency in agriculture is required in the context of lower availability of key resources and production factors, such as land or water in adequate quantity and quality (Singh *et al.*, 2020). Factors that influence the farming system efficiency can be distinguished as controlled (farmer's managerial skill) and uncontrolled factors (natural factors, price and agriculture institution). The integration of all of the variables together, will create the level of efficiency that can be achieved (Hidayah *et al.*, 2013). Farm level inefficiency is likely to be affected by exogenous factors, *i.e.* factors that are neither inputs nor outputs of the production process, but nonetheless affect the farm performance (Bhattacharyya, 2016). So, it is important to study the variables which influence inefficiency. It is also necessary to know the outcome of different cultivation systems to suggest adaptation strategies. In this climate change scenario, farmers must be able to adapt coping strategies to ensure long-term output and these adaptation measures can help people minimize their susceptibility and improve their 'socio-economic status' and 'quality of life' (Pooja *et al.*, 2022).

Cultivation system like SRI method is considerably more profitable than traditional method due to low input expenditure. The total cost of cultivation was higher in traditional method (Rs.14014.54/ac) than SRI method *i.e.* Rs.12154.63/ac (Agarwal *et al.*, 2018). Thus, cost efficiency analysis will reveal the method of cultivation to be practised. Vinay *et al.* (2016) analysed the impact of direct seeded rice (DSR) on economics of paddy crop in Haryana. The net return was higher in DSR (Rs.60105/ha) as compared to transplanted rice (Rs.57532.5/ha) and BC ratio was 2.13 in DSR while it was only 1.94 in transplanted rice. This indicates the decreased costs due to increase in efficiency. Moreover, efficiency analysis is also an important input to the policy makers. The aerobic rice cultivating farms were more technically and economically efficient compared to conventional rice cultivating farms. Effective policies to promote and create awareness about aerobic rice can boost the rice production and productivity sustainably. The focus

should be given to optimal allocation of resources which enhances the farm productivity and returns (Kumar *et al.*, 2021). This highlights the requisite of cost efficiency analysis.

In this regard, the study made an attempt to estimate the cost efficiency of different rice cultivation systems and identify the system which is cost or allocatively most efficient. It also examined the influence of socio-economic characteristics on cost inefficiency under different rice cultivation system.

### Study Area and Selection of Farmers

The study was carried out in Mandya district of Karnataka, which is one of the major producers of rice in Southern Karnataka. Purposive sampling was used to sample the farmers for the study. The primary data was collected from 120 farmers consisting of 30 farmers from each cultivation system namely, conventional rice cultivation, SRI (System of Rice Intensification), aerobic and DSR (Drum Seeded Rice) method. At first the villages practicing these cultivation systems were selected and then the farmers were selected randomly. The data was collected from the respondents through personal interview method using pre-tested, well-structured schedule to achieve the objectives of the study. The required information regarding age, education, land holdings, costs incurred, input usage *etc.* in rice cultivation was collected for the agricultural year 2021-22.

### Analytical Tools Used

#### Efficiency Analysis

Efficiency analysis orders decision-making units such as firm or a farm, by comparing all resources engaged in production and the costs incurred to produce a given set of outputs and building a frontier based on the input costs. The Cobb-Douglas Stochastic Frontier Cost (SFC) approach was used for assessing the cost efficiency of rice farmers under different rice cultivation system, following the Coelli (1996) model as follows:

$$\ln C_i = \alpha_0 + \sum_{j=1}^4 \alpha_j \ln X_{ji} + (v_i + u_i)$$

where,  $\ln$  denotes natural logarithm.  $C_i$  is the total production cost of the farm  $i$  measured in rupees per acre,  $X_{1i}$  is the cost of seeds (Rs./acre),  $X_{2i}$  is the cost of fertilizers used (Rs./acre),  $X_{3i}$  is human labour cost and  $X_{4i}$  is the machine labour cost.  $v_i$  is a symmetric, identically and independently distributed  $N(0, \sigma_v^2)$  error term. It represents random variation in production due to random exogenous factors, such as measurement errors and statistical noise.  $u_i$  is a non-negative error term. It reflects cost inefficiency relative to the stochastic frontier.

The computer programme FRONTIER Version 4.1 was used to estimate the model and to obtain the maximum likelihood estimates of the SFP function. The calculation of MLE requires (Coelli, 1996).  $\sigma^2 = \sigma_v^2 + \sigma_u^2$ . This indicates total variance is due to variance in error term ( $v$ ) and non-negative random variable ( $u$ ), where in  $v$  and  $u$  assumed to be independent of each other. The error term  $v_i$  represents the influence of factors outside the control of the farmer, while  $u_i$  represents the cost inefficiency factors because of poor management practices which are under control of the farmer. This variance parameter in model is represented by Gamma value, calculated using the following equation:

$$\gamma = \frac{\sigma_u^2}{\sigma_v^2 + \sigma_u^2}$$

### Factors Affecting the Cost inEfficiency

In order to assess the factors associated with cost inefficiency, the cost efficiency scores were used. It was analysed taking the degree of cost efficiency scores as dependent variable. The empirical specification of the cost inefficiency model is given by (Bettese and Coelli, 1995).

$$u_i = \delta_0 + \sum_{m=1}^7 (\delta_m Z_m)$$

Where  $Z_{mi}$  are socio-economic characteristics,  $Z_{1i}$  is age of the farmer.  $Z_{2i}$  is education (0 = Illiterate, 1 = primary, 2 = secondary, 3 = college, 4 = graduation),  $Z_{3i}$  is landholding of the farmer,  $Z_{4i}$  is the size of the family,  $Z_{5i}$  is the experience in farming,  $Z_{6i}$  is a binary variable equal to one if the farmer has membership in any organization and zero otherwise and  $Z_{7i}$  is binary variable equal to one if the farmer has access to extension services and to zero otherwise.

## RESULTS AND DISCUSSION

### Socio-Economic Characteristics of Rice Farmers

The socio-economic characteristics of the farmers is given in Table 1. The results indicated that average age of the farmers under conventional rice cultivation was 53 years and with respect to SRI, aerobic and DSR farmers, the average age was 48, 47 and 43 years, respectively. The average landholding was more than

TABLE 1

Socio-economic characteristics of rice farmers under different rice cultivation system

Variables	Conventional (n=30)	SRI (n=30)	Aerobic (n=30)	DSR (n=30)
Age (years)	53.00	48.00	47.00	43.00
Education level (years of formal education)	8.00	8.00	10.00	10.00
Landholding (acres)	4.83	3.08	3.33	3.40
Experience (years)	25.00	22.00	20.00	20.00
Family size (No.)	5.00	5.00	5.00	5.00
Membership (No.)	13.00 (43)	21.00 (70)	19.00 (63)	20.00 (67)
Access to extension services (No.)	6.00 (20)	25.00 (83)	23.00 (77)	27.00 (93)

Note: Figures in parentheses indicate percentage to total

3 acres for SRI, aerobic and DSR farmers but was more than 4 acres for farmers under conventional rice cultivation. The conventional farmers had around 8 years of formal education and SRI, aerobic and DSR farmers had 8, 10 and 10 years of formal education, respectively. The farmers under all the cultivation system had more than 20 years of experience in farming. It was also found that majority of the farmers belonged to the family size of five across all the systems.

It was noticed that around 43 per cent of the farmers under conventional rice cultivation had membership in organizations and 70, 63 and 67 per cent of the farmers under SRI, aerobic and DSR cultivation

had membership in organizations, respectively. It was also noted that more than 70 per cent of the farmers under SRI, aerobic and DSR had access to extension services but it was only 20 per cent for farmers under conventional method.

### Analysis of Cost Efficiency

The Cobb-Douglas cost function was estimated using the computer version FRONTIER 4.1 and the results of the maximum likelihood estimates of the stochastic cost frontier of rice farmers under conventional rice cultivation is given in Table 2. The results revealed that one per cent increase in the seed cost, human labour and machine labour cost will increase the total cost by 1.05, 1.96 and 0.9 per cent, respectively and was found significant. The estimated coefficient of the explanatory variables in the cost inefficiency model shows that all the coefficients have the expected signs except age and experience. With increase in age and experience by one per cent the cost inefficiency increased by 0.239 and 0.215 per cent indicating that the farmers who are old are reluctant to adopt the cost efficient technologies. This is in line with the findings of Singh *et al.* (2020) who reported that age is positively related to cost inefficiency. Similarly with respect to increase in experience, following conventional practices leads to increased cost inefficiency.

Sigma squared ( $\sigma^2$ ) on the other hand is 0.221 and statistically significant at one per cent indicating correctness of fit of the model as assumed for the composite error term. The estimated gamma parameter of 0.437 is highly significant at five per cent, indicating that around 44 per cent of the variation in the total cost of production among the sampled farmers is due to differences in their cost efficiency. Moreover, the presence of cost inefficiency was tested by LR (Likelihood Ratio) statistic, it was 14.054 which is lesser than the critical chi square value of 24.049, which implies the assumption of no cost inefficiency was rejected.

TABLE 2

Maximum likelihood estimates of the stochastic cost frontier of rice farmers (Conventional method)

Variables	Coefficients	t-ratio
Constant	- 9.309	-1.024
Seed cost	1.048 *	2.576
Fertilizer cost	-0.082	-0.335
Human labour cost	1.960 **	2.127
Machine labour cost	0.914 *	2.830
<i>Inefficiency model</i>		
Constant	1.597	0.624
Age	0.239 *	2.902
Education	0.144	0.878
Land holding	-0.022 **	- 2.074
Family size	-0.280	-0.734
Experience in farming	0.215 *	3.741
Membership in organization	-1.006	-0.270
Access to extension services	-0.359	-0.860
<i>Variance parameters</i>		
Sigma squared	0.221 *	3.867
Gamma	0.437 **	2.292
Log likelihood	19.989	
LR statistic	14.054	

Note: \*, \*\* indicates significance at one and five per cent probability level, respectively

TABLE 3

Maximum likelihood estimates of the stochastic cost frontier of rice farmers (SRI method)

Variables	Coefficients	t-ratio
Constant	2.452 *	3.501
Seed cost	0.054 *	5.977
Fertilizer cost	0.130 *	13.877
Human labour cost	0.340 *	12.923
Machine labour cost	0.373 *	9.033
<i>Inefficiency model</i>		
Constant	-0.106	-0.199
Age	0.009	0.158
Education	-0.078	-1.429
Land holding	-0.039	-0.279
Family size	-0.023	-0.828
Experience in farming	-0.011	-0.427
Membership in organization	-0.075 **	-2.389
Access to extension services	-0.289 *	-2.595
<i>Variance parameters</i>		
Sigma squared	0.636 *	3.733
Gamma	0.719 *	3.433
Log likelihood	70.878	
LR statistic	20.539	

Note : \*, \*\* indicates significance at one and five per cent probability level, respectively

The maximum likelihood estimates of the stochastic cost frontier of rice farmers under SRI method of rice cultivation is given in Table 3. The cost elasticities of all the input variables used in the cost analysis were positive which implies that an increase in the cost of seed, fertilizer, human labour and machine labour increases total production costs. The coefficients were positive and significant at one per cent. One per cent increase in costs of seed, fertilizer, human labour and machine labour increases the cost by 0.054, 0.130, 0.340 and 0.373 per cent, respectively.

The inefficiency effects of membership in organization and access to extension services was negative and

TABLE 4

Maximum likelihood estimates of the stochastic cost frontier of rice farmers (Aerobic method)

Variables	Coefficients	t-ratio
Constant	12.955 *	11.533
Seed cost	0.056 *	4.510
Fertilizer cost	0.140 *	5.540
Human labour cost	0.426 *	9.329
Machine labour cost	0.598 *	5.213
<i>Inefficiency model</i>		
Constant	-0.115	-0.167
Age	0.074	0.339
Education	-0.310 *	-2.77
Land holding	-0.002	-0.078
Family size	-0.048	-0.689
Experience in farming	0.060	1.940
Membership in organization	-0.039	-1.120
Access to extension services	-0.017	-1.130
<i>Variance parameters</i>		
Sigma squared	0.031 *	3.763
Gamma	0.89 *	190.080
Log likelihood	68.58	
LR statistic	23.346	

Note : \*, \*\* indicates significance at one and five per cent probability level, respectively

significant. This means that both the factors are contributing positively to cost efficiency. The farmers obtain required and necessary technical advice and knowledge, thereby produce at efficient costs. Increase in membership in organization and access to extension services by one per cent would lead to increase in cost efficiency by 0.075 and 0.289 per cent, respectively.

The sigma squared value was 0.636 and significant at one per cent level indicating the goodness of fit. The gamma parameter was estimated to be 0.719 and was significant at one per cent level. This reveals that approximately 72 per cent of the variation in the total

cost of production among the sampled farmers is due to differences in their cost efficiency. LR statistic was 20.539 and lesser than the critical chi square value of 24.049, depicting the presence of cost inefficiency.

The result of stochastic cost frontier for aerobic rice cultivation is depicted in Table 4. It was observed that coefficients of all the input variables *i.e.* cost of seeds, fertilizers, human labour and machine labour were highly significant at one per cent level. With respect to inefficiency effects, education was found negative and significant at one per cent level. Increase in education by one per cent decreases cost inefficiency by 0.37 per cent. This reveals that higher

the level of education, higher the cost efficiency of the farms. Aboaba (2020) reported that higher the level of education, the higher the allocative efficiency, which implies that educated farmers are allocatively efficient compared to their counterparts. The Sigma squared estimate (0.031) was also significant at one per cent level indicating the good fit of the model. The gamma value was 0.89 which revealed that 89 per cent of the variation in cost of production is attributed to the variation in costs among the rice farmers and is due to differences in cost efficiency.

It was observed from the stochastic cost frontier analysis under DSR rice cultivation that the cost of human labour was significant at one per cent whereas seeds and machine labour was significant at five per cent level. The coefficients obtained for the maximum likelihood estimates are given in Table 5. Even in the case of DSR method, increase in all the input variables increases the total cost. In the case of cost inefficiency effects, access to extension services and education were significant and one per cent increase in education and extension services decreases cost inefficiency by 0.20 and 0.08 per cent, respectively. Sigma squared and gamma was observed to be 0.092 and 0.69 and was significant at five per cent level.

TABLE 5

Maximum likelihood estimates of the stochastic cost frontier of rice farmers (DSR method)

Variables	Coefficients	t-ratio
Constant	7.634 *	7.931
Seed cost	-0.005 *	-3.005
Fertilizer cost	0.109	0.103
Human labour cost	0.169 *	3.085
Machine labour cost	0.022 **	2.045
<i>Inefficiency model</i>		
Constant	0.006	0.007
Age	0.003	0.004
Education	-0.200 *	-3.019
Land holding	-0.024	-0.092
Family size	-0.002	-0.012
Experience in farming	-0.022	-1.302
Membership in organization	-0.036	-0.113
Access to extension services	-0.083 *	55.770
Variance parameters		
Sigma squared	0.092 **	1.908
Gamma	0.69 **	2.502
Log likelihood	29.104	
LR statistic	7.528	

Note: \*, \*\* indicates significance at one and five per cent probability level, respectively

### Efficiency Distribution of Rice Farmers Under Different rice Cultivation Systems

The distribution of farmers according to the cost efficiency scores is presented in Table 6. It was observed that mean efficiency score was 0.55, 0.92, 0.94 and 0.88 for conventional, SRI, aerobic and DSR cultivation, respectively. This indicates that the cost efficiency was highest in aerobic rice cultivation. The cost efficiency obtained is supported by Aboaba (2020) which reported that mean allocative efficiency implies that rice farmers were 94 per cent cost-efficient, that is they were able to maximize their total output by minimizing 94 per cent of their total production cost, which shows that there is room for six per cent improvement.

TABLE 6  
Distribution and summary statistics for cost efficiency scores of farmers under different rice cultivation systems

Cost efficiency scores	Conventional	SRI	Aerobic	DSR
≤0.5	9.00	-	-	-
>0.50 ≤0.70	20.00	4.00	-	7.00
>0.70 ≤0.90	1.00	4.00	2.00	3.00
>0.90	-	22.00	28.00	20.00
Mean	0.55	0.92	0.94	0.88
Minimum	0.12	0.60	0.86	0.66
Maximum	0.60	0.97	0.98	0.95

The lowest cost efficiency was obtained under conventional rice cultivation indicating that around 55 per cent of the farmers were allocatively efficient. The mean efficiency scores obtained for SRI method of cultivation was higher than 83 per cent as reported by Mwatete *et al.* (2015). With respect to DSR rice cultivation, the efficiency scores obtained are in line with results of Maurice *et al.* (2015) which reported allocative efficiency of 0.84 for food crop production among small scale farmers. Thus, there was 45 per cent room for increasing efficiency for conventional farmers and only 2 per cent for aerobic farmers.

The adoption of a rice cultivation system which decreases cost inefficiency is the important aspect in the present context because with rice being a major staple food it has to be seen that there is sufficient and sustainable rice production in the country which can be produced at minimum costs. The results from the maximum likelihood estimates revealed that cost of seeds, fertilizers, human labour and machine labour were the various input variables influencing cost of production. The variables such as age, landholding and experience in farming significantly influenced cost inefficiency in conventional rice cultivation. Membership in organization and access to extension services were significant in explaining cost inefficiency of SRI method of cultivation. The major factor for aerobic farmers was education which

contributed significantly to cost efficiency. With respect to DSR method, age and access to extension services were the significant factors. Moreover, it was observed that the most cost efficient system was aerobic rice cultivation followed by SRI method and the system which had more scope for allocative efficiency was conventional method. Therefore, adopting aerobic rice cultivation or SRI method on a larger scale in the study area would help in increasing efficiency and sustainable management of resources.

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## Technology Driven Enhancement of Farmers Income in Karnataka : Lessons Learnt from Successful Farmers

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### ABSTRACT

Addressing the national agenda of doubling farmer's income, the Indian Council of Agricultural Research used its nation-wide network of Krishi Vigyan Kendras for enhancing farmer's income through technology centric approaches. Successful farmers who could enhance their income with the help and support of technological interventions have clearly demonstrated the possibility for enhancing farmers income. The average income of farm households increased through a multi-pronged approach. Horticulture crops provided the dominant source of total income as well as additional income. Percentage of increase was higher with farm and non-farm enterprises, followed by fisheries activities as the income from these sectors was relatively low during benchmark period. Change in income could be attributed to shift from field crops to horticulture crops and from crops-based farming to crops + livestock; crops + livestock + enterprises and crops + enterprises. Technical advisories based on diagnostic services and supply of critical inputs of new technologies were largely responsible for the shift towards high-income activities. Significant increase in income was evident among farmers from all land classes due to technological interventions. While the rate of increase in income was higher with smaller holdings, the additional income generated was greater with larger holdings.

*Keywords:* Farmer's income, Extension education, Landholding, Diversification, Technology uptake

**I**N India, early agricultural development strategies focused primarily on raising agricultural output for attaining food security (Chand, 2017). The announcement by the Prime Minister of India for doubling farmer's income propelled the momentum towards enhancing farmer's income. Since then, farmer's income has been at the centre stage in the debates on agriculture. In 2016, the Inter-Ministerial Committee set up by the Ministry of Agriculture and Farmers Welfare, Government of India examined issues related to farmers income and suggested a seven-point strategy: (i) Enhancing production of crops and livestock through intensification; (ii) Raising productivity through better management and irrigation; (iii) Reducing cost of production through adoption of technologies and conventional

practices; (iv) Higher realization of net income through modern/ electronic marketing; (v) Processing/ value addition to farm produce; (vi) Diversification into high-value crops; and (vii) Adoption of supplementary agricultural/ non-agricultural enterprises backed by skill development programs (Dalwai, 2018). The government initiated several steps in this direction with a focus on enhancing farmers income through intensification, diversification, shift from subsistence to commercialization and business orientation towards agro-based small-scale enterprises (ICAR, 2016).

The Indian Council of Agricultural Research (ICAR), being the premier organization in the field of agricultural research, education and extension,



initiated efforts to develop strategy documents for each state focusing on technologies, technology delivery mechanisms and market linkages. The ICAR utilized its nation-wide network of Krishi Vigyan Kendras (KVKs), which have a multidisciplinary team of specialists, to operationalize the strategies through scientific farming for enhancing farmers income in each district. Agriculture, horticulture, livestock production, sericulture, supplementary enterprises, processing and value addition and farm-based income generating activities comprised the focus of the KVKs to increase farmers income. Farmers are the ultimate deciders in the process of technology adoption as they have mastered the art of making the best use of technologies within the realm of their natural and socio-economic resources. Thus, an effort was made to analyze successful farmers across diverse agro-climatic and socio-economic situations in Karnataka. It was also felt necessary to assess the contribution of agriculture, horticulture, animal husbandry, fisheries and other enterprises to household income across different land holding categories so that the results

throw useful insights into opportunities embedded in increasing farmers income.

### METHODOLOGY

A simple format was designed by the ICAR and shared with all the KVKs for creating a database of farmers benefited with KVK interventions in each district. From among the list of contact farmers, about 110 farmers were randomly identified by each KVK for the present study. In all, a total of 3648 successful farmers of 33 KVKs functioning in all the rural districts of Karnataka constituted the sample. Annual net income during 2016-17 served as the benchmark data and was compared with the income levels during 2020-21, estimated at current prices. The income is assessed for the entire farm, not per unit area and by considering income from farm and non-farm enterprises managed by the farm family. Hence, income levels are reported as Rs/ household. Price effect due to higher minimum support price or enhancement in general prices is also included in the estimation. Technological interventions of KVKs that must have contributed to higher income of farm households under each sector are presented below:

Sector	Technological intervention
Field Crops	Introduction of improved varieties of paddy <i>viz.</i> , Gangavathi Sona, RNR-15048, MAS-26, KHP 13, KKP-5, direct seeding of rice (DSR), mechanical sowing and integrated crop management practices. Introduction of new varieties of finger-millet (ML-365, KMR-340, 630 and MR-6), sorghum (SPV-2217) and foxtail-millet (DHFT-109-3). Integrated pest management (IPM) of fall armyworm in maize. Introduction of new pigeonpea varieties <i>viz.</i> , BRG-3, 4, 5, GRG-811, TS3R, BSMR-736 and its intercropping in maize. Introduction of sugarcane variety VCF-517, adoption of nutrient management practices and biological control of root grub. Promotion of a new groundnut varieties GPBD-4, G2-52, DH-256, ICGV-03043, K-6, KDG-128 and cultivation of groundnut in paddy fallows. Introduction of new varieties of chickpea (JAKI-9218 and BGD 111-1), greengram (DGGV-2, BGS-9), blackgram (LBG791), horsegram (PHG-9, CRIDA-18), safflower (PBNS-12), sunflower (KBSH 53, RFSH 1887) and soybean (DSB-21). Promotion of micronutrient and pest management in cotton. Promotion of intercropping in sugarcane, cotton, maize, pigeonpea and groundnut. Adoption of dryland production technologies such as compartment bunding, seed hardening/treatment with CaCl <sub>2</sub> and farm pond supported protective irrigation, improved pulses production technologies such as pulse-magic and nipping.
Horticultural Crops	Introduction of new varieties/hybrids of chilli (Arka Kyathi and Arka Haritha), weed management and bio-intensive pest/disease management. Introduction of new hybrids of tomato (Arka Rakshak and Arka Abhed) and integrated pest/disease management practices. Promotion of new varieties of frenchbean (Arka Arjun, Sharat and Suvidha). Nutrient management in coconut to reduce nut dropping. Banana disease management and foliar nutrition. Nutrient and disease management in arecanut and arecanut husk decomposition. Mango pest disease management and mango-special

	as micronutrient supplement. Introduction of new onion varieties (Bhima Super and Bhima Shakti) and management of pests and diseases. Ginger rhizome rot management. Introduction of a new turmeric variety Pratibha and its processing at farm level for value addition. Promotion of new varieties of ridgegourd (Arka Prasan), okra (Arka Nikitha) and tuberose (Arka Prajwal)
Animal Husbandry	Promotion of balanced nutrition, area specific mineral mixture and clean milk production practices in dairy animals. Promotion of fodder varieties DHN-6, CoFS-29, 30, 31 Co-3, 4 and 5 and fodder seed production units. Introduction of breeds of backyard poultry (Swarnadhara), low-cost incubation, hatchery units and feed supplementation with azolla. Cost-efficient nutrition management with locally prepared feed formulations. Semi-intensive and intensive sheep and goat farming and micronutrient supplementation and deworming practices.
Farm and non-farm enterprises	Seed production of cereals, pulses, oilseeds and fodder crops. Horticultural nursery for seedlings of fruits, plantation, and vegetable crops. Mulberry cultivation and silkworm rearing for cocoon production. Bee keeping for honey production and its value addition. Millet processing and value addition. Value addition to Plate, direct and digital marketing strategies. Custom hiring of farm machinery and coconut climbing.

Percentage, frequency and weighted averages were used to decipher the data and present the results.

## RESULTS AND DISCUSSION

The impact in terms of household income under different components and the change in income for different land-class categories are presented and discussed in this section. The results in Table 1 compare the household income before and after the interventions by the KVKs. The average income of farm households was increased by 147 per cent between 2016-17 and 2020-21 in which horticulture crops (fruits, vegetables, flower crops, plantation crops, spices, medicinal and aromatic crops) provided the dominant source of household income (Rs.144549/ household during 2016-17

and Rs.364361/ household during 2020-21). Field crops comprising of food crops (cereals, pulses, oilseeds and millets) and cash crops (cotton, sugarcane, tobacco, jute and fodder crops) provided the next major amount of income (Rs.78925/ household during 2016-17 and Rs.153727/ household during 2020-21). The household income from livestock was moderate (Rs.71591/ household during 2020-21), but more than income from fisheries and other enterprises (bee keeping, mushroom production, seed / plant material production, food processing and value addition *etc.*).

In terms of percentage increase in income, the highest percentage increase was recorded among farm and non-farm enterprises (330.38%) during this period. The next highest increase in income

TABLE 1  
Level and change in household income

Crops and Enterprises	Net income (Rs/household at current prices)		% Increase in income	% Share in total income		Additional net income (Rs/ household)	% Share in additional income
	2016-17	2020-21		2016-17	2020-21		
Field crops	78925	153727	94.78	30.65	24.17	74802	19.76
Horticulture	144549	364361	152.07	56.13	57.28	219812	58.06
Livestock	23104	71591	209.75	8.98	11.26	48487	12.82
Fisheries	1921	7687	300.16	0.75	1.21	5766	1.52
Other enterprises	8985	38670	330.38	3.49	6.08	29685	7.84
Overall	257512	636099	147.02	100.00	100.00	378587	100.00

was observed in fisheries (300.16%). This is basically due to lower income levels during benchmark year (Rs.8233/- and Rs.1921/- per household respectively). Livestock income also increased substantially (209.75%) from Rs.23104/- household to Rs.71591/- household.

In terms of share in total income, horticulture was the major sector with 56.13 per cent of the total income during 2016-17, which further increased to 57.28 per cent during 2020-21. The share of field crops in total income declined from 30.65 per cent to 24.17 per cent, possibly due to crop diversification (from field crops to horticulture crops) and additional investment on livestock-based activities and other enterprises. The livestock sector consolidated its share in the household income to 11.26 per cent in 2020-21 from 8.98 per cent in 2016-17.

Share in additional income over the benchmark year was also highest from horticulture sector (Rs.219812 per household equivalent to 58.06 per cent), followed by field crops (19.76%) and livestock sector (12.82%). These statistics provide valuable insights to the approach of successful farmers for achieving higher income. These are in line with the statistics related to contribution of horticulture and livestock sector in national and state

GDP. As per the secondary sources (DES, 2022), horticulture component constituted 29.51 per cent of the total income from entire agriculture sector and contributed 3.24 per cent to Karnataka GSDP with Rs.52718 crore during 2019-20. Livestock sector employs 8.8 per cent of India’s population and contributed 16 per cent of the total income of small farm households (DAHD & F, 2019). Enhancing farmer’s income is possible (Chand, 2016) and the study results confirmed that the technology adoption is an important driver of enhancing farmer’s income.

Change in income could be attributed to shift from crops-based farming to (i) Crops + livestock which increased from 39.4 per cent to 46.7 per cent (ii) Crops + livestock + enterprises increased from 2.7 per cent to 8.2 per cent and (iii) Crops + enterprises increased from 2.3 per cent to 5.8 per cent as compared to benchmark year. Technical advisories and support in the form of diagnostic services, critical inputs and timely visits provided by KVKs were largely responsible for the shift towards high-income activities (Fig.1).

Practicing innovative cropping systems and activities are essential for increasing farmers income (Bankey *et al.*, 2019). Chand (2016) suggested that farmers income could be increased through diversification of production activities. Increase in

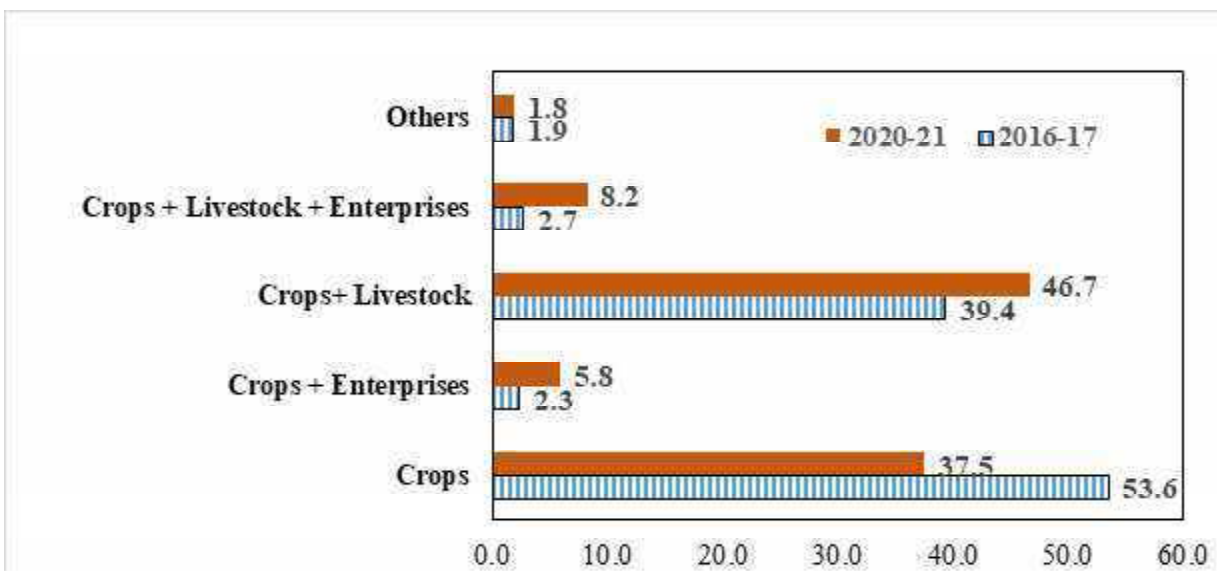


Fig.1 : Change in choice of crops and enterprises by farmers (%) due to KVK interventions

TABLE 2  
Income level and change in household income by land class

Land class	No. of households	% Share in total household	Income level (Rs/household at current prices)		Additional income (Rs/ousehold)	% Change in household income
			2016-17	2020-21		
Landless	20	0.55	78702	276401	197699	251.20
Marginal (<1.0ha)	790	21.65	109268	286108	176840	161.84
Small (1-2ha)	1479	40.54	175155	441184	266029	151.88
Medium (2-4ha)	865	23.72	296934	721890	424956	143.11
Large (>4ha)	494	13.54	679367	1643701	964334	141.95
Total	3648	100.00	257512	636099	378587	147.02

total factor productivity is essential for growth in output (Saxena *et al.*, 2017) and profitability in farming. Farmer's income was substantially enhanced when farm income was supplemented with other farm and non-farm activities (Sendhil *et al.*, 2017a).

The results in Table 2 depicted that farmers from all land classes were benefitted from the technological interventions. The income of the landless families increased by 251.20 per cent over benchmark year, which was highest among all categories. This was due to very low-income levels during the benchmark period (Rs.78702/ household per year) and higher additional income generated during the period (Rs.197699/household). Landless families were encouraged and supported to take up livestock and other enterprises with technological backing by the KVKs. Marginal landholder's income was increased by 161.84 per cent, but the additional income generated to each house hold was less (Rs.176840) compared to landless. This is a paradoxical situation where even the successful farmers cultivating marginal landholdings earned less than landless category farmers. Small holders were better than these two categories in terms of additional income generated (Rs.266029/household) although the percentage increase (151.88%) was comparatively less. Collective farming by smallholders with focus on integrated farming and mixed farming (Sendhil *et al.*, 2017b) could have resulted in higher income among small

farmers. The medium landholders earned almost double that of the above three categories (Rs.424956/ household) and large farm households could realize highest additional income (Rs.964334/household). Larger landholdings provided every possible opportunity for diversification, mechanization, economy of scale and risk taking for new activities, which are evident through higher income levels in the present analysis.

Technological breakthroughs that facilitate farmers to engage in the production of crops and commodities as per the demands of local markets is important for driving farmers income (Shivakumar and Chahal, 2018). Since there is huge gap between potential and actual yield being obtained (Swaminathan, 2016), increasing the income levels of farmers across the landholding categories is possible.

The results justify that scientific knowledge, when integrated with farmers experience, can contribute significantly to the income enhancement process in farming. The resilience of the farming sector during COVID proved its relevance to Indian economy beyond any doubt, as it was the only ray of hope for the livelihood amidst the pandemic that hit the economy hard.

The analysis of the success stories offers very important lessons. Diversification into high-value crops and commodities with location-specific technologies emerged as important paths for sustainable and climate-resilient agriculture. In such

cases, farmers information needs became complex and hence, there is a need to design a dynamic and single-window extension delivery system for the timely provision of appropriate advisory services. New technologies and enterprises require skills and therefore continuous upgradation of skills of both extension personnel and farmers needed. Hand holding of farmers in technology application and initial adoption also needs availability of quality inputs within the reach of farming community. This is particularly important during climatic aberrations that demand quick adaptation to contingency strategies. To supplement income from farm, there is a need to strengthen locally preferred agro-enterprises such as bee-keeping, seed production, nursery raising, custom-hiring services, mushroom production and food processing and value addition. Landless households could realize higher income by involving in animal husbandry and related enterprises. Beyond production, innovative marketing attempts played catalytic role in supplementing farmer's income. Successful direct marketing approaches through farmer's organizations and cooperatives need to be scaled up. Digital and online marketing strategies were useful even in remote areas but require further strengthening of internet connectivity and infrastructure.

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## Utilization of Psychomedicinal Plants in Udhampur District of Jammu and Kashmir

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### ABSTRACT

Psychomedicinal plants have been enormously used for traditional and commercial necessities in the Udhampur District of Jammu and Kashmir. The distinct household remedies were made and used for the treatment of neurological disorders and mental ailments such as anxiety, dementia, depression, epilepsy, headache, insomnia, improving cognition power, intelligence, migraine and snakebite. Since ancient times, these plants are being used in worshipping supernatural powers, marriages, prayers, exorcism, rituals and funerals. Sorcery practices like necromancy and voodoo, use some selected psychomedicinal plants for expelling the ghost influence out of an affected person and getting rid of negative energies by chanting mantras. In this communication, the authors collected information on different psychomedicinal plants used traditionally by the local people in Udhampur district, Jammu and Kashmir. An intensive and extensive field survey was conducted in 2021-2022, exploring such psychomedicinal plants. The tribal people, shepherds, medicinal practitioners, vaidyas, old folks and also other local residents provided useful information. This information was verified with the help of relevant literature. Collected plant species were correctly recognized and authenticated by regional herbaria. A total of 43 plant species from 40 distinct genera and 33 different families were identified. For each plant species, its scientific name, family, vernacular name, habit, part used and medicinal uses were described. The percentage of herbs were 60 per cent, shrubs were 14 per cent and trees were 26 per cent. Poaceae was the dominant family with five plant species. Entire natural resources and relevant indigenous knowledge are valuable and should be secured for the native people of this region where they don't have modern facilities for healthcare. Community knowledge should be raised through various awareness programmes about mental health issues. Further, phytochemical and pharmacological analysis needs to be done to examine the bioactive compounds responsible for treating neuro-disorders.

**Keywords:** Ethnobotany, Neuro-disorders, Mental health, Medicinal plants

THE word 'medicine' in Greek means 'cure or heal'. The medicinal plants used to treat the ailments of the psyche of living beings are known as psychomedicinal plants. In India, since the Vedic age period, psychomedicinal plants are used to cure the ailments of the mind through traditional medicinal practitioners, such as Hakims and Vaidyas, who used medicinal herbs for the treatment of mental illnesses. These herbs are also used for performing traditional

ceremonies, exorcisms, sorcery practices, hymns, making amulets, rituals, religious rites *etc.* (Badoni, 1987; Tiwari *et al.*, 2010; Anthwal *et al.*, 2010; Negi, 2010; Sardiana and Dinata, 2010; Mohanty *et al.*, 2011; Mathur and Joshi, 2013; Kandari *et al.*, 2014; Sharma *et al.*, 2014 and Bamin and Gajurel, 2015).

Jammu and Kashmir has peculiar topography and is wealthy in vegetation, peculiar traditions and customs.

It is famous for its aesthetic beauty and natural charms throughout the globe. Its high snow-covered mountains and dense forests attract tourists. A lot of work has been done on ethnobotany and ethnomedicine in Jammu and Kashmir (Aggarwal and Kotwal, 2009; Bhellum and Singh, 2012; Rashid, 2013; Bhardwaj *et al.*, 2019 and Pant and Wani, 2020) but studies on psychomedicinal plants were few numbered. District Udhampur, placed in the Jammu is hilly terrain and a vegetational-rich region comprising of different altitudinal zones *i.e.*, tropical, subtropical, temperate and alpine zones. The local residents of the district are still relying on their traditional medical knowledge and religious practices to get rid of mental health-related problems due to the remoteness of the area from modern healthcare facilities. Dangwal *et al.*, 2021 provided

ethnomedicinal uses of 27 plants used traditionally for curing mental illnesses in the Udhampur district.

The documentation of such valuable traditional knowledge is important because of its gradual disappearance from our society. In this communication, an effort has been made to gather information on distinct uses of psychomedicinal plants in Udhampur district of Jammu and Kashmir. Plants utilized directly as medicine and those used in religious and spiritual practices were documented in this study.

### STUDY AREA

The research was carried out in the district Udhampur of Jammu and Kashmir. The Shivalik range of the Himalayas encompasses the Udhampur district, which

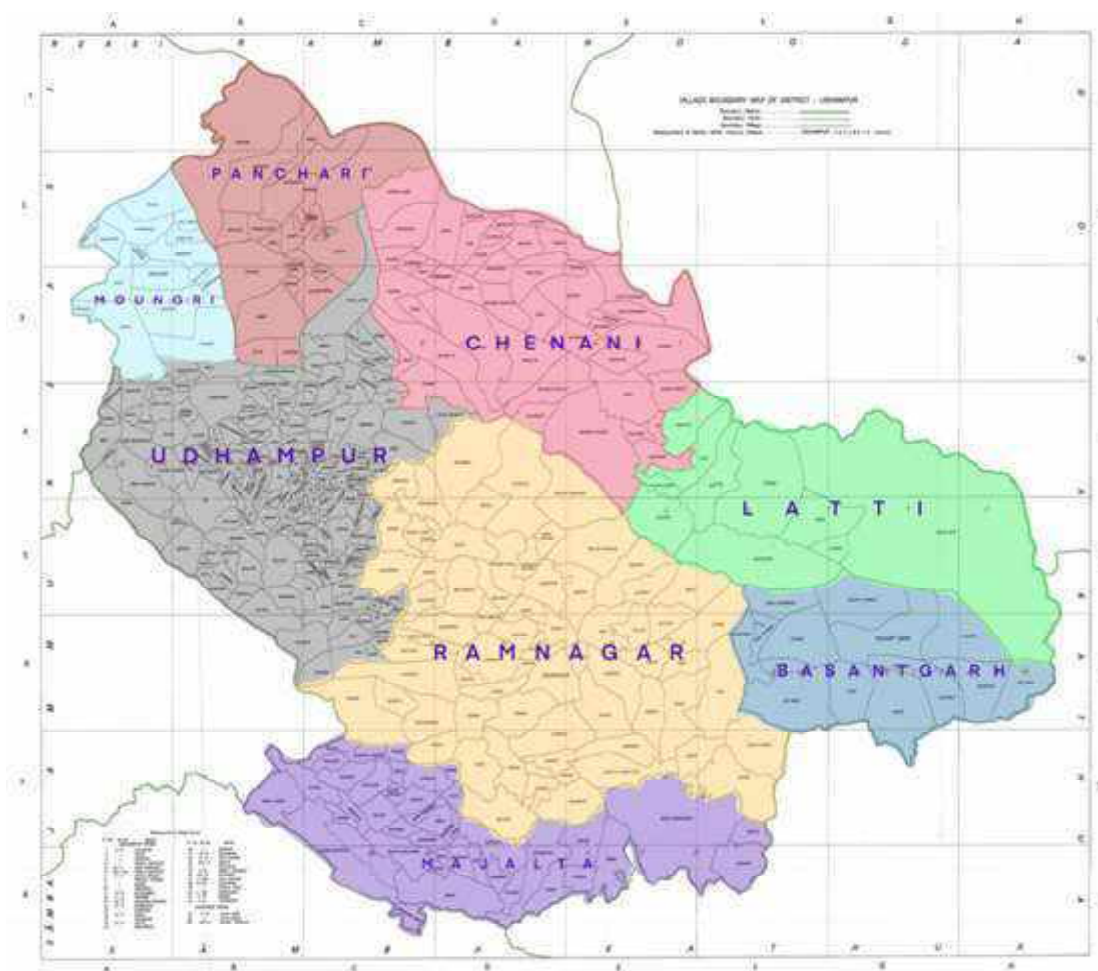


Fig. 1 : Map showing the tehsils of Udhampur district, Jammu and Kashmir (<https://udhampur.nic.in/map-of-district>)

is primarily mountainous. It is situated in the state of Jammu and Kashmir South-eastern region and is bordered by the districts of Rajouri in the West, Anantnag in the North, Doda in the North-East, Kathua in the South-East and Jammu in the South-West. (Murtaza *et al.*, 2019). Geographical coordinates range between 32°34' N to 39° 30' N latitudes and 74°16' E to 75°38' longitudes and encompasses an area of 4,550 square kilometres. Its altitude range varies between 600 to 3000 meters above sea level. During the winter, the district's upper reaches are blanketed in snow and temperature vary between 0° C to 35° C in the snowfall zone. In summer hot and humid weather, temperature ranges from 23° C to 42° C. Administratively the district is divided into 4 subdivisions, 8 tehsils, 17 blocks and 357 settlements. The distinct sampling sites included Basant Garh, Chowki, Beernoo, Dalsar, Dhiari, Dudu, Jandrore, Nalla Malion and Kutwalt (<https://udhampur.nic.in/map-of-district>).

#### METHODOLOGY

The present work has been done by making several visits to the study area during the time period of a year, *i.e.*, from 2021 to 2022. An intensive and extensive field survey was conducted and meetings with indigenous people of different villages were carried out during different seasons and months. The present communication is mainly concentrated on interviews and discussions with well-knowledgeable local people, Vaidyas, Hakims, shepherds, local inhabitants, aged peoples, nomadic, religious people, peasants and housewives with respect to medicinal plants used traditionally to cure mental illnesses or that used for religious rituals and sorcery practices. The point count method was used during the survey; time data was collected by direct count, dead specimens and some photographic records had been collected. The collected information on medicinal plants includes botanical names, families, vernacular names, habits, plant parts used and their medicinal uses. The relevant floras, like 'The Flora of Jammu and Kashmir' (Singh *et al.*, 2002), 'Flora of the District Garhwal, North West Himalaya' (Gaur, 1999), 'The Flora of British India' (Hooker, 1897), *etc.*, were consulted for verification.

#### RESULTS AND DISCUSSION

A total of 43 psychomedicinal plant species belonging to 40 genera and 33 families had been identified, out of which 31 species were dicotyledons and 12 monocotyledons. These species are enlisted alphabetically in (Table 1, 2 and 3) along with relevant information like botanical name, family common name, part used, uses, *etc.*. The most dominant family was Poaceae (monocotyledon) having 5 plant species, followed by Moraceae (dicotyledon) and Rosaceae (dicotyledon) with 3 plant species each. Other most dominant families were Acoraceae (2), Alliaceae (2) and Asteraceae (2). Least dominant plant families were Amaranthaceae, Acanthaceae, Asparagaceae, Apiaceae, Berberidaceae, Betulaceae, Nyctaginaceae, Bombacaceae, Cannabaceae, Zingiberaceae, Convolvulaceae, Phyllanthaceae, Geraniaceae, Euphorbiaceae, Musaceae, Solanaceae, Lamiaceae, Piperaceae, Santalaceae, Pedaliaceae, Urticaceae, Scrophulariaceae, Fabaceae, Verbenaceae, Rutaceae and Rhamnaceae. Among the total recorded species, 43 shrubs (60%), 19 were trees (26%) and 11 were herbs (14%) (Fig. 4). In (Fig. 5) the local priest performing exorcism in various steps by activating psychomedicinal plants to get rid off negative energies from ghostly effected humans body. Psychomedicinal plants were used in the treatment of epilepsy, anorexia, headache, hypertension, dementia and depression. Employment of these plants were also observed in practicing religious rites and rituals, worshipping supernatural powers, sorcery, exorcism, *etc.*. Local people believe in 'mantra' and 'tantra' practices to ward off evil eyes, expel ghost influence and sway away negative energy. They have faith in these religious and spiritual rituals to treat various mental health-related problems (Table 1,2,3). The method of preparation documented were *i.e.*, dried rhizome, decoction, paste, juice and dried powder. The common plant parts used were rhizome, bulb, tuber, bark, root, stem, leaves, flower, flower bud, fruit, seed and whole plant. (Table 1,2 and 3).



TABLE 1  
List of herbs used as psychomedicinal plants in Udhampur district

Botanical Name	Family	Common Name	Parts Used	Uses
<i>Acorus calamus</i> L.	Acoraceae	Barian, Sweet Flag	Rhizome	180 gm of dried rhizome powder mixed with 2 to 3 teaspoons of honey is highly effective to cure epilepsy and anxiety.
<i>Achyranthes aspera</i> L.	Amaranthaceae	Puthkanda, Prickles Chaff	Root, Stem, Seed	Decoction of root is given to cattle to treat anorexia. The stem is used in exorcism. The dried powder of seeds is used for inhalation to cure headaches and migraine.
<i>Allium cepa</i> L.	Alliaceae	Pyaz, Onion	Bulb, Seeds	Decoction of bulb mixed with seeds of <i>Coriandrum sativum</i> helps to improve intelligence. Seeds are used to prepare tea for curing insomnia.
<i>Allium sativum</i> L.	Alliaceae	Thom, Garlic	Bulb	Decoction of bulbs and leaves is used to cure hysteria and epilepsy.
<i>Arisaema flavum</i> (Forsk.) Schott.	Araceae	Saap-Kukdi, Yellow Cobra Lily	Tuber	Paste of tuber is used in the preparation of medicine for snake bite.
<i>Asparagus racemosus</i> Willd.	Asparagaceae	Sainsmaya, Shatavar	Whole plant	The entire plant is used to remove negative energies through sorcery and hymn practices.
<i>Centella asiatica</i> (Urb L.)	Apiaceae	Brahmi, Indian Penny Wort	Leaves	Decoction of leaves is used to improve cognition power and relieve anxiety. The paste of fresh leaves is applied to the forehead to get rid of headache.
<i>Bambusa arundinacea</i> Willd.	Poaceae	Chmathi, Thorny Bamboo	Root, Stem	Decoction of root is taken orally, releases kidney stones. The stem is used in funeral rites.
<i>Boerhavia diffusa</i> L.	Nyctaginaceae	Itt-sitt, Hog-Weed	Whole plant, Root	The entire plant is utilized for sorcery and ritual practices.
<i>Cannabis sativa</i> L.	Cannabaceae	Pang, True Hemp	Leaves	Decoction of leaves is used for the ailment of epilepsy, sleep disorders depression and anxiety.
<i>Curcuma longa</i> L.	Zingiberaceae	Aldhar, Turmeric	Rhizome	Rhizome powder is used in religious, rituals and skin care.
<i>Cuscuta reflexa</i> Roxb.	Convolvulaceae	Aandal, Dodder	Whole plant	Exorcism, sorcery and rituals involve the usage of this plant.

Botanical Name	Family	Common Name	Parts Used	Uses
<i>Cynodon dactylon</i> (L.) Pers.	Poaceae	Khabbal, Bermuda Grass	Whole plant	The whole plant is used for worshipping of Lord Ganesha and a clump of the shoot is used in the pooja ceremony.
<i>Geranium wallichianum</i> D. Don ex Sweet.	Geraniaceae	Laljari, Robert Geranium	Roots	Roots are used for exorcism.
<i>Hordeum vulgare</i> L.	Poaceae	Jou, Barley	Seed	Seeds are used for worship of supernatural powers.
<i>Musa paradisiaca</i> L.	Musaceae	Kela, Banana	Leaves, Fruit	The leaves are generally used to cure insanity. Leaves are generally used to drive away evil spirits (ghosts) from the houses. Fruit is directly used as remedy to remove hypertension.
<i>Nicotiana tabacum</i> L.	Solanaceae	Tmbaku, Wild Tobacco	Leaves	Tobacco leaves are smoked in sorcery practices.
<i>Oryza sativa</i> L.	Poaceae	Dhaan, Paddy	Seeds	During exorcism practice, rice grains mixed with urad dal are revolved over the head of the possessed person and then thrown in all four directions.
<i>Ocimum tenuiflorum</i> L.	Lamiaceae	Tulsi, Holy Basil	Whole plant, Leaves	The whole plant is used daily for morning Hindu prayers. Leaves are directly used in several religious ceremonies.
<i>Piper nigrum</i> L.	Piperaceae	Mirchi, Black pepper	Seeds	Heat the seeds, make a powder and mix with 1-2 teaspoons of honey, take it orally for dementia and sweetness of voice.
<i>Sesamum orientale</i> L.	Pedaliaceae	Till, Sesame	Seed oil	Seed oil is widely used in worshipping super natural powers.
<i>Tagetes erecta</i> L.	Asteraceae	Gutti, Marigold	Flower	It is used for offering the prayer to the local God for happiness and inner mental peace.
<i>Triticum aestivum</i> L.	Poaceae	Gehun, Wheat	Seeds	Grains are used in exorcism & sorcery practices.
<i>Urtica dioica</i> L.	Urticaceae	Jujuli, Stinging Nettle	Twigs, leaves	Twigs are used in exorcism. Leaves are used for cooking to cure hypertension and joint pain.
<i>Verbascum thapsus</i> L.	Scrophulariaceae	Desitmaku, Cow's Lungwort	Leaves	The leaves are used by ritual performers as a cloud of smoke to drive away evil spirits (ghosts) from the houses.
<i>Vigna mungo</i> (L.) Hepper	Fabaceae	Urd, Black Gram	Seeds	Seeds were mixed with rice grains and revolved overhead seven times of affected person to drive away evil spirits.

TABLE 2  
List of shrubs used as psycho-medicinal plants in Udhampur district

Botanical Name	Family	Common Name	Parts Used	Uses
<i>Adhatoda zeylanica</i> Medikus.	Acanthaceae	Brenker, Malabar Nut	Flower, Leaves	The leaves, heated and tied-on joints, fade away the pain. The honey is blended with dried powder of flower administered orally to treat epilepsy.
<i>Artemisia maritima</i> L.	Asteraceae	Mooiin, Sea Wormwood	Whole plant	Plants are considered sacred and through religious practices used in insanity.
<i>Berberis lycium</i> Royle.	Berberidaceae	Kamblu, Barberry	Stem, Root	Rituals are performed with the stem. The root decoction is administered orally for the treatment of internal wounds.
<i>Rosa macrophylla</i> Lindl.	Rosaceae	Gulab, Wild Rose	Twigs, Flower	Twigs are directly used for exorcism. The flower is used for fragrance.
<i>Rosa brunonii</i> Lindl.	Rosaceae	Karir, Himalayan Musk Rose	Stem with leaves	Stem with leaves is used for exorcism. It is highly used in rituals.
<i>Vitex negundo</i> L.	Verbenaceae	Bana, Chinese Chaste Tree	Twigs	The twigs are used in exorcism practices.
<i>Ziziphus mauritiana</i> Lam.	Rhamnaceae	Ber, Jujube	Fruits	Fruits are directly eaten to relieve stress and anxiety. In marriages, bride and groom revolve around the plants seven times. Delicious rice cooked and mixed with pulses (klichdi) and cooked roti of wheat flour (Dhropd) is given to crows for happiness of local deity (kuldevta).

TABLE 3  
List of trees used as psychomedicinal plants in Udhampur district

Botanical Name	Family	Common Name	Parts Used	Uses
<i>Betula utilis</i> D. Don	Betulaceae	Bhojpatra, Indian Birch	Bark	The bark is used as paper for writing religious texts and mantras and also used for making amulets for children to prevent evil energies.
<i>Bombax ceiba</i> L.	Bombacaceae	Simbal, Silk Cotton Tree	Bark & Flower bud	Bark decoction is orally taken to fight against fever. The bud of the flower and the calyx of not fully opened flowers are cooked and eaten as vegetable.
<i>Phyllanthus emblica</i> L.	Phyllanthaceae	Ambala, Indian Gooseberry	Fruit pulp	Fruit pulp is applied to the forehead to treat headaches and dizziness.
<i>Ficus religiosa</i> L.	Moraceae	Barh, Peepal	Leaves, Wood	Leaves and wood are used during worship of various supernatural powers to reduce its effect. It is considered a sacred plant.
<i>Ficus auriculata</i> Lour.	Moraceae	Trimbal, Elephant Ear Fig Tree	Whole plant, Leaves	The entire plant is used in religious rites. Leaves are directly used in rituals.
<i>Ficus benghalensis</i> L.	Moraceae	Borh, Banyan Tree	Whole plant	The whole plant is used for religious rituals and it is considered a sacred tree.
<i>Mallotus philippensis</i> (Lam.) Muell. Euphorbiaceae	Plu, Monkey Face Tree	Flower, Leaves	Flower, Leaves	Flowers are used for rituals and marriages. Leaves are directly used in marriage often during the "kanyadaan" ceremony.
<i>Prunus cerasoides</i> D. Don.	Rosaceae	Batarn, Himalayan Wild Cherry	Bark	An amulet is made from bark to prevent harm from bad energies.
<i>Santalum album</i> L.	Santalaceae	Chandan, East Indian Sandalwood	Wood	The wood is often used in rituals and marriages.
<i>Zanthoxylum armatum</i> DC.	Rutaceae	Timbru, Winged Prickly Ash	Wood	The wood is used for exorcism.

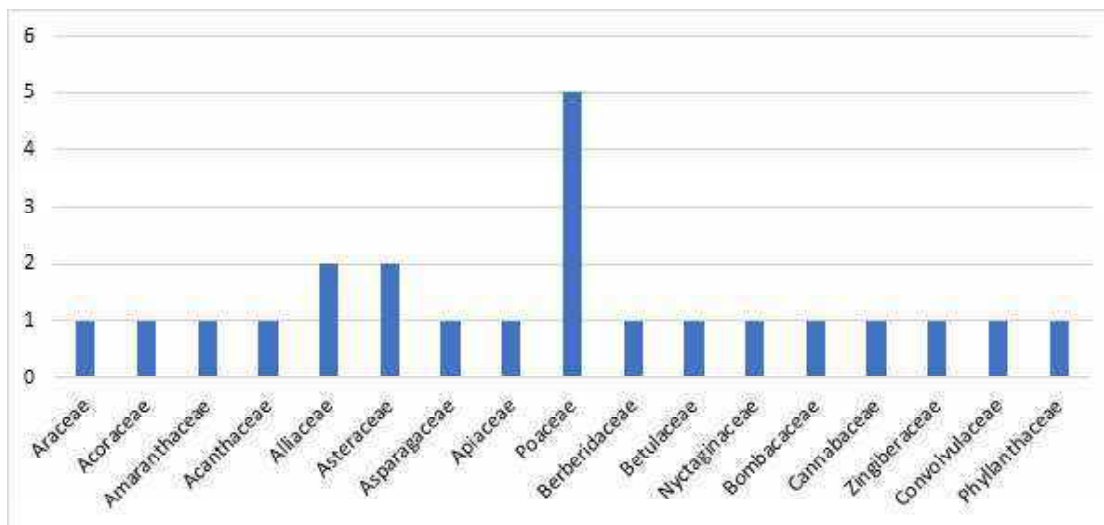


Fig. 2 : Graph chart showing the number of psychomedicinal plant species in each family

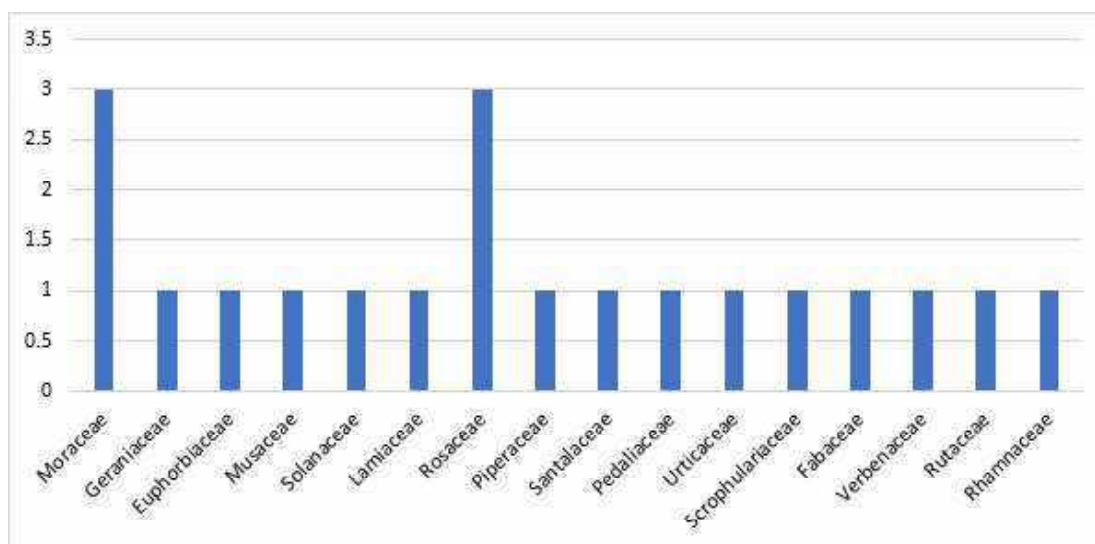


Fig. 3 : Graph chart showing the number of psychomedicinal plant species in each family

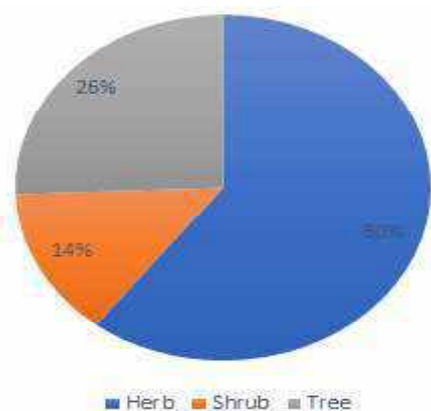


Fig. 4 : Pie chart showing the percentage of herbs, shrubs and trees

In the current research, the authors conclude that different psychomedicinal plants are traditionally used by Vaidyas, Hakims, tribes, shepherds, old age men and women for the purpose of treating different mental ailments in the study area. Necromancer and Voodooists used these psychomedicinal plants for treating various mental health problems through sorcery. Local inhabitants used these plants for worshipping supernatural powers and in various religious practices. These psychomedicinal plants have a huge scope for commercial purposes and may contribute to generating employment for local people.



Fig. 5 : Photographs show a priest performing exorcism using psychomedicinal plants

The traditional knowledge that is passed from generation to generation among the local inhabitants needs to be maintained, secured and protected. Further research on local informants is required to assess the traditional knowledge and its status among different generations. Community knowledge should be raised through various awareness programmes about mental health issues. Further, phytochemical and pharmacological analysis needs to be done to examine the bioactive compounds responsible for treating neuro-disorders.

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## Morphological Characterization of Selected Land Races of Rice (*Oryza sativa* L.)

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### ABSTRACT

Increasing interest in the descriptive characterization of plant varieties in the context of intellectual property rights is stimulated by the recent agreements within the framework of the World Trade Organization (WTO). The requirements of these activities vary, the varietal registration process (involving testing for distinctness, uniformity and stability-DUS) requires that a description of a newly bred variety be produced and compared with all existing varieties of common knowledge. Thirty one land races of rice (29 land races and 2 check varieties) maintained at Organic Farming Research Centre, UAHS, Navile, Shivamogga were utilized for the present study. These land races were raised in RCBD replicated thrice, they were evaluated for 43 morphological characters (9 quantitative & 34 qualitative) during *khari* 2021. The local landraces exhibited sufficient genetic variation for most of the traits. There were 43 descriptors studied, out of which, six characters were found monomorphic, while rest of the characters showed polymorphic variations among the landraces. The genetic potential of the land races for the desired traits can be utilized in hybridization programme to select promising genotypes.

*Keywords* : Morphological, Characterization, Land races, Rice, Qualitative, Quantitative

RICE (*Oryza sativa* L.) plays a vital role in the national food security in India and contributes major source of calories for the urban and rural population. Rice belongs to the family graminiae, recognized as 'Millennium Crop' expected to contribute towards food security in the world; It is one of the staple cereal crops and a primary source of food for more than half of the world's population (Ramesh Channannavar, 2019). The slogan 'Rice is Life' is most appropriate for India because this crop provides a living for millions of rural households (Renuka *et al.*, 2022). Rice has a large number of native varieties and landraces having unique characteristics and great adaptability and they are grown in different agro climatic zones. About 425,500 rice accessions conserved in various gene banks of the world are potential gene sources directed for crop improvement. India has a rich and wide range

of genetic wealth of rice. It has been estimated from various surveys that nearly 50,000 of rice land races is still being grown in the country (Roy *et al.*, 1985).

Major rice producing states in India are West Bengal, Uttar Pradesh, Punjab, Andhra Pradesh, Odisha, Tamil Nadu and Madhya Pradesh. In world rice is grown in an area of 167.1 million hectares with the production and productivity levels of 782 million tonnes and 4678 kg per hectare, respectively. In India, rice is grown in an area of 46.15 million hectares with the production and productivity levels of 116.47 million tonnes and 2638 kg per hectare, respectively. In Karnataka rice is grown in an area of 1.13 million hectares with the production and productivity levels of 3.43 million tonnes and 3012 kg per hectare, respectively (Anonymous, 2020).



Introduction of high yielding varieties were released with new technologies have become a great threat to the security of the age-old practice of growing traditional varieties and landraces which may have immense potential for different important traits (Song *et al.* 1999). The existing UPOV models of plant variety protection were not suitable for Indian requirements. The Government of India enacted our own legislation on the 'Protection of Plant Varieties and Farmers Act' (PPV & FRA) in 2001 for providing protection to plant varieties based on distinctness, uniformity and stability (DUS) test apart from novelty. Which is a unique and model act gives equal importance to the farmers and breeders and treats them as partners in their efforts for sustainable food security (Patra, 2000).

The ability to distinguish and clearly identify the varieties of cultivated species is fundamental for the operational aspects in the seed trade. The new varieties developed in agricultural and horticultural crops should be distinct from other varieties, with the introduction of Indian legislation on 'The Protection of Plant Varieties and Farmer's Rights (PPV & FR) Act, 2001'. Morphological characterization of the released varieties and landraces helped in developing the database based on which new varieties developed can be distinguished and the characterization would also help in assessment of genetic diversity existing in the landraces and released varieties. Internationally, DUS testing is co-ordinated by the International

Union for the Protection of New Varieties of Plants (UPOV), which produces guidelines detailing list of characters to be used for examination of different species. The present study was conducted to characterize the selected land races of rice based on DUS characters.

#### MATERIAL AND METHODS

A field experiment was conducted during *kharif*, 2021 at Organic Farming Research Centre (OFRC), University of Agricultural and Horticultural Sciences, Shivamogga. The research station is situated at 140 0' to 140 1' North latitude and 750 40' to 750 42' East longitude and at an altitude of 650 meters above the mean sea level. Morphological characterization of 31 land races (29 land races and 2 check varieties) of rice was done using 43 morphological traits (Plate 2 & 3). The land races were grown in RCBD design (Sundaraju *et al.*, 1972) with two checks and 29 landraces in three replications during *kharif* 2021 (Plate 1). Each land race was transplanted in five rows of 3m row length at spacing of 30 cm between rows and 10 cm between plants. Crop was raised by following recommended package of practices. Observations were recorded on 5 randomly chosen plants of each landrace per replication for 43 morphological traits.

Among the qualitative character, 34 visually assessed characteristics were observed according to the National Test Guidelines for DUS test in rice which



Plate 1 : General view of the experimental plot



Plate 2: Selected elite land races of rice seeds were used for the study



Plate 3: Selected elite land races of rice seeds used for the study

was developed by Directorate of Rice Research Rajendarnagar, Hyderabad (Shobha Rani *et al.* 2004). The observation of various characteristics was recorded at different stages of growth with appropriate procedures as per the DUS test guide lines of PPV & FR Act, 2001. Like UPOV, in PPV and FR Act, a variety must fulfill the criteria of Distinctness, Uniformity, Stability and Novelty to get protection under this act (Anonymous 2001). The characters studied were Coleoptile colour, Basal leaf sheath colour, Intensity of green colour of leaf, Anthocyanin colouration of leaf, Distribution anthocyanin colouration of leaf, Anthocyanin colouration of leaf sheath, Pubescence of Leaf blade surface, Auricles, Anthocyanin colouration of collar, Leaf ligule, Shape of ligule, Colour of ligule, Culm attitude, Attitude of Flag leaf blade (early and late), Spikelet sterility, Spikelet: Density of pubescence of lemma, Anthocyanin colouration of keel, Anthocyanin colouration of area below apex, Anthocyanin colouration of apex, Colour of stigma, Anthocyanin colouration of nodes, Anthocyanin colouration of inter nodes, Curvature of Panicle main axis, Colour of tip of lemma, Panicle awns, distribution of awns, Presence of secondary branching, Attitude of branches, Panicle exertion and Sterile lemma colour, Leaf: Length of blade, Leaf: Width of blade, Time of heading (50% of plants with panicles), Stem: thickness, Stem: Length (excluding panicle), Panicle: length of longest awns, Panicle: length of main axis, Panicle: number per plant & Time of harvest (days).

## RESULTS AND DISCUSSION

Qualitative and quantitative characters were considered as marker characters in the identification of land races of rice, which are less influenced by environmental fluctuations. The 43 morphological characters (Table 2) were recorded in 31 rice land races. The qualitative characters which are less influenced by the environmental factors are used as morphological markers for the identification of rice land races (Rao *et al.*, 2013 and Kalyan *et al.*, 2017).

TABLE 1

List of land races of rice used for the study

Sl. No.	Name of Land races	Sl. No.	Name of Land races
1	Jasmine	17	Mysurumallige
2	Ratnachudi	18	Rajamudi
3	Nazarbad	19	Gowrisanna
4	Rajabhoga	20	Barmablack
5	Gandhasale	21	Doddabairunellu
6	Bangarasanna	22	Kempusale
7	Champakali	23	Navara
8	Dappavalya	24	Kempujiddu
9	Raichur sanna	25	Anekombinabhatta
10	Madras sanna	26	Kiruvani
11	Karigajavale	27	Rathnasagara
12	Karijiddu	28	Misebhatta
13	Neregulibhatta	29	Ambemohari
14	Puttabhatta	30	Jyothi-C
15	Jeerigesanna	31	MTU-1001-C
16	Gilisale		

The coleoptile colour varied among 31 rice land races studied as colorless and purple. Among the 31 land races 29 (94%) showed colorless and 2 (6%) (Nazarbad & Puttabhatta) land races had purple. Similar results were reported earlier by Priyanga *et al.* (2020). While colour of basal leaf sheath, among the 31 landraces 23 (74%) exhibited presence of green, 2 (7%) (Karigajavale and Puttabhatta) light purple, 5 (16%) purple lines and 1 (3%) (Nazarbad) land race showed uniform purple. Intensity of green colour, 20 (64%) land races found light green colour, 7 (23%) were medium and 4 (14%) were dark green colour. Lahkar and Tanti (2017) also reported similar results in land races. Out of thirty-one landraces 5 had leaf anthocyanin colouration, while distribution of anthocyanin coloration out of 5 land races, 4 (80%) landraces showed in margin and 1 (20%) (Nazarbad) had uniform purple colour similar result was reported earlier by Umarani *et al.* (2017). Anthocyanin colouration of leaf sheath was present in nine (29%) land races and absent in 22 (71%) land races (Table 3 & Plate 4). Pubescence of leaf blade surface, 15 (48%) were weak and 16 (52%) were medium. Leaf auricle (100%), collar (100%) and ligule (100%) present in all 31 landraces. For anthocyanin colouration of auricle, 28 (90%) land

TABLE 2  
Essential characters along with DUS descriptor

Characters	Strategies
Coleoptile colour	Green
Basal leaf: Sheath colour	Light purple
Leaf: Intensity of green colour	Medium
Leaf: Anthocyanin Coloration	Present
Leaf: Distribution of anthocyanin coloration	On tips only
Leaf Sheath: Anthocyanin colouration	Absent
Leaf: Pubescence of blade surface	Absent
Leaf: Auricles	Absent
Leaf: Anthocyanin colouration of auricles	Colourless
Leaf: Collar	Absent
Leaf: Anthocyanin colouration of collar	Absent
Leaf: Ligule	Absent
Leaf: Shape of Ligule	Truncate
Leaf: Colour of ligule	Membrane white
Leaf: Length of blade	Short (<30 cm)
Leaf: Width of blade	Narrow (<1 cm)
Flag leaf: Attitude of blade (early observation)	Erect
Flag leaf: Attitude of blade (late observation)	Erect
Culm: Attitude	Erect
Time of heading (50% of plants with panicles)	Very early(<71 days)
Spikelet sterility	Absent
Stem: Thickness	Thin
Spikelet: Density of Pubescence of lemma	Absent
Lemma anthocyanin coloration of keel	Absent
Lemma anthocyanin coloration of area below apex	Absent
Lemma anthocyanin coloration of area apex	Absent
Spikelet: Colour of stigma	White
Stem: Length (excluding panicle)	Very short (<91 cm)
Stem: Anthocyanin colouration of nodes	Absent
Stem: Anthocyanin colouration of inter- nodes	Absent
	Colour less
	Green
	Light purple
	Medium
	Present
	On margins only
	Present
	Weak
	Present
	Light purple
	Present
	Present
	Acute
	Light purple
	Medium (30-45 cm)
	Medium (1-1.5 cm)
	Semi-erect
	Semi-erect
	Semi-erect
	Early (71-90 days)
	Present
	Medium
	Weak
	Weak
	Weak
	Weak
	Light green
	Short (91-110 cm)
	Present
	Present
	Purple
	Purple lines
	Dark
	-
	In blotches only
	-
	Medium
	-
	Purple
	-
	-
	Split
	Purple
	-
	Long (>45 cm)
	Broad(>1.5cm)
	Horizontal
	Horizontal
	Open
	Medium (91-110 days)
	-
	-
	Thick
	Medium
	Weak
	Weak
	Weak
	Weak
	Light green
	Short (91-110 cm)
	Present
	Present
	Yellow
	Medium (111-130 cm)
	-
	-
	-
	Uniform purple
	-
	-
	-
	Uniform
	-
	Strong
	-
	Very strong
	-
	Deflexed
	Deflexed
	Spreading
	Late (111-130 days)
	-
	-
	Very strong
	Very strong
	Very strong
	Very strong
	Very strong
	Light purple
	-
	-
	-

Table 2 Continued....

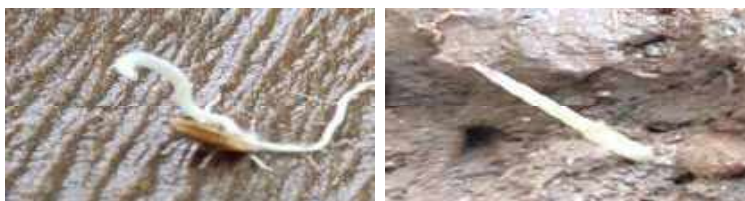
Characters	Strategies			
Panicle: Curvature of main axis	Straight	Semi-straight	Drooping	Deflexed
Spikelet: Colour of tip of lemma	White	Yellowish	Brown	Purple
Panicle: Awns	Absent	Present	-	-
Panicle : Distribution of awns	Tip only	Upper half only	Whole length	-
Panicle: length of longest awns	Very short	Short	Medium	Long
Panicle : Presence of secondary branching	Absent	Present	-	-
Panicle: Secondary branching	Weak	Strong	Clustered	-
Panicle: Exertion	Partly exerted	Mostly exerted	Well exerted	-
Sterile lemma: Colour	Straw	Gold	Red	Purple
Panicle: Attitude of branches	Erect	Erect to semi-Erect	Semi-erect	Semi-erect to spreading
Panicle: length of main axis	Very short (<16 cm)	Short (16-20 cm)	Medium (21-25 cm)	Long (26-30 cm)
Panicle: number per plant	Few (<11)	Medium (11-20)	Many (>20)	-
Time of harvest (days)	Early (101-120)	Medium (121-140)	Late (141-160)	Very late (>160)

racess were colourless, 2 (Barmablack & Nazarbad) land races were purple (7%) and one (Karigajavale) land race was light purple (3%) and it was similar with earlier reports by Sakthi Avinash *et al.* (2019). Among the 311 and racestwo (Karijiddu & Karigajavale) land races had anthocyanin colouration at collar of leaf (6%) and 29 (94%) had no anthocyanin colouration at collar of leaf. All the land races were having split shape of ligule (100%). Two (Barmablack & Nazarbad) land race posses purple colour of ligule (7%), 24 (77%) were membranous white and 5 (16%) were of light purple. With respect to length of leaf blade, the 31 land races classified 3 (19%) were short, 22 (71%) were medium and 6 (10%) were long, whereas, for width of leaf blade, 26 (84%) were narrow, 4 (13%) were medium and 1 (3%) was broad (Table 3A, 4 & Plate 5 & 6). Culm attitude of land races, five (16%) were erect, 8 (26%) were semi erect, 10 (32%) were open type and 8 (26%) were spreading. For time of 50 per cent heading two (6%) land races were very early, 15 (48%) land races early, 11 (36%) land races were medium and three (10%) land races were late. Flag leaf attitude of blade (early), 18 (58%) land races were erect, 12 (39%) land races were semi erect and one (Ambemohari) land race was deflexed (3%). However, flag leaf attitude of blade (late), 16 (51%) land races were erect, 8 (26%) land races were semi erect, four (13%) land races were horizontal and three (10%) (Ambemohari, Puttabhatta & Rajamudi) landraces weredeflexed (Table 3A & 4). Similar results were notified by (Kalyan *et al.*, 2017 and Keerthivarman *et al.*, 2019). Spikelet sterility was absent in all 31 land races.

For anthocyanin colouration of lemma keel was absent for 22 (71%) land races while present in nine (29%) land races, out of nine, seven (23%) had medium anthocyanin colouration and two (6%) (Karigajavale & Barmablack) had strong intensity. Anthocyanin colouration of area below apex of lemma was absent in twenty land races (65%), while nine (29%) land races were medium coloured and two (6%) (Karigajavale and Barmablack) land races showed strong colour. Anthocyanin colouration of apex was absent in 19 (61%) land races, whereas, ten (32%) had medium, two (7%) (Karigajavale & Barmablack)

**TABLE 3**  
Per cent distribution in selected land races of rice for various morphological characters

Characters	Status	No. of rice landraces	Contribution of number of land races (%)
Coleoptile colour	Colour less	29	94
	Purple	2	6
Basal leaf: Sheath colour	Green	23	74
	Light purple	2	7
	Purple lines	5	16
	Uniform purple	1	3
Leaf: Intensity of green colour	Light	20	64
	Medium	7	23
	Dark	4	13
Leaf: Anthocyanin Coloration	Absent	26	84
	Present	5	16
Leaf: Distribution of anthocyanin coloration	On margins only	4	80
	Uniform	1	20
Leaf Sheath: Anthocyanin colouration	Absent	22	71
	Present	9	29
Leaf: Pubescence of blade surface	Weak	15	48
	Medium	16	52
Leaf: Auricles	Absent	0	0
	Present	31	100
Leaf: Anthocyanin colouration of auricles	Colourless	28	90
	Light purple	1	3
	Purple	2	7



Colourless

Purple

**Coleoptile colour**



Green

Light Purple

Purple lines

Uniform purple

**Basal leaf sheath colour**

Plate 4: Coleoptile colour, basal leaf sheath colour and intensity of green colour of land races of rice

TABLE 3A

Per cent distribution in selected land races of rice for various morphological characters

Characters	Status	No. of rice land races	Contribution of number of land races (%)
Leaf: Collar	Absent	0	0
	Present	31	100
Leaf: Anthocyanin colouration of collar	Absent	29	94
	Present	2	6
Leaf: Ligule	Absent	0	0
	Present	31	100
Leaf: Shape of Ligule	Split	31	100
Leaf: Colour of ligule	White	24	77
	Light purple	5	16
	Purple	2	7
Leaf: Length of blade	Short (<30 cm)	3	19
	Medium(30-45 cm)	22	71
	Long (>45 cm)	6	10
Leaf: Width of blade	Narrow (<1 cm)	26	84
	Medium (1-1.5 cm)	4	13
	Broad(>1.5cm)	1	3
Flag leaf: Attitude of blade (early observation)	Erect	18	58
	Semi-erect	12	39
	Horizontal	0	0
	Deflexed	1	3
Flag leaf: Attitude of blade(late observation)	Erect	16	51
	Semi-erect	8	26
	Horizontal	4	13
	Deflexed	3	10
Culm: attitude	Erect	5	16
	Semi-erect	8	26
	Open	10	32
	Spreading	8	26

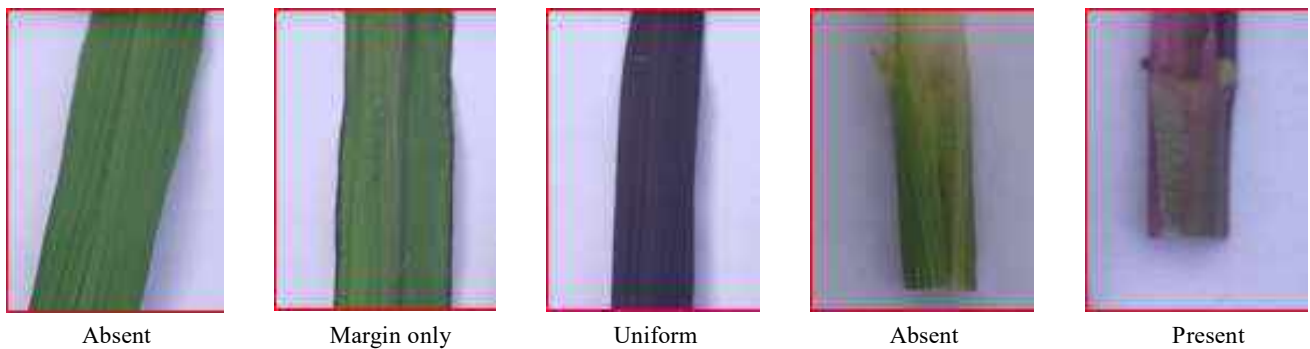
Characters	Status	No. of rice land races	Contribution of number of land races (%)
Time of heading (50% of plants with panicles)	Very early(<71 days)	2	6
	Early(71-90 days)	15	48
	Medium(91-110 days)	11	36
	Late(111-130 days)	3	10
Spikelet sterility	Absent	31	100
	Present	0	0

TABLE 3B  
Per cent distribution in selected land races of rice for various morphological characters

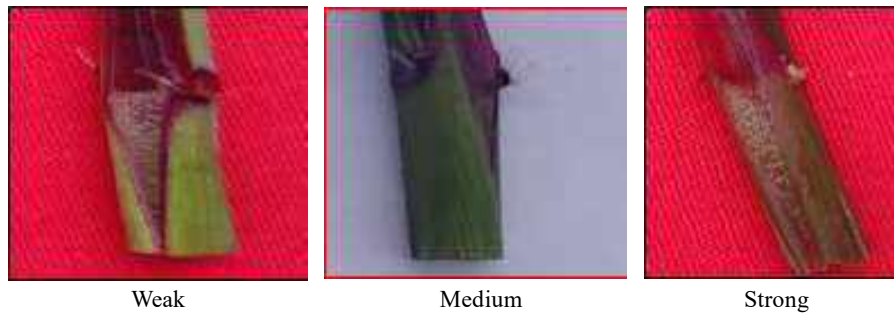
Characters	Status	No. of rice Land races	Contribution of number of land races (%)
Lemma anthocyanin coloration of keel	Absent	22	71
	Weak	0	0
	Medium	7	23
	Strong	2	6
Lemma anthocyanin coloration of area below apex	Absent	20	65
	Weak	0	0
	Medium	9	29
	Strong	2	6
Lemma anthocyanin coloration of area apex	Absent	19	61
	Weak	0	0
	Medium	10	32
	Strong	2	7
Spikelet: Colour of stigma	White	25	81
	Purple	6	19
Stem: Thickness	Thin (<0.45 cm)	16	52
	Medium (0.45-0.60 cm)	9	29
	Thick (>0.60 cm)	6	19
Stem: Length(excluding panicle)	Very short (<91 cm)	17	55
	Short (91-110 cm)	12	39
	Medium (111-130 cm)	2	6
Stem: Anthocyanin colouration of nodes	Absent	29	94
	Present	2	6
Stem: Anthocyanin colouration of inter- nodes	Absent	24	77
	Present	7	23
Panicle: Length of main axis	Very short(<16 cm)	6	19
	Short(16-20 cm)	6	19
	Medium(21-25 cm)	12	39
	Long (26-30 cm)	7	23



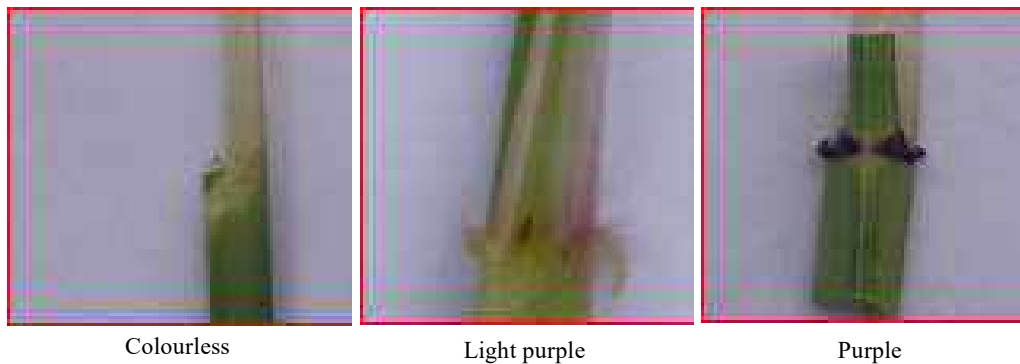
Characters	Status	No. of rice Land races	Contribution of number of land races (%)
Panicle: Curvature of main axis	Straight	2	6
	Semi-straight	4	13
	Dropping	13	42
	Deflexed	12	39
Panicle: Number per plant	Few (<11)	14	45
	Medium (11-20)	16	52
	Many (>20)	1	3



**Leaf anthocyanin colouration Leaf sheath anthocyanin colouration**



**Leaf sheath intensity of anthocyanin colouration**



**Leaf anthocyanin colouration of auricles**

Plate 5 : Anthocyanin colouration of leaf, leaf sheath and auricles of land races of rice



Absent



Present

**Leaf anthocyanin colouration of collar**



Membranous white



Light purple



Purple

**Leaf colour of ligule**



Long Medium Short



Narrow Medium Broad

**Leaf : length of the blade Leaf : width of the blade**

Plate 6 : Anthocyanin colouration of leaf collar, ligule and length of leaf blade and leaf width of land races of rice

had strong (Table 3B & 6). Similar results were reported by Ramesh Channannavar *et al.* (2020). Among the 31 land races 16 (52%) land races were thin, nine (29%) were medium and six (19%) land races had thick stem. Stem length was very short in 17 (55%) land races, short in 12 (39%) and medium

in two (6%) land races. For spikelet density of pubescence of lemma, 9 (29%) were weak, 18 (58%) were medium and 4 (13%) were strong. For colour of stigma, 25 (81%) were white and 6 (19%) were purple. The similar results were obtained by Subbu Rao *et al.* (2013).

TABLE 4  
Quantitative characters in selected land races of rice

Landraces	Length of leaf blade (cm)	Leaf: Width of blade (cm)	Time of heading (50% of plants with panicles)
Jasmine	63.00	1.00	115.00
Ratnachudi	49.90	0.90	99.00
Nazarbad	35.40	0.80	95.00
Rajabhoga	38.20	1.00	119.00
Gandhasale	24.30	0.60	94.00
Bangarasanna	30.50	0.80	105.00
Champakali	34.00	0.60	83.67
Dappavalya	38.60	1.00	83.00
Raichursanna	32.50	0.90	83.00
Madrassanna	32.50	0.80	90.00
Karigajavale	42.90	0.90	99.00
Karijiddu	44.80	0.80	103.00
Neregulibhatta	55.10	1.00	82.00
Puttabhatta	57.20	0.70	105.00
Jeerigesanna	38.70	0.80	95.00
Gilisale	42.00	0.90	95.00
Mysurumallige	25.00	0.60	78.00
Rajamudi	42.00	0.90	110.00
Gowrisanna	37.50	1.10	95.00
Barmablack	39.50	1.10	115.00
Doddabairunellu	37.40	0.70	79.00
Kempusale	32.50	0.90	78.00
Navara	23.00	0.50	67.00
Kempujiddu	41.60	0.60	83.00
Anekombinabhatta	31.00	1.10	83.67
Kiruvani	38.00	1.00	83.00
Rathnasagara	37.50	1.10	88.00
Misebhatta	46.60	0.90	83.00
Ambemohari	49.00	0.70	75.00
Jyothi	30.50	0.90	68.00
MTU-1001	31.00	1.00	85.00
Mean	38.76	0.86	90.85
SEm±	0.867	0.054	0.761
CD (P=0.05)	2.454	0.153	2.152
CV (%)	3.875	10.890	1.450

Anthocyanin colouration of node was absent in 29 (94%) landraces and present in two (Neregulibhatta & Misebhatta) landraces (6%). Anthocyanin colouration of inter node was present in seven (23%) landraces and absent for in 24 (77%) landraces. The identical results were summarized by (Ramesh Channannavar *et al.*, 2020). For panicle curvature of main axis, two (Doddabirunellu & Navara) were straight (6%), four (13%) were semi straight, 13 (42%) were drooping and 12 (39%) were deflexed. Among 31 land races with respect to length of panicle main axis, six (19%) were very short, six (19%) were short, 12 (39%) were medium and seven (23%) had long panicle. Fourteen (45%) land races were identified with few panicles per plant, 16 (52%) were medium and one (3%) had much number of panicles per plant. Based on the spikelet colour of tip of lemma, landraces were grouped into 13 (42%) white, five (16%) yellowish, five (16%) brown, three (10%) purple and five (16%) brown tawny (Table 5).

Panicle awns were present only in 5 (16%) land races out of 31 and absent in 26 (84%) land races. Distribution of awns in panicle, two (40%) had awns in tip only and three (Madrassanna, Kempusale & Misebhatta) had in whole length (60%). Out of the 5 awned landraces, one (20%) had very short, one (20%) had short, one (20%) had medium, one (20%) had long and one (20%) had very long in length of awns. Manjunatha *et al.* (2018) also reported the absence of awns in the landraces taken for the study whereas Chakravorty and Ghosh (2012) reported panicle distribution of awns at tip only in the landraces studied. All 31 land races showed presence of secondary branching of panicle, out of which, 12 (39%) land races were weak, 10 (32%) were clustered and 9 (29%) were strong. For the panicle attitude of branches, three (Doddabirunellu, Navara & Raichursanna) were erect (10%), five (Nazarbad, Rajabhoga, Dappavalya, Gilisale & Kiruvani) were erect to semi erect (16%), 10 (32%) were semi erect, 11 (36%) were semi erect to spreading and two (Bangarasanna & Gandasale) were spreading (6%). Based on the exertion of panicle, two (Bangarasanna & Kempusale) were mostly exertion (6%) and 29

TABLE 5  
Per cent distribution in selected land races of rice for various morphological characters

Characters	Status	No. of rice Land races	Contribution of number of land races (%)
Spikelet: Density of Pubescence of lemma	Absent	0	0
	Weak	9	29
	Medium	18	58
	Strong	4	13
Spikelet: Colour of tip of lemma	White	13	42
	Yellowish	5	16
	Brown	5	16
	Purple	3	10
	Brown tawny	5	16
Panicle: length of longest awns	Very short	1	20
	Short	1	20
	Medium	1	20
	Long	1	20
	Very long	1	20
Panicle: Awns	Absent	26	84
	Present	5	16
Panicle : Distribution of awns	Tip only	2	40
	Whole length	3	60
Panicle : Presence of secondary branching	Absent	0	0
	Present	31	100
Panicle: Secondary branching	Weak	12	39
	Strong	9	29
	Clustered	10	32
Panicle: Exertion	Partly exerted	0	0
	Mostly exerted	2	6
	Well exerted	29	94
Panicle: Attitude of branches	Erect	3	10
	Erect to semi-Erect	5	16
	Semi-erect	10	32
	Semi-erect to spreading	11	36
	Spreading	2	6
Sterile lemma: Colour	Straw	30	97
	Purple	1	3
Time of harvest (days)	Early (101-120)	1	3
	Medium(121-140)	18	58
	Late (141-160)	11	36
	Very late (>160)	1	3

TABLE 6  
Quantitative characters in selected land races of rice

Landraces	Stem: Thickness (cm)	Stem: Length (excluding panicle) cm	Panicle: Length of main axis (cm)	Panicle: Number per plant
Jasmine	0.49	70.50	27.20	15.00
Ratnachudi	0.39	92.00	23.00	20.00
Nazarbad	0.56	88.00	17.00	8.00
Rajabhoga	0.57	70.20	23.20	17.00
Gandhasale	0.51	90.00	9.10	13.00
Bangarasanna	0.42	92.00	24.00	12.00
Champakali	0.48	73.00	19.20	10.00
Dappavalya	0.67	104.00	23.00	8.00
Raichursanna	0.60	80.10	23.00	14.00
Madrassanna	0.43	61.00	17.20	7.00
Karigajavale	0.52	105.00	25.20	12.00
Karijiddu	0.10	85.00	22.20	17.00
Neregulibhatta	0.41	104.00	26.00	13.00
Puttabhatta	0.31	94.00	26.00	17.00
Jeerigesanna	0.44	127.00	25.20	11.00
Gilisale	0.61	116.00	26.20	17.00
Mysurumallige	0.41	44.00	20.00	13.00
Rajamudi	0.42	84.20	24.20	15.00
Gowrisanna	0.55	92.20	26.10	11.00
Barmablack	0.70	93.50	25.00	8.00
Doddabairunellu	0.32	70.00	16.00	8.00
Kempusale	0.36	52.00	18.30	11.00
Navara	0.31	65.00	15.00	9.00
Kempujiddu	0.27	77.50	17.20	9.00
Anekombinabhatta	0.63	98.50	24.10	11.00
Kiruvani	0.32	64.00	24.00	15.00
Rathnasagara	0.41	94.50	23.20	12.00
Misebhatta	0.60	100.00	16.00	15.00
Ambemohari	0.65	111.50	27.00	27.00
Jyothi	0.44	50.60	15.00	10.00
MTU-1001	0.62	62.00	23.30	11.00
Mean	0.47	84.24	21.65	12.77
SEm±	0.001	0.759	0.866	0.737
CD (P=0.05)	0.003	2.148	2.451	2.085
CV (%)	0.401	1.561	6.933	9.992

TABLE 7  
Quantitative characters in selected land races of rice

Landraces	Panicle: length of longest awns (cm)	Time of harvest (days)
Jasmine	0.707	163.00
Ratnachudi	0.707	150.00
Nazarbad	1.047	147.00
Rajabhoga	0.707	132.00
Gandhasale	0.707	143.00
Bangarasanna	0.707	153.00
Champakali	0.707	139.00
Dappavalya	0.707	140.00
Raichursanna	0.707	136.00
Madrassanna	2.423	142.00
Karigajavale	0.707	147.00
Karijiddu	0.707	154.00
Neregulibhatta	0.707	136.00
Puttabhatta	0.707	157.00
Jeerigesanna	0.707	135.00
Gilisale	0.707	138.00
Mysurumallige	0.707	134.00
Rajamudi	0.707	159.00
Gowrisanna	0.707	139.00
Barmablack	0.707	161.00
Doddabairunellu	0.707	136.00
Kempusale	1.394	134.00
Navara	0.707	125.00
Kempujiddu	0.707	138.00
Anekombinabhatta	0.947	136.00
Kiruvani	0.707	139.00
Rathnasagara	0.707	139.00
Misebhatta	3.029	132.00
Ambemohari	0.707	142.00
Jyothi	0.707	120.00
MTU-1001	0.707	137.00
Mean	0.88	141.39
SEm±	0.00046	0.955
CD (P=0.05)	0.00131	2.700
CV (%)	0.092	1.169

Note : Square root transformation for the parameter Panicle: length of longest awns (cm)

(94%) were well exertion of panicle. The correspondent results were recited by Subbu Rao *et al.* (2013). For sterile lemma colour, 30 (97%) had straw colour and one (Ambemohari) had purple (3%). For time of harvest, one (3%) landrace was early, 18 (58%) landraces were medium, 11 (36%) land races were late and one (3%) landrace was very late (Table 5 & 7).

The present study was conducted with 31 landraces and they exhibited 43 (9 quantitative & 34 qualitative) distinctive essential characters. Majority of them showed clear cut distinctive characters. Hence, these characters will be useful for developing future Varieties/ hybrids through conventional breeding methods. The information generated on these landraces also supports their registration with the PPV and FRA.

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## Morphological Screening of Jackfruit (*Artocarpus heterophyllus* Lam.) Genotypes for Vegetable Purpose

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### ABSTRACT

Twenty jackfruit genotypes with six and eight weeks old harvested fruits were screened for qualitative and quantitative characters that are suitable for vegetable purpose. Good diversity was noticed for qualitative characters like fruit shape and fruit rind colour. Among the quantitative characters studied, fruit length was found to be higher in genotype NSP-3 (33.67 cm) of six week old fruit and HV-1 (33.67 cm) of eight week old fruit. The fruit width was more in GTC-3 (14.27 cm) of six week old fruit and in Swarna (15.67 cm) of eight week old fruit. The fruit weight was found to be higher in both six and eight week old fruits of GTC-3 (1.93 kg and 2.32 kg respectively). The rind thickness was found lesser in both six and eight week fruits of NSP-2 (0.33 cm and 0.37 cm respectively) and core thickness was lesser in both six and eight week fruits of KV-1 (3.75 cm and 3.77 cm, respectively). Mesocarp thickness of six week fruit was found maximum in GTC-3 (3.67 cm) and in eight week old fruit; Lalbagh Madhura recorded the higher (3.77 kg) mesocarp thickness. The lesser rind weight was found in KV-2 of both six and eight week fruits (0.13 kg and 0.17 kg respectively) and the four genotypes of eight week fruit recorded the minimum core weight viz., KV-1, KV-2, KV-3 and HV-2 each of 0.11 kg. The mesocarp thickness in both six and eight week old fruit of GTC-3 found the highest (1.29 and 1.63 kg respectively) and the higher edible portion recovery of both six and eight old week fruits was recorded in KV-2 (71.33% and 75.07%).

**Keywords:** Tender jackfruit, Vegetable jackfruit, Vegan meat, Morphological characterization

JACKFRUIT (*Artocarpus heterophyllus* Lam.) is one of the important fruit crops of tropical region, belonging to moraceae family. It is believed to have originated from the forests of Western Ghats of India and later the cultivation has been distributed throughout the tropical lowlands of northern and southern hemisphere around the globe. In India, it is widely distributed in the states of Assam, Bihar, Tripura, West Bengal, Uttar Pradesh, Kerala, Tamil Nadu and Karnataka (Rai *et al.*, 2003). Presently in India, jackfruit is cultivated in an area of 1.87 Lakh ha with the production of 1.87 Million MT (Anonymous 2022). It is largest tree borne fruit on

earth and is a heavy yielder than any other fruit trees. (Shyamamma *et al.*, 2017 and Bharathi *et al.*, 2022).

The jackfruit is utilized at different stages for various purposes viz., at tender stage, it can be used as vegetable in many culinary dishes and can be processed into pickles, chutney, powders, canned slices and this is the best meat alternative. In mature stage, it is used for making papad, chips and various culinary dishes. The ripe fruits are often relished fresh and also processed into jam, jelly, juice, squash, fruit powder and pulp is used as natural ingredient in ice-cream *etc.* Due to increasing adaptability of vegan lifestyle, there is huge demand for dummy meat or



meat substitute products. Jackfruit at tender stage will be having meat like texture because of its fiber. It is often deliberated as super food and is in high demand because of its immense nutritive value. The nutritional composition per 100 g of tender fruit include 84 per centage of moisture, 2.6 g of protein, 0.3 g of fat, 0.7 g of minerals, 2.8 g of crude fiber, 9.4 g of carbohydrates, 51 Kcal. of energy, 287-323 mg of potassium, 40 mg of phosphorous, 30 mg of calcium and 0.567 mg of iron (Gopalan *et al.*, 1989). These phytochemicals possess anti-inflammatory, anti-bacterial, anti-tumor and other nutraceutical properties with low glycemic index and almost zero cholesterol compared to its ripe stage (Swami *et al.*, 2012).

The variations witnessed in the qualitative (fruit shape) and quantitative (fruit length, width and weight of the fruit; thickness and weight of the rind, mesocarp and core; recovery of edible portion) traits of the tender jackfruit genotypes can be attributed to their virtue of cross pollination and seed propagation (Wangchu *et al.*, 2013 and Sampath *et al.*, 2019). The screening of the jackfruit genotypes for the vegetable purpose will help in selection of elite trees possessing high yield potential with better nutritional and cooking quality, this will further lead to the development of good varieties resulting in commercialization of the tender jackfruit as a vegetable.

## MATERIAL AND METHODS

The present study was conducted during the off season (October to December, 2020) and main fruit bearing season of 2020-21 (March to June, 2021) based on the survey at different jackfruit growing areas of Karnataka. Among the surveyed trees, the genotypes established at different locations in GKVK campus and Kachahalli village of Doddaballapura taluk of Bengaluru Rural district were finalized for the study based on bearing conditions and fruit availability.

### Morphological Characters

The morphological characters were recorded as per the guidelines of Bioversity International, Rome

(formerly, IPGRI) jackfruit descriptors (IPGRI, 2000). The morphological characters were grouped into qualitative and quantitative characters.

### Qualitative Characters

The characters whose data is non quantifiable (fruit shape) was grouped as qualitative characters. Such parameters were documented and analyzed based on the score given for each character in jackfruit descriptor.

### Quantitative Characters

The quantitative characters such as fruit length, fruit width, fruit weight; rind, core and mesocarp thickness; rind, core and mesocarp weight along with recovery percentage of the 20 genotypes (Table 1) were recorded at six and eight weeks after fruit set (opening of female inflorescence). The genotypes HV-1 and HV-2 were kept for reference as standard checks because of their use as vegetable types (Shyamamma *et al.*, 2008).

### Statistical Analysis

The data for the above said quantitative characters were recorded at six and eight weeks after fruit set. Three replications in each genotype (Three fruits per tree) were used for recording all quantitative traits. The data was computed using one way ANOVA (Kavya *et al.*, 2019) and analysis was done in Microsoft Excel separately for the fruits of both six and eight weeks after fruit set.

## RESULTS AND DISCUSSION

### Fruit Shape

Fruit shape is one of the major traits, which determines the tender jackfruit recovery per cent. Thus a good fruit shape will determine better recovery. The fruits with irregular shape and bumpy surfaces contribute to more wastage while cutting the tender jackfruit. Good diversity was recorded among the genotypes studied where, spheroid shape was noticed in two genotypes Swarna and GTC-3; oblong shape was seen in three genotypes *i.e.*, Lalbagh Madhura,

TABLE 1  
List of jackfruit genotypes screened for vegetable types

Genotype/ Variety	Abbreviation Used	Location
Lalbagh Madhura	LM	Jackfruit orchard -2 Department of Horticulture, GKVK
Byrachandra	BC	Hithakari Nursery, Shivakote, Bengaluru
Swarna	SW	Jackfruit orchard -1 Department of Horticulture, GKVK
Muttamvarika	MV	Jackfruit orchard -1 Department of Horticulture, GKVK
Gumless	GL	Jackfruit orchard -1 Department of Horticulture, GKVK
Horticulture Vegetable type -1	HV-1	Dept. of Horticulture, GKVK
Horticulture Vegetable type -2	HV-2	Dept. of Horticulture, GKVK
Kachahalli Vegetable type -1	KV-1	Kachahalli village, Doddaballapura Tq., Bengaluru Rural dist.
Kachahalli Vegetable type -2	KV-2	Kachahalli village, Doddaballapura Tq., Bengaluru Rural dist.
Kachahalli Vegetable type -3	KV-3	Kachahalli village, Doddaballapura Tq., Bengaluru Rural dist.
GKVK Tissue Culture lab-1	GTC-1	Dept. of Horticulture, GKVK
GKVK Tissue Culture lab-3	GTC-3	Dept. of Horticulture, GKVK
GKVK Avenue-18	GA-18	GKVK campus
GKVK Avenue-20	GA-20	GKVK campus
National Seed Project-1	NSP-1	NSP, GKVK
National Seed Project-2	NSP-2	NSP, GKVK
National Seed Project-3	NSP-3	NSP, GKVK
GKVK Horticulture orchard-9	GH-9	Jackfruit orchard -1 Department of Horticulture, GKVK
GKVK Horticulture orchard-11	GH-11	Jackfruit orchard -1 Department of Horticulture, GKVK
GKVK Horticulture orchard-15	GH-15	Jackfruit orchard -1 Department of Horticulture, GKVK

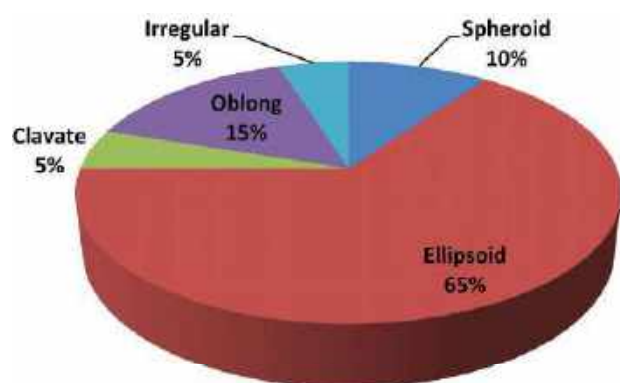


Fig. 1 : Diversity in fruit shape noticed among the screened tender jackfruit genotypes

Byrachandra and GH-15. NSP-1 exhibited clavate shape and HV-1 with irregular shape; remaining 13 genotypes were ellipsoid fruit shape (Fig. 1).

Similar type of diversity in fruit shape among different jackfruit genotypes was also reported by Rai

*et al.* (2003), Dey and Baruah (2019), Akter and Rahman (2017). The variations in the fruit shape are mostly attributed to genotypic character.

### Fruit Rind Colour

Rind or outer surface colour is the primary thing that catches attention in any fruit. Generally, dark green shades of tender fruit are preferred over its lighter ones. Majority of the genotypes exhibited green colour, where as three genotypes *i.e.*, GA-18, GA-20 and NSP-1 tender fruits were greenish yellow colour. The genotypes NSP-4, NSP-3 and HV-2 had brownish yellow, brown and pale green colours respectively (Fig. 2).

Variation in fruit skin colour is because of accumulation of one or more combination of colour pigments such as chlorophyll, anthocyanins and

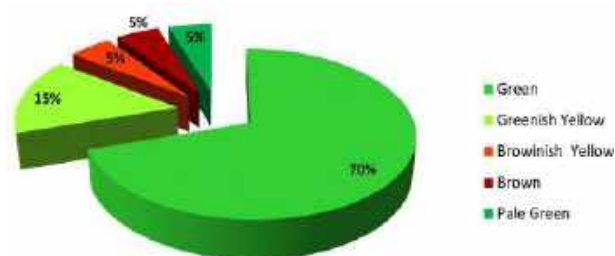


Fig. 2 : Diversity in fruit rind colour noticed among the screened tender jackfruit genotypes

carotenoids in the fruit rind (Akter and Rahman, 2017). They also reported good range of variability in fruit skin colour of jackfruit genotypes.

### Quantitative Characters

#### Fruit Length

Fruit length, which is convenient to hold in a single hand is preferred in tender jackfruit. The fruit length among the studied genotypes differed significantly ranging from 23.77 cm to 33.67 cm with a mean of 29.48 cm in six week's fruits and a range of 28.07 cm to 37.67 cm with a mean fruit length of 33.62 cm was recorded in eight week's fruit (Table 2). NSP-3 had the higher fruit length (33.67 cm) and two genotypes *viz.*, Swarna (31.9 cm) and NSP-2 (31.47 cm) were found on par with in the six week old fruits. In the eight week old fruits, the genotype HV-1 recorded the higher fruit length (37.67 cm) and the tree genotypes

*viz.*, Swarna (36.80 cm), GTC-3 (36.77 cm) and Gumless (36.07 cm) were found on par with HV-1. The least was noticed in both 6 week and eight week old fruit of KV-2 (23.77 cm and 28.07 cm respectively).

The variations in fruit length could be due to genetic and environmental factors. Rai *et al.* (2003), Chandrashekar *et al.* (2018), Dey and Baruah (2019) and Kavya *et al.* (2019) have also noticed good diversity in fruit length of jackfruit genotypes in their study.

#### Fruit Width

The width of the tender jackfruit visually determines the suitability of fruit for vegetable type. A significant difference in the fruit width was noticed among the genotypes ranging from 7.5 cm to 14.27 cm with the mean of 10.72 cm in the six week old fruits. In the eight week old fruits, the range was 9.49 cm to 15.67 cm was noticed with a mean fruit width of 12.29 cm (Table 2). The genotype GTC-3 recorded the highest fruit width (14.27 cm) followed by Swarna (13.10 cm) in six week old fruits. In the eight week old fruits, Swarna recorded higher fruit width (15.67 cm) followed by GTC-3 (14.60 cm). The least fruit width was recorded from KV-2 (7.50 cm) in the six week old fruit and in the eight week old fruit, KV-1 exhibited the least fruit width (9.49 cm).

TABLE 2  
Variations noticed in fruit length, width and weight of jackfruit genotypes at six and eight week old tender fruits

Genotypes	Fruit length (cm)		Fruit width (cm)		Fruit weight (kg)	
	6 week	8 week	6 week	8 week	6 week	8 week
Lalbagh Madhura	31.23	33.17	11.57	12.80	1.21	1.67
Byrachandra	30.47	33.17	11.93	13.62	1.40	2.19
Swarna	31.90	36.80	13.10	15.67	1.61	1.87
Muttamvarika	30.67	32.20	11.50	13.23	1.41	2.23
Gumless	29.37	36.07	10.20	12.67	1.34	1.78
HV-1	30.83	37.67	11.00	14.07	1.20	2.14
HV-2	29.33	33.90	10.27	11.93	1.05	1.84
KV-1	25.27	30.81	10.53	9.49	1.03	1.30

Genotypes	Fruit length (cm)		Fruit width (cm)		Fruit weight (kg)	
	6 week	8 week	6 week	8 week	6 week	8 week
KV-2	23.77	28.07	7.50	9.93	0.54	0.86
KV-3	27.00	32.50	9.67	10.87	0.87	1.40
GTC-1	26.27	30.60	10.23	11.93	1.02	1.48
GTC-3	31.50	36.77	14.27	14.60	1.90	2.32
GA-18	30.33	34.37	10.93	12.30	1.20	1.56
GA-20	30.63	34.40	11.23	12.87	1.13	1.42
NSP-1	30.90	33.70	11.00	12.20	1.29	1.56
NSP-2	31.47	33.17	10.23	12.53	1.39	1.59
NSP-3	33.67	36.67	11.33	12.97	1.16	1.53
GH-9	29.23	31.13	9.47	10.63	0.90	1.20
GH-11	29.27	33.33	9.83	10.67	1.11	1.55
GH-15	26.53	33.87	8.57	10.90	0.75	1.44
Grand Mean	29.48	33.62	10.72	12.29	1.17	1.65
Range Maximum	33.67	37.67	14.27	15.67	1.90	2.32
Minimum	23.77	27.07	7.5	9.49	0.54	0.86
SEm ±	0.82	0.82	0.33	0.31	0.06	0.06
CD at 5%	2.35	2.34	0.95	0.89	0.16	0.18
F test	*	*	*	*	*	*

Dey and Baruah (2019) reported that, the variations in the fruit width can be attributed to genetic factors and soil nutrition. Diversity in fruit width was noticed from the studies of Rai *et al.* (2003), Dey and Baruah (2019), Kavya *et al.* (2019), Akter and Rahman, (2017).

### Fruit Weight

It is one of the important characters in selecting the fruit for vegetable purpose. The fruit weight differed significantly in the genotypes ranging from 0.54 kg to 1.90 kg with the mean of 1.17 kg in the six week old fruit and the range of 0.86 kg to 2.32 kg with a mean fruit weight of 1.65 kg was noticed in eight week old fruits (Table 2). Higher fruit weight in both six and eight week old fruit was noticed in GTC-3 (1.90 kg and 2.32 kg respectively), followed by Swarna (1.61 kg) in six week's fruit and Muttamvarika (1.23 kg) in eight week old fruit.

The fruit weight has inverse relation with total bearing habit in a tree (Rai *et al.*, 2003). More the fruit number in the trees, lesser will be the individual fruit weight and vice-versa. Similarly, variation in fruit weight has been reported by Anu *et al.* (2015), Chandrashekar *et al.* (2018) and Phaomei & Mathew (2019).

### Rind Thickness

Rind thickness differed significantly among the genotypes ranging from 0.33 cm to 1.57 cm with a mean of 0.63 cm in six week old fruits. In eight week old fruits, the range was from 0.37 cm to 1.5 cm (Table 3). Lesser rind thickness was noticed in both six and eight week old fruits in genotype NSP-2 (0.33 cm and 0.37 cm respectively) followed by KV-2 (0.37 cm and 0.40 cm, respectively) and HV-2 (0.40 cm and 0.47 cm, respectively). The higher rind thickness was recorded in both six and eight old fruits of GTC-3 (1.57 and 1.5 cm, respectively). Thinner, less tough and less weighed rind type of genotype is more preferred for vegetable type.

TABLE 3  
Variations noticed in fruit rind, core and mesocarp thickness of jackfruit genotypes at six and eight week old tender fruits

Genotypes	Rind thickness (cm)		Core thickness (cm)		Mesocarp thickness (cm)	
	6 week	8 week	6 week	8 week	6 week	8 week
Lalbagh Madhura	0.63	0.67	5.07	6.07	3.60	3.77
Byrachandra	0.57	0.63	4.80	5.80	2.97	3.43
Swarna	0.42	0.63	6.13	7.10	2.57	3.70
Muttamvarika	0.93	0.90	6.17	8.10	1.97	2.37
Gumless	0.90	0.97	5.23	5.67	2.23	2.50
HV-1	0.53	0.87	5.93	7.13	1.73	2.83
HV-2	0.40	0.43	4.40	4.70	2.53	3.23
KV-1	0.57	0.50	3.75	3.77	2.57	2.29
KV-2	0.37	0.40	4.40	5.13	1.43	2.23
KV-3	0.47	0.53	4.17	4.93	2.10	2.33
GTC-1	0.77	0.70	5.00	5.13	2.03	2.57
GTC-3	1.57	1.50	4.77	4.73	3.67	3.30
GA-18	0.73	0.80	5.23	5.83	2.20	2.63
GA-20	0.60	0.60	4.50	5.57	2.57	2.97
NSP-1	0.47	0.47	5.53	5.93	2.07	3.00
NSP-2	0.33	0.37	5.13	5.53	1.97	2.17
NSP-3	0.73	0.70	4.97	5.60	2.07	2.37
GH-9	0.60	0.67	5.00	5.90	1.53	1.67
GH-11	0.50	0.47	4.50	5.13	1.93	2.37
GH-15	0.57	0.53	4.33	4.87	1.57	2.10
Grand Mean	0.63	0.67	4.95	5.63	2.27	2.69
Range Maximum	1.57	1.5	6.17	8.1	3.67	3.77
Minimum	0.33	0.37	3.77	3.77	1.43	1.67
SEm ±	0.06	0.04	0.15	0.23	0.09	0.13
CD at 5%	0.16	0.12	0.44	0.66	0.26	0.36
F test	*	*	*	*	*	*

Maximum rind thickness of 2 cm and minimum of 0.5 cm in the genotypes of cluster 1 and 3 respectively was noticed in a study conducted by Anu *et al.* (2015). Mahalakshmi (2017) reported the average rind thickness of 0.86 cm and 1.13 cm in the 30 and 60 days old tender jackfruits respectively.

#### Core Width

The core width plays a major role in per cent recovery of edible portion. In the present study,

a significant difference was noticed ranging from 3.77 cm to 6.17 cm with a mean of 4.95 cm in the six week old fruits. In eight week old fruits, the range was from 3.77 cm to 8.1 cm with a mean core thickness of 5.63 cm (Table 3). Both six and eight week old fruits of KV-1 recorded minimum core width (3.75 cm and 3.77 cm respectively) followed by KV-3 (4.17 cm) and KV-2 (4.40) in six week old fruit; HV-2 (4.70 cm) and GTC-3 (4.73 cm) in eight week's fruits, respectively.

Core is having insoluble fiber, which is indigestible in human body and hence considered as non edible. Genotypes with lesser core thickness and weight are extremely helpful in identifying a vegetable type. Wangchu *et al.* (2013) from West Bengal and Kavya *et al.* (2019) from Karnataka also reported diversity in core width among the genotypes studied.

### Mesocarp Thickness

It is another important visual character that determines the suitability of the fruit for vegetable type. A significant difference in the mesocarp thickness among the genotypes was noticed with a range of 1.43 cm to 3.67 cm and the mean of 2.27 cm in the six week old fruits. In eight week old fruits, the range was from 1.67 cm to 3.77 cm with the mean of 2.69 cm (Table 3). The Highest mesocarp thickness in the six week old fruits was noticed in GTC-3 (3.67 cm) and the genotype Lalbagh Madhura (3.60 cm) was found on par with it. The least was seen in KV-2 (1.43). In the eight weeks fruit, Lalbagh Madhura was recorded highest mesocarp thickness (3.77 cm) and the genotypes Swarna (3.70 cm) and Byrachandra (3.43 cm) were found on par with it. The least was noticed in GH-9 (1.67 cm). For screening a

genotype for vegetable type, more mesocarp thickness and weight should be taken in to consideration.

The variations in the mesocarp thickness has been reported by Wangchu *et al.* (2013). They attributed that, the variation in mesocarp thickness could be due to cross pollination nature of the crop and also pertaining to genotypic characters.

### Rind Weight

One of the important factors that contribute to the total weight of fruit and also the edible portion recovery hence the genotypes with least rind recovery should be considered to prefer for vegetable types. The rind weight exhibited a significant difference among the genotypes ranging from 0.13 kg to 0.42 kg with a mean rind weight of 0.25 kg in six week old fruits and in eight week old fruits, the range was from 0.17 kg to 0.48 kg with a mean of 0.31 kg (Table 4). The least rind weight was recorded in KV-2 and GH-15 (0.13 kg) and seven genotypes found on par with them in the six week old fruits. The KV-2 also recorded least rind weight in eight week old fruits (0.17 kg) and four genotypes were on par with it. The higher rind weight was recorded in NSP-3 (0.42 kg) of six week old fruit and GTC-3 (0.48 kg) of eight week fruit.

TABLE 4  
Variations noticed in fruit rind, core, mesocarp weight and recovery of jackfruit genotypes at six and eight week old tender fruits

Genotypes	Rind weight (kg)		Core weight (kg)		Mesocarp weight (kg)		Recovery (%)	
	6 week	8 week	6 week	8 week	6 week	8 week	6 week	8 week
Lalbagh Madhura	0.25	0.31	0.25	0.29	0.69	1.06	57.64	63.27
Byrachandra	0.25	0.26	0.24	0.30	0.92	1.62	65.48	73.80
Swarna	0.28	0.29	0.25	0.23	1.05	1.34	65.52	71.74
Muttamvarika	0.31	0.42	0.26	0.38	0.83	1.36	58.84	61.09
Gumless	0.32	0.38	0.17	0.21	0.84	1.16	62.80	65.05
HV-1	0.20	0.37	0.21	0.39	0.79	1.37	66.27	63.93
HV-2	0.20	0.28	0.11	0.17	0.72	1.37	69.04	74.58
KV-1	0.18	0.24	0.11	0.19	0.73	0.97	71.33	75.07
KV-2	0.13	0.17	0.11	0.17	0.30	0.54	55.55	63.24
KV-3	0.20	0.24	0.11	0.20	0.52	0.90	59.64	63.88
GTC-1	0.25	0.40	0.15	0.21	0.61	0.92	60.09	62.06

Genotypes	Rind weight (kg)		Core weight (kg)		Mesocarp weight (kg)		Recovery (%)	
	6 week	8 week	6 week	8 week	6 week	8 week	6 week	8 week
GTC-3	0.41	0.48	0.13	0.15	1.29	1.63	67.93	70.63
GA-18	0.19	0.26	0.21	0.27	0.77	1.03	64.34	65.87
GA-20	0.24	0.23	0.12	0.14	0.76	1.04	67.48	73.24
NSP-1	0.21	0.23	0.18	0.19	0.88	1.14	68.01	73.04
NSP-2	0.35	0.34	0.21	0.24	0.84	1.02	60.25	64.31
NSP-3	0.42	0.47	0.14	0.16	0.52	0.73	44.78	48.17
GH-9	0.18	0.19	0.15	0.22	0.55	0.78	61.31	64.59
GH-11	0.25	0.35	0.14	0.16	0.69	0.98	61.56	62.87
GH-15	0.13	0.22	0.12	0.17	0.48	1.01	63.04	70.15
Grand Mean	0.25	0.31	0.17	0.22	0.74	1.10	62.55	66.53
Range Max.	0.42	0.48	0.26	0.39	1.29	1.63	71.33	75.07
Min.	0.13	0.17	0.11	0.14	0.30	0.54	44.78	48.17
SEm ±	0.03	0.02	0.02	0.01	0.04	0.05	1.07	1.13
CD at 5%	0.09	0.06	0.05	0.03	0.11	0.15	3.05	3.22
F test	*	*	*	*	*	*	*	*

Rind weight should be less and higher and more rind weight leads to increase in total fruit weight and the recovery of edible portion will be less. Variations in rind weight from 2.70 kg to 6.41 kg were reported from Kavva *et al.* (2019) and the rind weight of 1.15 kg to 4.7 kg was reported from Akter and Rahman, (2017) in their study.

### Core Weight

The core weight plays a prominent role in determining the suitability of a genotype for vegetable purpose. Most of the genotypes with higher core weight would be unsuitable for vegetable types and hence less core weight is preferred. In the present study, a significant difference was recorded for core weight ranging from 0.11 kg to 0.26 kg and a mean of 0.17 kg in six week old fruits and the range of 0.14 kg to 0.39 kg with a mean core weight of 0.22 kg was recorded in eight week old fruits (Table 4). The genotypes KV-1, KV-2, KV-3 and HV-2 had the least core weight of 0.11 kg. The highest was seen in Muttamvarika (0.26 kg), Lalbagh Madhura and Swarna (0.25 kg) in the six week fruits. The genotype GA-20 had the least core weight (0.14 kg) followed by GTC-3 (0.15 kg)

and the highest was noticed in HV-1 (0.39 kg) followed by Muttamvarika (0.38 kg).

Azad *et al.* (2007) noticed a poor correlation between core percentage of fruits with the environmental factors. Akter and Rahman (2017) reported the core weight ranged from 167 g to 935 g with a mean core weight of 371.22 g.

### Mesocarp Weight

Mesocarp is the most important trait in determining the suitability of a genotype for vegetable purpose. The higher mesocarp weight differed significantly, ranging from 0.30 kg to 1.29 kg with a mean mesocarp weight of 0.74 kg in six week fruits and in eight weeks old fruits, the range was from 0.54 kg to 1.63 kg with a mean fruit weight of 1.10 kg (Table 4). The genotype GTC-3 recorded the highest mesocarp content in both six and eight weeks old fruits (1.29 kg and 1.63 kg, respectively) followed by Swarna (1.05 kg) and Byrachandra (0.92 kg) in six week old fruits and Byrachandra (1.62 kg) of eight week fruit. The least mesocarp weight was noticed in KV-2 at six and eight weeks old fruits (0.30 kg and 0.54 kg, respectively).

This is the edible portion in the tender jackfruit after separating the core and rind. Thus the mesocarp weight should be higher than core and rind weight in order to get good recovery. Presence or absence of seeds in the mesocarp is also taken into consideration. Mesocarp without seed is preferred but slightly immature or presence of soft seeds is also acceptable.

### Recovery

Recovery of edible portion is calculated based on mesocarp weight over total fruit weight. A significant difference was observed in edible portion recovery ranging from 44.78 per cent to 71.33 per cent with a mean of 62.55 per cent in six week old fruit and the range of 48.17 per cent to 75.07 per cent with a mean of 66.53 per cent in eight week old fruits (Table 4). The genotype KV-1 recorded higher recovery in both six and eight week old fruits (71.33% and 75.07% respectively) and the edible portion recovery of four genotypes found on par *viz.*, HV-2 (69.04%), Swarna (65.52%), Byrachandra (65.48%) in the six old fruits. In eight week old fruits, four genotypes *i.e.*, HV-2 (74.58%), Byrachandra (73.80%), GA-20 (73.24%) and NSP-1 (73.04%) were found on par with KV-1. The least edible portion was recorded in NSP-3 genotype of six and eight week old fruits (44.78% and 48.17%, respectively).

The recovery of the edible portion is directly proportional to higher mesocarp weight in the total fruit weight. Genotype with good fruit weight might not get good recovery because of contribution of higher rind and core weight. Akter and Rahman, (2020) also reported a huge diversity of 18.39 per cent to 60.74 per cent recovery of edible portion among the jackfruit germplasm of their study.

Based on the morphological characters assessed in the experiment, the parameters such as minimum rind weight and thickness, less core weight and thickness, higher mesocarp weight and thickness at the early growth stages (six to eight weeks) plays the key role in identifying the genotypes/varieties that are suitable for vegetable types. However, the gum/latex exudation was seen throughout the observed stages, it prevails up to the fruit maturity or onset of fruit ripening in Gumless also and its content may be

reduced in some genotypes due to biochemical conversions. Among different genotypes evaluated, the genotype KV-1 performed the best. The check variety HV-2 also performed well and stood in the second position. Further, KV-1 and HV-1 also recorded higher mesocarp recovery because of less rind weight, core weight and also less core and rind thickness even though they had less total fruit weight. The genotypes NSP-3 and KV-2 got the least recovery because of the more rind weight which added to the total fruit weight and hence reduced in edible portion. The genotypes with higher edible portion recovery could be studied further for its nutritional properties and cooking quality to recommend for vegetable jackfruit type.

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## Effect of Long-term Application of Graded Levels of Boron on Growth, Yield and Oil Content of Sunflower (*Helianthus annuus* L.)

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### ABSTRACT

A field experiment was conducted to study the levels of boron application on growth, yield and oil content of sunflower (*Var- KBSH-78*) during *kharif-2020* at Zonal Agricultural Research Station, Gandhi Krishi Vignana Kendra, Bengaluru. The experiment was laid out in a RCBD with eight treatments, replicated thrice. Treatments consisting of the application of recommended dose of fertilizer (POP-90:90:62.5 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O ha<sup>-1</sup> and 7.5 t ha<sup>-1</sup> FYM), POP along with soil application of graded levels of boron. The results revealed that due to the application of higher dose of boron, germination of sunflower was completely affected from 2 kg B ha<sup>-1</sup> to 16 kg B ha<sup>-1</sup> applied plots even at 13<sup>th</sup> DAS. But, after gap filling with the seedlings grown in polythene pockets, crop stand was good however, crop growth and yield was affected as the boron level increased. The sunflower yield data indicated that continuous application of boron from 2 kg ha<sup>-1</sup> to 16 kg ha<sup>-1</sup> has drastically reduced the seed yield as compared to no boron applied plots. Stalk yield and other growth and yield attributes of sunflower also followed the same trend. Significantly higher seed yield (20.47 q ha<sup>-1</sup>) and stalk (30.88 q ha<sup>-1</sup>) in FYM+ RDF to 16 kg B ha<sup>-1</sup> (13.28 q ha<sup>-1</sup>) has considerably lowered the seed yield of sunflower.

*Keywords:* Sunflower, Long term application of boron, Growth, Yield

SUNFLOWER (*Helianthus annuus* L.), one of the major oilseed crops, is widely cultivated in the world. This crop has gained importance because of its short duration of maturity, excellent quality oil, photo insensitivity, wide adaptability in different agro-climatic regions, kind of cropping patterns and drought tolerance. Sunflower production is about 50.22 million tons from 27.87 million ha of global coverage India accounts for about 15-20 per cent of global oilseeds area (32 lakh ha) with seed production of 21.3 lakh tons (FAO. 2020). Karnataka, Maharashtra and Odisha are the major states, which accounts for about 75 per cent of the total area under sunflower cultivation in India. In Karnataka it is grown in an area of 1.29 lakh ha with production of 1.03 lakh tons and productivity of 802 kg ha<sup>-1</sup>, which

is lower than national (931 kg ha<sup>-1</sup>) and much lower than world's (2048 kg ha<sup>-1</sup>) average productivity (APEDA. 2020).

Sunflower requires a high amount of B as compared to other crops and has been used as a good indicator of B deficiency. It is one of the most sensitive crop to low boron (B) supply, showing B-deficiency symptoms on leaves, stems and reproductive organs. Yield reduction due to B deficiency is frequent, even if typical visual symptoms on leaves and heads are not evident. Furthermore, an increase in the incidence of B deficiency can be expected with continued cropping and increased yields because very little B is supplemented as fertilizer.

For many plant species there is a very narrow range in critical tissue concentrations of boron between

deficiency and toxicity. Boron toxicity exerts different effects on vascular plants, such as reduced root cell division, lower photosynthetic rates and decreased lignin and suberin levels. Accordingly, a reduced growth of shoots and roots is typical of plants exposed to higher levels of boron. Boron toxicity in the sunflower is indicated first by a mottling of the tips and edges of the lowest leaves. Later these mottled areas die. In severe cases of injury practically the entire leaf may be affected. However, the exact tissue concentration or soil boron level showing the boron toxicity symptoms is rarely known specially in sunflower crop. Hence, a study was taken on boron dynamics with the objective to know the effect of long term application of different levels of boron on growth, yield and oil content of sunflower.

#### MATERIAL AND METHODS

A long term field experiment has been in progress since 2016 on *Afisols* belonging to *Vijayapura* soil series at 'F' block, STCR field unit, Zonal Agriculture Research Station, University of Agricultural Sciences, Gandhi Krishi Vignyan Kendra, Bengaluru located in Eastern Dry Zone of Karnataka at 13° 04' 55.2" N latitude, 77° 34' 10.0" E longitude with an altitude of 930 meters above mean sea level (MSL). The experiment consists of eight treatments with graded levels of boron application and replicated thrice. The different crops raised during different years are sunflower-maize-sunflower-ragi. In the present study, sunflower as a test crop was taken up during *kharif* 2020.

A germination study was conducted prior to the long-term study at various levels of B and found germination at 40 mg kg<sup>-1</sup> B also. From the results obtained, a pot experiment was conducted till 45 DAS from 1, 2, 4, 8, 10, 20 kg B ha<sup>-1</sup> upto 80 kg B ha<sup>-1</sup>. In that particular pot study also there were no toxicity symptoms till 20 kg B ha<sup>-1</sup> application. Hence the experiment was designed with regard to the results since 2016 and the treatment details are as follows below.

Treatment details :

- T<sub>1</sub> : Absolute control
- T<sub>2</sub> : POP + without boron
- T<sub>3</sub> : POP + 2 kg B ha<sup>-1</sup>
- T<sub>4</sub> : POP + 4 kg B ha<sup>-1</sup>
- T<sub>5</sub> : POP + 6 kg B ha<sup>-1</sup>
- T<sub>6</sub> : POP + 8 kg B ha<sup>-1</sup>
- T<sub>7</sub> : POP + 12 kg B ha<sup>-1</sup>
- T<sub>8</sub> : POP + 16 kg B ha<sup>-1</sup>

Note: POP - UAS-B Package of practice for sunflower is 90:90:62.5 (N, P<sub>2</sub>O<sub>5</sub>, K<sub>2</sub>O kg ha<sup>-1</sup>) + FYM 7.5 t ha<sup>-1</sup>

A composite soil sample was taken from 0-15 cm depth from each plot after laying out the field plan before start of experiment during 2016. The soil was air dried, pounded and then passed through 2 mm sieve and was analyzed for physical and chemical properties following standard procedure. The analytical techniques followed for the estimation of physical and chemical properties of soil and the results are presented in Table 1.

The germination of sunflower seeds was observed till 13 DAS and the failure in germination was compensated with gap filling with the seedlings grown in pockets, crop stand was found good after that.

The sunflower was raised as per the treatment details. Borax was used as boron source. The biometric observations like growth (at 30 DAS, 60 DAS and at harvest) and yield parameters of sunflower were recorded. The oil content in seeds was assessed using nuclear magnetic resonance spectrophotometer (NMR, model Minispec 20 pi).

#### RESULTS AND DISCUSSION

##### Boron Concentration in Soil with Long Term Boron Fertilization

With the continuous application of graded levels of B from 2 to 16 kg ha<sup>-1</sup> resulted in an increase in available boron content in soil over a period of five years in all treatments except control and POP treatments where boron was not applied. Both before and after the sunflower crop *kharif* 2020, the available

TABLE 1  
Initial physico-chemical properties of the experimental site (2016)

Particulars	Values	Methodology
Physical properties of soil		
Sand (%)	65.60	International pipette method(Piper, 1966)
Silt (%)	16.60	
Clay (%)	17.80	
Texture	Sandy loam	
Bulk density (Mg m <sup>-3</sup> )	1.52	Keen Raetzowski cup method (Piper,1966)
Chemical properties of soil		
pH (1:2.5)	5.45	Potentiometry (Jackson, 1973)
Electrical conductivity (dS m <sup>-1</sup> )	0.04	Conductometry (Jackson, 1973)
Organic carbon (g kg <sup>-1</sup> )	4.0	Wet oxidation method (Walkley and Black, 1934)
Available N (kg ha <sup>-1</sup> )	231.37	Alkaline permanganate method(Subbiah and Asija, 1956)
Available P <sub>2</sub> O <sub>5</sub> (kg ha <sup>-1</sup> )	176.84	Bray's method (Jackson, 1973)
Available K <sub>2</sub> O (kg ha <sup>-1</sup> )	250.26	Flame photometry method(Page <i>et al.</i> , 1982)
Available S (mg kg <sup>-1</sup> )	27.30	Turbidometry method (Jackson, 1973)
Exchangeable calcium [c mol (p <sup>+</sup> ) kg <sup>-1</sup> ]	0.60	Versenate titration method(Jackson, 1973)
Exchangeable magnesium [c mol (p <sup>+</sup> ) kg <sup>-1</sup> ]	1.85	
DTPA iron (mg kg <sup>-1</sup> )	8.66	DTPA extraction method(Lindsay and Norvell, 1978)
DTPA manganese (mg kg <sup>-1</sup> )	3.91	
DTPA copper (mg kg <sup>-1</sup> )	1.36	
DTPA zinc (mg kg <sup>-1</sup> )	1.03	
Hot watersoluble boron(mg kg <sup>-1</sup> )	0.64	

soil boron was found to be statistically significant. In the initial stage (*kharif* 2020) of the experiment soil boron increase was from 3.05 to 5.51 mg kg<sup>-1</sup> with application of B from 2 to 16 kg ha<sup>-1</sup>, respectively.

The available boron content after the harvest was also increased due to the continuous application of boron to a higher concentration (3.46 to 5.57 mg kg<sup>-1</sup>) from 2 to 16 kg ha<sup>-1</sup>, respectively. Higher doses of B at 12 and 16 g ha<sup>-1</sup> showed on par results at the end of this experiment. The application of higher levels of boron to previous crop increased the hot water soluble B in the soils at harvest of residual crop. This might be due to slow release of B in to soil pool (Priyanka 2018). Similarly, in a study conducted by Bhattacharya *et al.* (2015) soil B was increased from 33.33 to 70.37 per cent after sunflower harvest with graded levels of B application.

TABLE 2  
Effect of long-term application of graded levels of boron on available B of soil initial (*kharif* 2020) and after harvest of sunflower

Treatment details	Initial ( <i>kharif</i> 2020) Soil available B (mg kg <sup>-1</sup> )	After harvest Soil available B (mg kg <sup>-1</sup> )
T <sub>1</sub> Control	0.46	0.54
T <sub>2</sub> POP without Boron	0.58	0.56
T <sub>3</sub> POP+ 2 kg B ha <sup>-1</sup>	3.05	3.46
T <sub>4</sub> POP+ 4 kg B ha <sup>-1</sup>	3.56	3.69
T <sub>5</sub> POP+ 6 kg B ha <sup>-1</sup>	3.56	3.88
T <sub>6</sub> POP+ 8 kg B ha <sup>-1</sup>	3.81	4.45
T <sub>7</sub> POP+ 12 kg B ha <sup>-1</sup>	4.07	5.54
T <sub>8</sub> POP+ 16 kg B ha <sup>-1</sup>	5.51	5.70
S Em ±	0.39	0.35
C D @ 5%	1.19	1.06

### Status of Post-harvest Soil Available N, P and K

As shown in Table 3, the varying quantities of boron application had no discernible effect on the post-harvest soil available N, P and K status.

After the sunflower crop was harvested, the available nitrogen content of the soil did not significantly differ across different boron treatments. However, in the treatments the available N increased compared to initial nitrogen level of soil. The soil available N content (Table 3) was numerically increased compared to control (252.97 kg ha<sup>-1</sup>) and as the boron level increased from 2 kg (257.15) to with 16 kg B ha<sup>-1</sup> (270.92 kg ha<sup>-1</sup>). The available phosphorus was quite high in all of the treatments but, compared to initial phosphorous, it was increased. After the sunflower crop was harvested, there was no significant variation in the available phosphorus level of the soil between the various treatments. But, the available P<sub>2</sub>O<sub>5</sub> level increased numerically as the boron level increased. The 16 kg B ha<sup>-1</sup> along with the POP-treated plot (T<sub>8</sub>) had the numerically higher available phosphorus content (189.49 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>). Wherein absolute control (T<sub>1</sub>) had numerically lower amount of available phosphorus (102.73 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>).

TABLE 3

Effect of long-term application of graded levels of boron on available N, P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O of soil after harvest of sunflower

Treatment details	N (kg ha <sup>-1</sup> )	P <sub>2</sub> O <sub>5</sub> (kg ha <sup>-1</sup> )	K <sub>2</sub> O (kg ha <sup>-1</sup> )
T <sub>1</sub> Control	252.97	102.73	131.25
T <sub>2</sub> POP without Boron	258.86	159.24	147.84
T <sub>3</sub> POP+ 2 kg B ha <sup>-1</sup>	257.15	169.53	151.93
T <sub>4</sub> POP+ 4 kg B ha <sup>-1</sup>	260.03	172.84	154.71
T <sub>5</sub> POP+ 6 kg B ha <sup>-1</sup>	265.06	176.82	157.45
T <sub>6</sub> POP+ 8 kg B ha <sup>-1</sup>	261.33	179.28	161.00
T <sub>7</sub> POP+ 12 kg B ha <sup>-1</sup>	268.86	184.37	162.33
T <sub>8</sub> POP+ 16 kg B ha <sup>-1</sup>	270.92	189.49	163.51
S Em ±	13.72	18.94	10.79
C D @ 5%	NS	NS	NS

Due to differing levels of boron application, the soil available potassium concentration did not significantly differ between the treatments either but, the K status of soil after post-harvest has increased in the different levels of boron application compared to control. However, with 16 kg B ha<sup>-1</sup> combined with POP (T<sub>8</sub>), a numerically larger amount of available potassium (163.51 kg K<sub>2</sub>O ha<sup>-1</sup>). From 2 kg to 16 kg B there was slight increase in available potassium. While in the absolute control (T<sub>1</sub>), had lower potassium levels (131.25 kg K<sub>2</sub>O ha<sup>-1</sup>).

Different boron application levels had no significant effect on the amount of available nitrogen, phosphorous and potassium in the soil. However numerically higher nitrogen and phosphorus availability were seen in the treatment receiving 16 kg B ha<sup>-1</sup>, also potassium concentration in the soil increased up to the same level of B. These results concur with those of Banasode and Channakeshava (2021), who noted an increase in available nitrogen due to the mineralization of additional FYM along with Borax application. Higher dose of boron in the soil induces the desorption of H<sub>2</sub>PO<sub>4</sub><sup>-</sup> and NO<sub>3</sub><sup>-</sup> from the adsorption sites by H<sub>3</sub>BO<sub>3</sub><sup>-</sup> makes them available in the soil. Higher amounts of boron may interact favourably with an increase in K, balancing cations and anion in the soil (Sathi Babu *et al.*, 2017). Similar to this, Das *et al.* (2014) found that a high B concentration in the soil enhances the availability of potassium to plants.

### Boron Fertilization and Sunflower Crop Growth

The observations relating to selected growth attributing characteristics (e.g., plant height and no. of leaves) as influenced by graded levels of B fertilization at different growth stages *viz.*, 30 DAS, 60 DAS and at harvest have been recorded and data are given in Table 4. Significantly highest mean plant height (27.07, 113.73 and 124.07 cm) at 30, 60 DAS and at harvest, respectively were recorded in treatment T<sub>2</sub> where POP was applied without boron. However, gradual decrease in the plant height and no of leaves at all the intervals of growth period was observed due to the application of boron from 2 kg ha<sup>-1</sup> to

TABLE 4  
Influence of graded levels of boron application on plant height and no. of leaves of sunflower crop at different stages

Treatment details	Plant height (cm)			No. of leaves		
	30 DAS	60 DAS	Harvest	30 DAS	60 DAS	Harvest
T <sub>1</sub> Control	18.20	66.67	75.07	10.40	17.27	25.00
T <sub>2</sub> POP without Boron	27.07	113.73	124.07	13.53	24.47	30.40
T <sub>3</sub> POP+ 2 kg B ha <sup>-1</sup>	23.73	111.00	120.87	12.93	24.00	27.60
T <sub>4</sub> POP+ 4 kg B ha <sup>-1</sup>	22.73	92.07	96.20	12.07	21.27	27.27
T <sub>5</sub> POP+ 6 kg B ha <sup>-1</sup>	18.53	76.67	83.33	10.27	20.00	25.67
T <sub>6</sub> POP+ 8 kg B ha <sup>-1</sup>	17.87	76.60	87.47	10.07	19.67	25.13
T <sub>7</sub> POP+ 12 kg B ha <sup>-1</sup>	16.87	73.27	85.60	9.67	19.53	24.93
T <sub>8</sub> POP+ 16 kg B ha <sup>-1</sup>	14.93	68.13	77.93	9.00	19.40	24.87
S.Em. ±	2.12	7.60	6.97	0.37	0.80	0.88
C.D. @ 5%	6.43	23.06	21.15	1.11	2.44	2.66

16 kg ha<sup>-1</sup>. But, the plant height in treatment T<sub>3</sub> treated with POP + 2 kg B ha<sup>-1</sup> (23.73, 111.11 and 120.87 cm) was found on par with POP + without boron (T<sub>2</sub>) throughout the growth period. Significantly lower plant height was recorded in absolute control (T<sub>1</sub>) (66.67 and 75.07 cm) followed by T<sub>8</sub> where POP + 16 kg B ha<sup>-1</sup> (68.13 and 77.93 cm) was applied at 60 DAS and at harvest, respectively, whereas at 30 DAS lower plant height was recorded in T<sub>8</sub> where POP + 16 kg B ha<sup>-1</sup> (14.93 cm) followed by T<sub>7</sub> (POP + 12 kg B ha<sup>-1</sup>).

There was a significant differences in number of leaves per plant at all intervals with application of different levels of boron. Significantly higher number of leaves (13.53, 24.47 and 30.40) at 30, 60 DAS and at harvest, respectively were recorded with the application of POP without boron (T<sub>2</sub>), whereas at 30 DAS and harvest number of leaves was significantly lower (9.00 and 24.87, respectively) in T<sub>8</sub> (POP + 16 kg B ha<sup>-1</sup>) followed by T<sub>7</sub> (POP + 12 kg B ha<sup>-1</sup>). At 60 DAS, lowest number of leaves was recorded in absolute control (T<sub>1</sub>) (17.27) followed by T<sub>8</sub> where POP + 16 kg B ha<sup>-1</sup> (19.4) was applied. Continuous application of B increased the level of boron in the soil and toxic conditions began to set in, thereby

exerting adverse effects on plant metabolic activities that consequently affect plant height negatively. Regular application of more than 2 kg ha<sup>-1</sup> B fertilizer annually or application of irrigation water high in B leads to B toxicity in plants and reduced crop yields (Singh *et al.*, 2005). Lower boron levels undergo rapid cells division and differentiate hence increase in plant height and photosynthetic rate (Bonilla *et al.*, 2004). The significant increase in growth parameters at low concentration of boron could be due to its involvement in cell elongation or cell division and meristematic growth (Khan *et al.*, 2006).

Increase in different levels of boron up to 16 kg B ha<sup>-1</sup> along with POP, decreased all the growth and yield attributes beyond 2 kg B ha<sup>-1</sup> along with POP. It was due to excessive amount of boron appears to inhibit the formation of starch from sugars in turn results in the formation of carbohydrates complexes, and thus fails to increase growth and yield attributes (Soad Soliman EI- Feky *et al.*, 2012).

#### Boron Fertilization and Sunflower Yield Attributes and Oil Content

The initial soil boron content shows that, the continuous application of boron increased the boron

content to a toxic level with increasing dose of application (Table 3). The highest initial boron content was observed in T<sub>8</sub> (5.51 mg kg<sup>-1</sup>) where, 16 kg B ha<sup>-1</sup> with POP was applied and lowest soil boron status was found in absolute control (0.46 mg kg<sup>-1</sup>) followed by treatment (T<sub>2</sub>) no boron with POP (0.58 mg kg<sup>-1</sup>). Due to the application of heavy dose of boron, the germination was affected from 2 kg B ha<sup>-1</sup> to 16 kg B ha<sup>-1</sup> applied plots at 7 and 13 DAS (Table 6). The per cent germination of sunflower recorded at 7 and 13 DAS indicates, that significantly higher germination was observed in absolute control treatment (73.3 and 93.33%, respectively) and it was on par with treatment (T<sub>2</sub>) no boron with POP (85%) at 13 DAS. But, after gap filling with the seedlings grown in pockets, crop stand was good however, crop growth and yield was affected as the boron levels increased.

The long-term application of boron on sunflower crop recorded a decrease in test weight, number of seeds per head, head weight and head diameter with increase in levels of boron (Table 5). A significant higher test weight (75.19g), number of filled seeds per head (586.6) and head diameter (18.8cm) in treatment T<sub>2</sub> (POP without Boron) were recorded whereas the lowest test weight (47.13g) was recorded in higher dose of boron applied treatment (T<sub>8</sub>: POP +

16 kg B ha<sup>-1</sup>) but, absolute control treatment recorded lowest in number of filled seeds (298.2) and head diameter (51.17 cm) in T<sub>7</sub> (POP+ 12 kg B ha<sup>-1</sup>). The capitulum was often mal-formed with poor seed set due to the toxicity effect on continuous application of boron over the years. Similarly, Ceyhan *et al.* (2008) reported that B application as average of years, decreased diameter of head and 1000 seed weight with increasing B dose.

The number of chaffy and total seeds as well as head weight of sunflower crop after harvest was showing non significant results with the application of graded levels of boron. However, highest number of chaffy seeds per head (486.1) was recorded in treatment of (T<sub>7</sub>) POP+ 12 kg B ha<sup>-1</sup> and number of total seeds per head (963.1) and head weight (17.8 kg) were recorded in the treatment of POP without Boron (T<sub>2</sub>). Lowest number of chaffy seeds per head (337.0), number of total seed per head (612.8) and head weight (11.7kg) were recorded at absolute control, (T<sub>1</sub>) POP + 6 kg B ha<sup>-1</sup> and (T<sub>8</sub>) POP + 16 kg B ha<sup>-1</sup>, respectively. Shirur *et al.* (2021) observed, significantly higher number of tubers per plant, tuber weight and tuber yield per hectare were recorded in potato with the application of 150 kg Gypsum ha<sup>-1</sup> + Foliar spray of 0.5 per cent Boron along with RDF +FYM.

TABLE 5

Effect of long-term application of graded levels of boron on sunflower seeds test weight, number of chaffy, filled and total seeds per head, head weight and diameter after harvest

Treatment details	Test weight (g)	Number of seeds per head			Head weight (kg)	Head diameter (cm)
		Chaffy seeds	Filled seeds	Total		
T <sub>1</sub> Control	47.13	337.0	298.2	635.2	5.2	12.4
T <sub>2</sub> POP without Boron	75.19	343.1	586.6	963.1	12.0	17.8
T <sub>3</sub> POP+ 2 kg B ha <sup>-1</sup>	70.10	218.7	361.4	580.1	10.1	13.5
T <sub>4</sub> POP+ 4 kg B ha <sup>-1</sup>	62.69	207.7	358.7	566.3	10.2	14.3
T <sub>5</sub> POP+ 6 kg B ha <sup>-1</sup>	51.72	260.8	352.0	612.8	7.3	12.4
T <sub>6</sub> POP+ 8 kg B ha <sup>-1</sup>	48.02	375.5	347.9	723.3	7.2	11.8
T <sub>7</sub> POP+ 12 kg B ha <sup>-1</sup>	39.61	486.1	342.9	829.0	6.3	12.0
T <sub>8</sub> POP+ 16 kg B ha <sup>-1</sup>	35.53	482.6	338.2	820.9	5.8	11.7
S.Em. ±	0.39	55.7	47.4	86.8	1.20	1.3
C.D. @ 5%	1.19	NS	145.0	NS	3.62	NS

TABLE 6

Effect of long-term application of graded levels of boron on germination percentage, seed yield, stalk yield and oil content of sunflower crop with initial soil B concentration

Treatment details	Germination per cent		Seed yield (q ha <sup>-1</sup> )	Stalk yield (q ha <sup>-1</sup> )	Oil content (%)
	7 DAS	13 DAS			
T <sub>1</sub> Control	73.33	93.33	11.64	12.91	32.98
T <sub>2</sub> POP without Boron	57.50	85.00	20.47	30.88	34.22
T <sub>3</sub> POP+ 2 kg B ha <sup>-1</sup>	39.17	56.67	18.71	27.17	35.72
T <sub>4</sub> POP+ 4 kg B ha <sup>-1</sup>	20.83	32.50	17.76	26.96	36.69
T <sub>5</sub> POP+ 6 kg B ha <sup>-1</sup>	13.33	21.67	16.34	24.32	35.64
T <sub>6</sub> POP+ 8 kg B ha <sup>-1</sup>	10.00	15.00	15.14	22.32	33.51
T <sub>7</sub> POP+ 12 kg B ha <sup>-1</sup>	2.50	4.17	14.98	21.45	32.38
T <sub>8</sub> POP+ 16 kg B ha <sup>-1</sup>	0.83	0.83	13.28	15.44	31.81
S.Em. ±	2.79	3.49	0.77	8.42	8.42
C.D. @ 5%	5.98	10.60	2.34	2.78	2.78

The perusal of the data in (Table 6) related to seed yield indicated that there was a significant difference due to application of different levels of boron. Among the different levels of boron application to sunflower crop the treatment POP without boron application (T<sub>2</sub>) recorded significantly higher grain yield (20.47 q ha<sup>-1</sup>) compared to other treatments. However, it was found to be on par with T<sub>3</sub> (POP + 2 kg B ha<sup>-1</sup>) (18.71 q ha<sup>-1</sup>) was applied. Among boron treatments, significantly lower grain yield (11.64 q ha<sup>-1</sup>) was observed in absolute control where no POP and no boron was applied. The yield reduction was to the tune of 8.6 to 35.1 per cent from 2 kg B to 16 kg B ha<sup>-1</sup>. Statistically stalk yield was significantly higher (30.88 q ha<sup>-1</sup>) in treatment (T<sub>2</sub>) without boron + POP and was decreasing with increased application of boron in the soil. The significantly lower stover yield (73.72 q ha<sup>-1</sup>) was recorded in absolute control (T<sub>1</sub>) when compared to other treatments. Even though the available N, P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O increased after harvest of sunflower with increasing dose of B, the results showed that the toxicity effect of B was prominent in retarding the growth and yield of sun flower due to higher levels of B in soil.

The oil content in sunflower seeds significantly increased with application of POP + 4 kg B ha<sup>-1</sup> (T<sub>4</sub>) (36.69%) followed by T<sub>3</sub>, where 2 kg B ha<sup>-1</sup> with

POP (35.72%) and (T<sub>2</sub>) no boron with POP (34.22). The oil content was significantly reduced with application of higher dose of boron (T<sub>8</sub>) @ 16 kg B ha<sup>-1</sup> (31.81%) compared to absolute control (32.98%).

The Fig. 1. showing effect of long-term application of graded levels of boron on seed yield, stalk yield and oil content of sunflower crop with initial soil B content illustrated the adverse effect of toxicity of B in soil on sunflower seed and stalk yield. Excess of B deteriorated the quality of sunflower seeds by lowering the plant height, number of leaves, head diameter number of filled grains and test weight. The B toxicity might be due to direct effect of B on pollen viability, fertility and seed set in plant (Fang *et al.*, 2016). High B concentration in leaves can cause

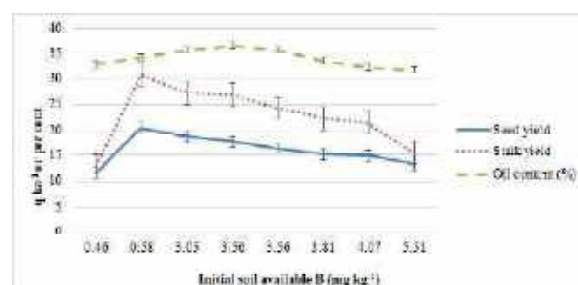


Fig. 1 : Effect of long-term application of graded levels of boron on seed yield, stalk yield and oil content of sunflower crop with initial soil B concentration



a decrease in photosynthesis as reported for olives (Chatzissavvidis and Therios, 2010). A reason for this decline could be the reduction of N concentration in leaves because most of this element in green tissues is utilized in the photosynthetic machinery (Simon *et al.*, 2013). Residual effects of B on yield was reported by Dongale and Zende (1977) in wheat, while Ratna Kalyani *et al.* (1993) reported that the application of B to the pigeon pea crop played a critical role in reducing flower and pod drop by preventing the formation of an abscission layer. At lower dose of boron, the application Zn and B along with NPK and FYM increased growth and yield of paddy as well as residual crop cowpea (Banasode and Channakeshava, 2021). The increase in oil content may be due to the positive influence of B on bio-synthesis of oil and fatty acids (Malewar *et al.* 2001 and Mallick & Raj 2015). The increase in oil content of mustard due to B application have also been reported by Mandal and Das (2014) and Jaiswal *et al.* (2015) Yadav *et al.* (2016).

The essential tissue boron concentrations for many plant species fall within a very small range between deficiency and toxicity. Boron toxicity has a variety of consequences on vascular plants, including lowered photosynthetic rates and impaired root cell division. As a result, plants exposed to increased levels of boron typically exhibit lower development of shoots and roots. A mottling of the lowest leaves tips and edges is the earliest sign of boron toxicity in sunflower and these spotted regions eventually perish. The entire plant growth is hampered in cases of severe injury, which leads to low yields and poor quality oil. Due to continued application of boron for five years, POP along without boron practise, showed initial soil boron status at its ideal level, which led to good plant growth at various phases and a high yield of sunflowers. In the initial stage (*kharif* 2020) of the experiment soil boron increase was from 3.05 to 5.51 mg kg<sup>-1</sup> and after the harvest was also increased to a higher concentration (3.46 to 5.57 mg kg<sup>-1</sup>) from 2 to 16 kg ha<sup>-1</sup>, respectively. Thus, toxicity was more severe at the higher dose of boron where, the test weight, number of filled seeds per head, head weight, and head diameter per plant was decreased finally,

yield was lowered by 8.6 to 35.1 per cent from 2 kg B to 16 kg B ha<sup>-1</sup>. Also with treatment of a greater dose of boron the oil content was significantly decreased (T8) @ 16 kg B ha<sup>-1</sup> (31.81%) compared to the absolute control (32.98%). Different boron application levels had no significant effect on the amount of available nitrogen, phosphorous and potassium in the soil. However numerically higher nitrogen, phosphorus and potassium availability were seen in the treatment receiving 16 kg B ha<sup>-1</sup>. But, higher B treatments partially hindered a variety of cellular processes, which could prevent roots from absorbing nutrients. Hence the treatment where POP without B showed the highest seed production (20.47 q ha<sup>-1</sup>) and stalk yield (30.88 q ha<sup>-1</sup>) whereas it considerably decreased with an increase in B application rate. This makes it quite evident that not even the recommended 2 kg B ha<sup>-1</sup>, should be administered consistently without first assessing the soil, since this could lead to boron accumulation, toxicity and decreased crop yield. The B toxicity might be due to direct effect of B on germination, pollen viability, fertility and seed set which reduced the sunflower seed yield and oil content also the plant growth of sunflower crop.

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## Effect of Temperature, Soil Moisture and Elevated CO<sub>2</sub> on Growth of *Rhizoctonia bataticola* and Dry Root Rot Disease of Chickpea in Karnataka State

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### ABSTRACT

Dry root rot caused by *Rhizoctonia bataticola* is becoming an emerging disease as per the new reports and considered as potential threat to chickpea productivity and production under changing climatic scenario. The identity of pathogen was confirmed molecularly using ITS-1 and ITS-4 primers which produced amplified product size of 500-650 bp in all three isolates indicating that all the isolates belonged to species *Rhizoctonia bataticola*. The maximum colony growth of *R. bataticola* and the dry root rot disease severity was recorded at 30-35 °C which is considered as optimum temperature range for growth of pathogen and development of disease. Highest severity of dry root rot and lesser plant growth parameters such as root length, shoot length and total biomass were observed at 40-60 per cent soil moisture regimes, irrespective of type of soil. Further, elevated CO<sub>2</sub> @ 550 ± 25 ppm with 2 °C rise in temperature recorded higher dry root rot well as reduced growth parameters of chickpea. With respect to correlation studies, increase in the temperature lead to decreased radial growth of pathogen and dry root rot disease incidence. It is also inferred that increase in the soil moisture led to increase in growth parameters in both black as well as red soils.

**Keywords :** Dry root rot of chick pea, Climate change, Temperature, Soil moisture, Disease severity

CHICKPEA is one of the most important food legumes being cultivated in almost all over the world including temperate and sub-tropical regions. The crop faces various problems throughout the growing areas, some related to specific regions and some under wider range of climatic conditions. Chickpea cultivation is often subjected to significant yield losses due to insects and diseases ranging from 5-10 per cent in temperate and 50-100 per cent in tropical regions (Van Emden *et al.*, 1988).

The recent reports indicated that dry root rot is emerging as a potential threat to chickpea productivity and production (Ghosh *et al.*, 2013). The disease is more prevalent during hot temperature 30 to 35 °C and low soil moisture conditions (Pande *et al.*, 2010). Dry root rot caused by *Rhizoctonia bataticola* (Taub.) Butler [Pycnidial stage: *Macrophomina phaseolina* (Tassi) Goid] is a soil and seed borne necrotrophic

fungal pathogen that has a global distribution, which can infect more than 284 plant species throughout the world including monocot and dicots (Farr *et al.*, 1995). *Rhizoctonia bataticola* does not produce spores, but are composed of hyphae and sclerotia (hyphal propagules) acting as facultative plant pathogen causing complete loss in grain yield if chickpea crop is infected.

Environmental conditions like temperature, soil moisture and carbon dioxide play an important role in the viability and growth of *R. bataticola* (Khan, 2007). *R. bataticola* is able to produce microsclerotia under relatively low water conditions while viability of microsclerotia is drastically reduced at high water potentials (Olaya and Abawi, 1996). Temperature and soil moisture are the two important weather parameters influencing the dry root rot infection, colonization and development in chickpea. In the

changing climatic scenario, studies on impact of climatic factors on pathogen and disease are scanty. Moreover, the crop is largely grown in rainfed environment and change in climatic factors within the rainfed ecologies may lead to varying degrees of growth of pathogen and intensities of dry root rot. Keeping this in view, studies on impact of climatic variables on growth of pathogen and disease incidence is felt necessary under changing climatic scenario. Hence, the effect of three climatic change variables viz., temperature, moisture and carbon dioxide was studied on growth of pathogen and development of disease during present investigation.

## MATERIAL AND METHODS

### Isolation, Purification and Detection of *R. bataticola* Isolates

Chickpea plants showing typical dry root rot symptoms were collected from three different geographic regions (Region 1, Region 2 and Region 3) of Karnataka and used for isolation of pathogen. Infected roots were cut into pieces of 5-6 mm and were transferred on to sterilized potato dextrose agar (PDA) medium in Petri dishes and incubated at  $25 \pm 2$  °C to obtain mycelial growth. After 48h of incubation, hyphal tips of the growing mycelium were marked on the underside of the petri dish with a glass marker by viewing through a light microscope. The cultures were purified by placing single sclerotial body transferred to PDA slants. Later, Koch postulates were carried out for all three isolates to confirm the pathogenicity of pathogen.

### Molecular Detection of Isolates

Total DNA of three isolates of pathogen was extracted using Cetyl-Trimethylammonium Bromide (CTAB) method from young vegetative mycelium using the procedure given by Murray and Thompson (1980). The Internal Transcribed Spacers region was sequenced from three isolates belonging to three different geographic locations to confirm the identity of isolates. PCR (Mastercycler, Hamburg, Germany) amplifications of the ITS of rDNA was performed by using universal primers

ITS-1 (52CCTGTGCACCTGTGAGACAG-32 ) as forward primer and ITS-4 (52 - TGTCCAAGTCAAT GGACTAT-32 ) as reverse primer. The PCR product was sequenced using forward and reverse primers at Medauxin Biotech. Ltd., Bengaluru. Homology search was done using BLAST algorithm available at the <http://www.ncbi.nlm.nih.gov>,

### Effect of Climate Change Variables on *R. bataticola* and Dry Root Rot Incidence

The effect of three climatic change variables viz., temperature, moisture and carbon dioxide was studied on growth of pathogen and development of disease during present investigation.

#### Effect of Temperature on *R. bataticola*

Three isolates of pathogen namely, Rb1, Rb2 and Rb3 were used to study the effect of temperature on pathogen virulence. The isolates were inoculated on PDA medium and kept for incubation at different temperature regimes, 15 °C, 20 °C, 25 °C, 30 °C, 35 °C, 40 °C and 45 °C. Later, the radial growth of mycelium of pathogen was recorded.

#### Effect of Temperature on Dry Root Rot Disease

Chickpea (Annigeri-1) seedlings were raised in small plastic containers which were filled with pre-autoclaved sand. Chickpea seeds are sown in these containers with 2 cm deep. Moisture was maintained by watering on daily basis, after ten days seedlings were used for the study. To study the effect of temperature on dry root rot, each isolate of pathogen was inoculated to ten days old seedlings separately, grown under *in vitro* conditions using paper towel technique. Inoculated seedlings were placed in folded, moist blotting paper with the shoots left outside and then incubated at different temperature regimes 15, 20, 25, 30, 35, 40, 45 and 50 °C with a 12 h photoperiod. The experiment was conducted with Completely Randomized Block Design (CRBD) with three replications (each replication consisted of 5 plants). Total 15 plants per treatment were scored for disease severity of dry root rot and the disease severity was recorded using 1-9 rating scale (Sharma and Pande, 2013) (Table 1).

### Effect of Soil Moisture on Dry Root Rot Disease

Pot experiment was conducted to know the effect of soil moisture on dry root disease in glasshouse conditions. To obtain large amount of inoculum, the fungus was multiplied on sorghum grain medium and sufficient inoculums was applied to pots. The pots were incubated for 4 days to allow the pathogen to multiply in soil. The effect of seven soil moisture regimes, *i.e.* 40, 50, 60, 70, 80, 90 and 100 per cent was studied with black and red soils separately for the development of disease. Each treatment was replicated four times and each replication consisted of three pots (five plants/pot). Deionized water was used for maintaining the soil moisture content (SMC) in each treatment. The SMC was determined using the gravimetric method on an oven-dry basis. The method includes saturation of soil sample followed by removal of available soil moisture by oven drying (100 - 110 °C) until the weight remains constant. After removing from oven, samples were cooled slowly to room temperature and weighed again. The difference in weight was amount of moisture in the soil. The available SMC in the soil was calculated by the following formula.

$$\text{SMC (\%)} = \frac{\text{Saturated soil weight} - \text{oven dry soil weight}}{\text{Oven dry soil weight}} \times 100$$

### Effect of Carbon Dioxide (CO<sub>2</sub>) on Dry Root Rot

The study was conducted in the open top chambers maintained by Centre for Climatic Studies, University of Agricultural Sciences, Raichur. Totally, five sets of treatments were made to study the effect of carbon dioxide on dry root rot of chickpea.

- T<sub>1</sub> : Elevated CO<sub>2</sub> @ 550 ± 25 ppm alone
- T<sub>2</sub> : Elevated CO<sub>2</sub> @ 550 ± 25 ppm with 2 °C rise in temperature
- T<sub>3</sub> : Ambient CO<sub>2</sub> @ 390 ± 25 ppm with 2 °C rise in temperature
- T<sub>4</sub> : Reference Open Top Chamber
- T<sub>5</sub> : Open plot

Under the open top chamber, susceptible chickpea variety (Annigeri-1) was sown in the sick pots. Each

sick pot was sown with five seeds and for each treatment five replications were maintained. Observations on growth parameters and disease severity of dry root rot were recorded at 75 DAS.

### Statistical Analysis

The data obtained in the laboratory as well as open top chamber experiments through Factorial Completely Randomized Design were analyzed by Statistical Package for Social Sciences (SPSS V.20). Further, correlation analysis was carried out to understand the relationship between the parameters.

## RESULTS AND DISCUSSION

### Isolation, Purification and Detection of *R. bataticola* Isolates

The results indicated that *R. bataticola* pathogen isolates produced black, brown to grey coloured mycelium that become darker with age (Fig. 1). The young hyphae were thin, hyaline, septate and dichotomously branched and later produce typical black sclerotia. The characteristic features of *R. bataticola* were right angle branching of the mycelium and constriction of the branch near the point of origin. The sclerotia formed were black, smooth, varying from spherical through oblong to irregular shapes. Later, the pathogenicity was proved successfully by following Koch postulates. In molecular detection, both ITS-1 and ITS-4 primers produced amplified product size of 500-650 bp in all the three isolates indicating that all the isolates are *Rhizoctonia bataticola*. Further, nucleotide sequencing was done for ITS region of 18S rRNA. The BLAST data results revealed that the *R. bataticola* species matched with the reference strains of NCBI results and identified as *Rhizoctonia bataticola*. The sequences are deposited in NCBI GeneBank, Maryland, USA along with location of the isolates and accession number Rb1 (KX270355.1), Rb2 (MG001962.1) and Rb3 (HQ392772.1) were obtained. Similarly, Aghakhani and Dubey (2009) isolated 23 isolates of *R. bataticola* from chickpea plants showing characteristic dry root rot symptoms from different chickpea growing states of India.

TABLE 1  
Disease rating scale of 1-9 for dry root rot of chickpea

Rating	Observation
1	No infection on roots
>1 and <3	Very few small lesions (black discoloration) on roots
>3 and <5	Lesions (black discoloration) on roots clear but less; new roots free from infection
>5 and <7	Lesions (black discoloration) on roots more; many new roots generally free from lesions
>7 and 9	Roots infected and completely discoloured (black)

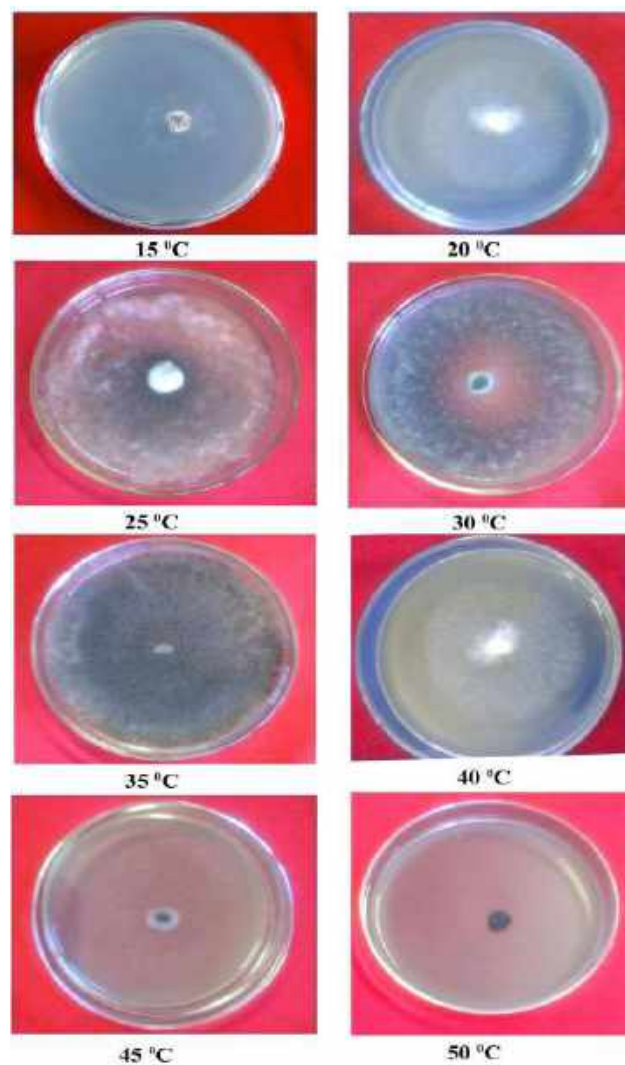


Fig. 1: Effect of temperature on radial growth of *R. bataticola*

Further, Sharma *et al.* (2012) also proved the pathogenicity of 50 isolates of *R. bataticola* using chickpea cultivar BG 212.

### Effect of Climate Change Variables on *R. bataticola* and Dry Root Rot Incidence

The effect of three climatic change variables *viz.*, temperature, moisture and carbon dioxide was studied on growth of pathogen and development of disease during present investigation was studied and results are presented and discussed here under.

### Effect of Temperature on Pathogen

Among three isolates of pathogen, the results indicated that the maximum colony growth was observed in Rb3 (4.5, 31, 47, 52, 68 and 15 mm at 15, 20, 25, 30, 35 and 40 °C, respectively) at 48 h after inoculation. However, the least growth of pathogen was observed in Rb1 (1.40, 29, 32, 40, 50 and 10 mm at 15, 20, 25, 30, 35 and 40 °C respectively). The results also indicated that the temperature levels such as 45 and 50 °C recorded lesser growth in all the three isolates (Table 2 and Fig. 2).

At 96 h after inoculation, the maximum colony growth was observed in Rb3 (18.15, 61.00, 75.50, 86.50, 90.00 and 18.00 mm at 15, 20, 25, 30, 35, 40 °C, respectively) and this is followed by isolate Rb1 (12.55, 50.00, 64.00, 85.00, 90.00 and 17.00 mm) and least growth was observed in Rb2 (10.20, 34.20, 37.00, 47.70, 80.00 and 15.00 mm at 15, 20, 25, 30, 35 and 40 °C, respectively). All three pathogen isolates did not grow at 45 and 50 °C as observed at 48 h after inoculation (Table 2).

The present investigations are in line with Srinivas (2016) who reported that significant difference in the radial growth among the isolates of *R. bataticola* ranging from 17.7 mm to 80.0 mm at 72 h after incubation. Isolate Rb 14, Rb 17, Rb 22, Rb 26, Rb 49 and Rb 54 showed significantly highest colony growth (80 mm). The least colony diameter was observed in the isolate Rb 20 (17.7 mm). The

TABLE 2

Effects of temperature regimes on radial growth of *R. bataticola* at 48 and 96 hours after inoculation

Temperature regimes (°C)	Rb1		Rb2		Rb3		Mean	
	48 HAI	96 HAI	48 HAI	96 HAI	48 HAI	96 HAI	48 HAI	96 HAI
15	2.50	12.55	1.40	10.20	4.50	18.15	2.80	13.66
20	30.00	50.00	29.00	34.20	31.00	61.00	30.00	48.40
25	43.00	64.00	32.00	37.00	47.00	75.50	40.66	58.80
30	50.00	85.00	40.00	47.70	52.00	86.50	47.33	73.06
35	65.00	90.00	50.00	80.00	68.00	90.00	61.00	86.66
40	12.00	17.00	10.00	15.00	15.00	18.00	12.33	16.66
45	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
50	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Mean	25.31	39.81	20.31	28.01	27.18	43.64	24.26	37.15

\*Mean of three replications HAI- Hours after inoculation

Factors	S.Em±	CD @ 1%
Temperature (T)	0.30	0.95
Isolate (I)	0.32	0.98
T x I	0.34	1.11

maximum radial growth (90 mm) of the *R. bataticola* was recorded at 30-35 °C as observed in present investigation (Veerendra Kumar, 2004)

**Effect of Temperature on Dry Root Rot**

The maximum disease severity was recorded in Rb3 with 1.7, 3.1, 7.4, 8.2, 9.0 and 3 grade on 1-9 scale followed by Rb1 (1.3, 3.0, 7.2, 8.0, 9.0 and 3.0) at 15, 20, 25, 30, 35 and 40 °C, respectively. Whereas the least disease severity rating was recorded in Rb2 (1.0, 2.9, 6.7, 7.2, 8.5 and 2.5 at 15, 20, 25, 30, 35 and 40 °C, respectively (Table 3 and Fig. 2). However, there was dry root rot development at 45 and 50 °C (Table 3).

The correlation results indicated that the negative correlation coefficients (-0.29 and -0.38 for 48 and 96 hours, respectively) between temperature regimes and radial growth of *R. bataticola* suggest a weak negative relationship between the two variables (Table 4). This means that as the temperature regime increases, the radial growth of *R. bataticola* is likely to decrease. However, the same pattern was observed

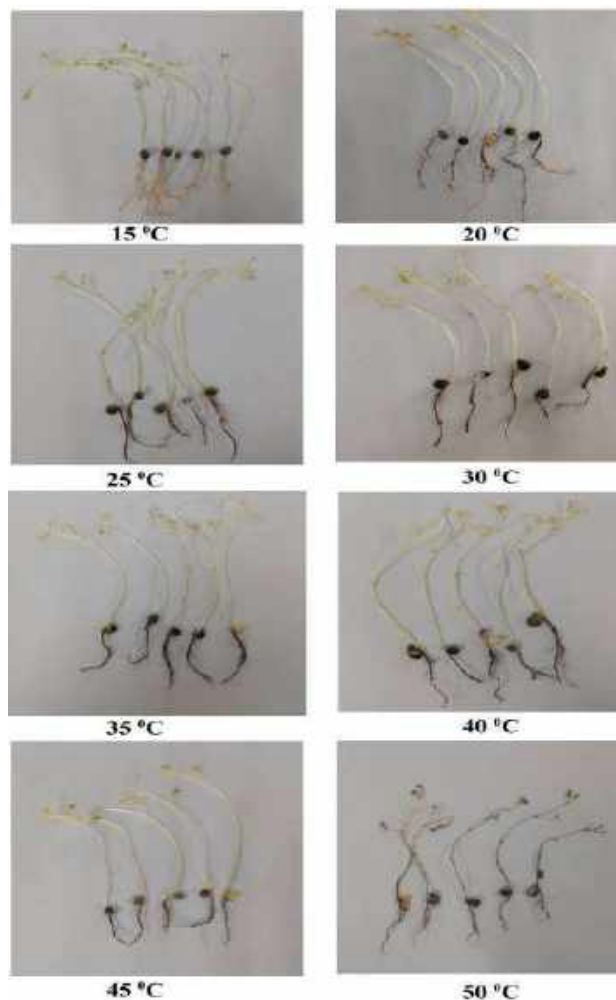


Fig. 2 : Effect of temperature regimes on dry root rot disease in chickpea



TABLE 3  
Effects of temperature regimes on dry root rot disease caused by *R. bataticola* isolates on Annigeri-1 variety

Temperature (°C)	*Disease severity (1-9 scale)		
	Rb1	Rb2	Rb3
15	1.3	1.0	1.7
20	3.0	2.9	3.1
25	7.2	6.7	7.4
30	8.0	7.2	8.2
35	9.0	8.5	9.0
40	3.00	2.5	3.0
45	0.00	0.00	0.00
50	0.00	0.00	0.00
Mean	3.93	3.60	4.05

\*Mean of three replications

Factors	S.Em±	CD @ 1%
Temperature (T)	0.20	0.65
Isolate (I)	0.22	0.72
T x I	0.16	0.69

with disease severity in *R. bataticola* isolates that is as the temperature regime increases, the disease severity by *R. bataticola* isolates is likely to decrease (Table 4). Sharma and Pande (2013) reported that the disease incidence of dry root rot was significantly affected by high temperature. Out of five temperature levels viz., 15, 20, 25, 30 and 35 °C tested, chickpea predisposed to dry root rot early and severity was more at 35 °C. Savary *et al.* (2011) observed the effect

TABLE 4  
Correlation Analysis of temperature regimes on radial growth of *R. bataticola* and Disease severity *R. bataticola* isolates on Annigeri-1 variety

Temperature Levels	Radial growth	Correlation Coefficient
Temperature	48 HAI(mm)	-0.29*
	96 HAI(mm)	-0.38*
	<i>Disease severity</i>	
	Rb1	-0.28*
	Rb2	-0.28*
	Rb3	-0.31*

\*\* Significant at 5% LOS

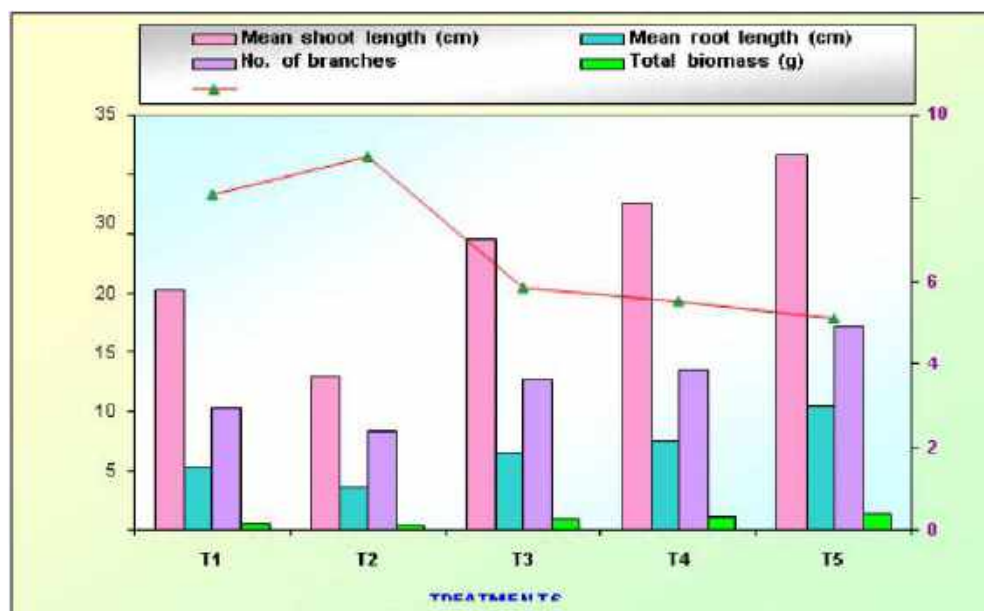


Fig. 3: Effect of carbon and temperature on dry root rot disease

**Treatment details:**

T<sub>1</sub> - Elevated CO<sub>2</sub> @ 550 ± 25 ppm alone; T<sub>2</sub> - Elevated CO<sub>2</sub> @ 550 ± 25 ppm with 2 °C rise in temperature  
T<sub>3</sub> - Ambient CO<sub>2</sub> @ 390 ± 25 ppm with 2 °C rise in temperature T<sub>4</sub> - Ambient CO<sub>2</sub> @ 390 ± 25 ppm; T<sub>5</sub> - Open plot

TABLE 5

Effect of soil moisture levels on growth parameters of chickpea in black soil inoculated with *R. bataticola*

Moisture (%)	Growth parameters											
	Root length (cm)				Shoot length (cm)				Total biomass (g)			
	Rb1	Rb2	Rb3	Mean	Rb1	Rb2	Rb3	Mean	Rb1	Rb2	Rb3	Mean
40	2.05	2.10	2.00	2.14	8.20	8.67	8.50	8.45	0.37	0.40	0.38	0.38
50	2.76	2.80	2.70	2.75	10.00	11.00	10.07	10.35	0.45	0.55	0.37	0.45
60	3.10	3.45	3.00	3.18	11.23	11.50	11.00	11.24	0.48	0.65	0.35	0.49
70	3.57	3.55	3.38	3.50	20.10	23.00	19.19	20.76	0.67	0.70	0.61	0.66
80	4.56	4.80	4.00	4.16	25.04	26.00	24.59	25.21	1.10	1.54	1.33	1.32
90	4.16	4.20	4.15	4.45	29.54	29.70	29.20	29.48	3.40	3.27	3.51	3.45
100	4.88	5.23	4.80	4.97	29.81	29.88	29.00	29.56	3.77	3.90	3.80	3.82
Mean	3.59	3.74	3.43	3.60	19.13	19.96	18.79	19.29	1.36	1.89	1.35	1.42

\* Mean of three replications

Factors	S.Em±	CD @ 1%
Temperature (T)	0.29	0.93
Isolate (I)	0.32	1.01
T x I	0.41	1.31

**Root Length**

The highest mean root length of three isolates (Rb1, Rb2 and Rb3) was observed in red soil (3.67 cm) compared to the black soil (3.60 cm), but there was no significant difference (Table 5 and 6). Among the three isolates, maximum mean root length of 3.74 cm and 3.90 cm was recorded in both the soils by Rb2 followed by Rb1 (3.59 cm and 3.60 cm). Whereas Rb3 recorded the least root length in two types of soils (3.43 cm and 3.54 cm).

of temperature on increasing incidence of dry root rot at various locations over the years which suggests a strong influence of rising temperature (above 30 °C) on *R. bataticola*.

**Effect of Soil Moisture on Dry Root Rot**

The effect of dry root rot on different growth parameters such as root length, shoot length and total biomass was studied at different moisture levels and results are presented in Table 5 and 6.

Among the seven soil moisture regimes tested against the dry root rot and its effect on plant root length, the maximum mean root length was recorded in both black and red soil at 100 per cent moisture level (4.97 cm and 5.29 cm) followed by 90 per cent (4.45 cm and

TABLE 6

Effect of soil moisture levels on growth parameters of chickpea in red soil inoculated with *R. bataticola*

Moisture (%)	Growth parameters											
	Root length (cm)				Shoot length (cm)				Total biomass (g)			
	Rb1	Rb2	Rb3	Mean	Rb1	Rb2	Rb3	Mean	Rb1	Rb2	Rb3	Mean
40	1.84	2.00	2.1	1.98	7.50	7.65	7.00	7.30	0.35	0.50	0.25	0.36
50	2.64	2.67	2.63	2.60	9.53	10.89	9.55	10.00	0.66	0.85	0.70	0.73
60	2.60	3.40	3.00	3.00	11.46	11.66	11.38	11.54	0.60	0.64	0.57	0.60
70	3.33	3.60	3.30	3.46	20.76	26.23	18.00	21.66	0.98	1.00	0.83	0.93
80	4.50	4.75	4.56	4.60	27.20	27.24	27.00	27.14	1.39	1.43	1.35	1.39
90	5.00	5.06	4.35	4.80	29.07	29.00	28.50	28.85	3.54	3.45	3.10	3.35
100	5.24	5.85	4.8	5.29	29.33	29.20	29.00	29.13	3.88	3.90	3.90	3.81
Mean	3.60	3.90	3.54	3.67	19.26	20.26	18.63	19.38	1.61	1.68	1.52	1.59

Factors	S.Em±	CD @ 1%
Moisture (M)	0.21	0.70
Isolates (I)	0.26	0.82
M x I	0.32	1.06

4.80 cm) and 80 per cent (4.16 cm and 4.60 cm). The least root length was observed at 40 per cent (2.14 cm and 1.98 cm) soil moisture regime (Table 6).

### Shoot Length

The highest mean shoot length of three isolates (Rb1, Rb2 and Rb3) was observed in red soil (19.38 cm) when compared to the black soil (19.29 cm). Among the three isolates, maximum mean shoot length was recorded Rb2 (19.96 cm and 20.26 cm in black and red soil, respectively) followed by Rb1 (19.13 cm and 19.26 cm) and least was observed in Rb3 (18.79 cm and 18.63 cm) (Table 5 and 6).

With respect to seven soil moisture regimes tested against the dry root rot, the maximum mean shoot length was recorded in both black and red soil at 100 per cent (29.56 cm and 29.13 cm, respectively) followed by 90 per cent (29.48 cm and 28.85 cm) and 80 per cent (25.21 cm and 27.14 cm) when compared to least shoot length of 8.45 cm and 7.30 cm at 40 per cent in both soils (Table 5 and 6).

### Total Biomass

The highest mean total biomass of three isolates (Rb1, Rb2 and Rb3) was observed in red soil (1.59 g) compared to the black soil (1.42 g). Among the three isolates, maximum mean total biomass was in Rb2 (1.89 g and 1.68 g in black and red soils, respectively) followed by Rb1 (1.36 g and 1.61 g) and Rb3 (1.35 g and 1.52 g) (Table 5 and 6).

Among the seven soil moisture regimes, the maximum mean total biomass was recorded at 100 per cent soil moisture (3.82 g and 3.81 g in black and red soils, respectively) followed by 90 per cent (3.45 g and 3.35 g) and 80 per cent (1.32 g and 1.39 g) in comparison to least moisture level of 40 per cent (0.38 g and 0.36 g) (Table 5 and 6).

Mayek *et al.* (2002) reported that the stress prone and infected plants had poor growth compared to healthy and irrigated plants. Drought stress showed higher negative effects coupled with *R. bataticola* which attack in vegetative growth of the plant which decreased leaf area and dry weight of all vegetative structures significantly. Srinivas (2016) studied different soil moisture levels on growth of chickpea and reported that 100 and 90 per cent soil moisture levels recorded the highest root length, shoot length and total dry weight of chickpea plants in as observed in present investigation.

### Effect of Soil Moisture on Disease Incidence

The results indicated that there was significant difference between moisture levels but not with respect to soil types. In black soil the highest disease severity rating was 9.0 grade, 9.0, 8.91, 8.03, 5.62, 4.86 and 3.30 while in red soil, it was 9.0, 9.0, 8.95, 8.33, 5.73, 5.40 and 3.56 at 40, 50, 60, 70, 80, 90 and 100 per cent soil moisture, respectively. The disease severity decreased as the soil moisture increased in both the types of soils (Table 7).

Among the seven levels of moisture content, the disease severity decreased slowly with respect to the increase in soil moisture. At 40 and 50 per cent soil moisture, there was early development of disease symptoms. Yellowing of leaves was started at ten days after sowing and at twenty days after sowing the plants completely dried and root system was completely black. The plants recorded the highest disease severity grade of 9.0 in both the soils at 40 and 50 per cent soil moisture. While in 60 per cent (8.91 and 8.95) and 70 per cent (8.03 and 8.33) soil moisture, the dry root symptoms such as yellowing and upward turning of leaflets was started after 20 days after sowing and recorded the moderate disease severity in both black and red soils. The lesser disease severities were recorded at 100 per cent (3.30 and 3.56) followed by 90 per cent (4.86 and 5.40) and 80 per cent (5.62 and 5.73) soil moisture levels in black soil and red soil, respectively (Table 7).

TABLE 7  
Effect of soil moisture levels on disease severity of dry root rot caused by *R. bataticola* isolates in black soil and red soil

Moisture (%)	Disease severity (1-9 scale)							
	Black soil				Red soil			
	Rb1	Rb2	Rb3	Highest scale	Rb1	Rb2	Rb3	Highest scale
40	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0
50	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0
60	8.90	8.35	8.91	8.91	9.0	8.45	8.95	8.95
70	7.80	6.60	8.03	8.03	8.00	7.82	8.33	8.33
80	5.00	4.87	5.62	5.62	5.25	5.22	5.73	5.73
90	4.55	4.32	4.86	4.86	5.08	4.25	5.40	5.40
100	3.10	2.80	3.30	3.30	3.16	3.00	3.56	3.56

\*Mean of three replications

Factors	S.Em±	CD @ 1%
Soil type (S)	0.20	0.81
Isolate (I)	0.23	0.93
Moisture (M)	0.20	0.42
S x I x M	0.35	1.17

likely to increase. The high negative correlation is observed with -0.94 and -0.93 coefficients values for black and red soil between moisture levels percentage and disease severity. This means that as the moisture level increases, the severity of disease in plants is likely to decrease.

The results (Table 8) on correlation analysis of moisture levels percentage and growth parameters indicated that, the high correlation coefficients of 0.98, 0.95, and 0.88 in black soil and 0.98, 0.98, and 0.90 in red soil suggest a strong positive relationship between the two variables. This means that as the moisture level increases, the growth parameters such as root length, shoot length and total biomass are also

The present investigations are in line with Srinivas (2016) who also observed difference in the dry root rot incidence with change in soil moisture content. The plants grown in 50%, 60% soil moisture recorded highest disease severity grade compared to the 80%, 90% and 100% soil moisture. Further, Sharma and Pande (2013) reported that plants exposed to 40% and 60% soil moisture, dry root rot severity was maximum,

TABLE 8  
Correlation analysis of soil moisture levels on growth parameters of chickpea in black and red soil inoculated with *R. bataticola* and disease severity

Moisture Levels	Type of Soil	Growth Parameters	Correlation Coefficient	Disease severity
Moisture (%)	Black	Root length (cm)	0.97**	-0.94**
		Shoot length (cm)	0.96**	
		Total biomass (g)	0.88**	
	Red	Root length (cm)	0.98**	-0.93**
		Shoot length (cm)	0.95**	
		Total biomass (g)	0.90**	

\*\* Significant at 1% LOS

showed higher mortality as compared to 80 and 100 per cent.

### Effect of Carbon Dioxide Combined with Temperature on Growth Parameters

The study was conducted in the open top chambers with five sets of treatments (carbon dioxide levels) on growth parameters and dry root rot of chickpea and results are presented in Fig 3.

#### Shoot length

Elevated carbon dioxide combined with temperature on phenology of chickpea crop revealed that the highest shoot length of 31.66 cm was observed in open plot followed by 27.66 cm in ambient CO<sub>2</sub> @ 390 ± 25 ppm. These two treatments significantly differed with respect of shoot length in elevated CO<sub>2</sub> @ 550 ± 25 ppm with 2 °C rise in temperature (12.91 cm) followed by elevated CO<sub>2</sub> @ 550 ± 25 ppm alone (20.33 cm) and 24.58 cm in ambient CO<sub>2</sub> @ 390 ± 25 ppm with 2 °C rise in temperature. The results indicated that the carbon dioxide alone and coupled with increased temperature has detrimental impact on shoot length of chickpea plants.

#### Root Length

There was a significant difference between root length of plants in the different carbon dioxide levels tested. Highest root length of 10.41 cm was observed in open plot followed by 7.45 cm in ambient CO<sub>2</sub> @ 390 ± 25 ppm while the least growth of 3.66 cm was observed in elevated CO<sub>2</sub> @ 550 ± 25 ppm with 2 °C rise in temperature. Further, the treatment that is elevated CO<sub>2</sub> @ 550 ± 25 ppm alone recorded root length of 5.30 cm and it was 6.61 cm in ambient CO<sub>2</sub> @ 390 ± 25 ppm with 2 °C rise in temperature. Finally, it is inferred that the elevated carbon dioxide combined with increased temperature of 2 °C resulted in increased the severity of disease and reduced the root length of plants (Fig. 3).

#### Number of Branches

The results indicated that there was a significant difference between number of branches of plants in the different sets of treatments. The maximum number

of branches (17.26) was observed in open plot followed by in ambient CO<sub>2</sub> @ 390 ± 25 ppm (13.33) and ambient CO<sub>2</sub> @ 390 ± 25 ppm with 2 °C rise in temperature (12.55). In contrast to this, there was a reduction in the number of branches in elevated CO<sub>2</sub> @ 550 ± 25 ppm with 2 °C rise in temperature (8.30) followed by elevated CO<sub>2</sub> @ 550 ± 25 ppm alone (0.29) (Fig. 3).

#### Total Biomass

The maximum total biomass (1.43 g) was recorded in open plot followed by 1.07 g in ambient CO<sub>2</sub> @ 390 ± 25 ppm and 0.85 in ambient CO<sub>2</sub> @ 390 ± 25 ppm with 2 °C rise in temperature (Fig. 3). However, the reduced biomass of chickpea was recorded in elevated CO<sub>2</sub> @ 550 ± 25 ppm with 2 °C rise in temperature (0.36 g) followed by elevated CO<sub>2</sub> @ 550 ± 25 ppm alone (0.54 g).

The present investigations on effect of carbon dioxide with temperature levels are supported by Jagadish *et al.* (2007) who reported that reduction in the weight of grains of pigeon pea under increased temperature with high levels of CO<sub>2</sub>. Further, Baker (2004) also studied the effect of elevated CO<sub>2</sub> (700 ppm) under different temperature regimes (24, 28, 32, 36 and 40 °C) and found that there was no increase in grains weight of chickpea under enriched CO<sub>2</sub> combined with high temperature.

#### Severity of Dry Root Rot

The results revealed that at higher concentration of carbon dioxide alone and in combination with increased temperature of 2 °C has aggravated the disease. Among the five treatments, elevated CO<sub>2</sub> @ 550 ± 25 ppm with 2 °C rise in temperature showed higher disease severity (9.00 grade) with early infection showing drying of leaves and wilting of entire plant and roots were completely rotten. This treatment was followed by elevated CO<sub>2</sub> @ 550 ± 25 ppm alone with disease rating of 8.10, here also similar kind of symptoms but expression of symptoms was slow compared to elevated CO<sub>2</sub> @ 550 ± 25 ppm with 2 °C rise in temperature. The least disease severity was observed in open plot

(5.10 grade). Apart from this, the treatment that is Ambient CO<sub>2</sub> @ 390 ± 25 ppm showed moderate disease severity of 5.50 grade and ambient CO<sub>2</sub> @ 390 ± 25 ppm with 2 °C rise in temperature recorded 5.83 grade (Fig. 3).

Similarly, Jagadish *et al.* (2007) studied the effect of carbon dioxide on root rot of pigeon pea grown in elevated CO<sub>2</sub> condition of 550 ± 25 ppm with raised temperature which increased disease incidence in pigeon pea. Further, Chakraborty and Datta (2003) studied the effects of increasing atmospheric CO<sub>2</sub> (increasing CO<sub>2</sub> from 365 ppm to 550 ppm) on plant-pathogen interactions and reported that there was an increased incidence of root rot and reduced yields of pulses in controlled conditions.

It is concluded from the present study that there was a significant impact of temperature, soil moisture and elevated CO<sub>2</sub> on growth of pathogen as well as dry root rot Disease of chickpea grown in different parts of Karnataka State. The temperature of 30-35 °C was optimum for growth of pathogen. The maximum colony growth of *R. bataticola* and the dry root rot disease severity was recorded at 30-35 °C which is considered as optimum temperature range for growth of pathogen and development of disease. Highest severity of dry root rot and lesser plant growth parameters such as root length, shoot length and total biomass were observed at 40-60 per cent soil moisture regimes, irrespective of type of soil. Further, elevated CO<sub>2</sub> @ 550 ± 25 ppm with 2 °C rise in temperature showed higher dry rot severity and reduced growth parameters of chickpea.

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## Seed-inhabiting Endophytic Bacteria of Finger Millet [*Eleusine coracana* (L.) Gaertn] Enhance Early Seedling Growth and Development

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### ABSTRACT

Seed microbiome has a strategic importance as it connects the maternal microbiome with next generation, seedling microbiome. Seed microbiome (bacteria & fungi) can be viewed as the transfer of microbial memory of life history adaptations to the next generation. Against this background, present study was taken up to study the role of seed inhabiting bacterial endophytes on finger millet seedling growth and development. Two bacterial endophytes were isolated from surface-sterilized seeds of finger millet and molecularly identified by 16S rRNA sequencing as *Paenibacillus quercus* (EC1) and *Bacillus cereus* (EC2). Enrichment of seed bacterial isolates enhanced seedling growth while removal of the endophytic bacteria from finger millet seeds compromised seedling growth and development. In summary our studies indicated that seed borne endophytic bacteria are crucial for early seedling establishment.

*Keywords:* Seed, Endophytes, Finger millet, Seedling establishment

IN recent years, increasing attention has been paid to endophytes and the role they play in modulating plant growth and development. Both, endophytic fungi and bacteria seem to enjoy a unique relationship with the plant that range from mutualism to symbiosis. In fact, their interactions with the plant are nested so deeply and intricately, that often the endophytes and other plant associated microbiome is also referred to as 'second genome' of the plant (Berendsen *et al.*, 2012). Their colonization does not cause any apparent symptoms and neither are they rejected by the plants.

Endophytes have been reported to promote plant fitness, either by directly enhancing nutrient availability, production of phytohormones, organic acids or indirectly by preventing the growth or activity of phytopathogen (Manasa *et al.*, 2015). The microbiome spans the entire plant, ranging from the rhizosphere to the phyllosphere, including the seed. While abundant literature exists to document the

nature of microbiome in rhizosphere and phyllosphere, relatively a little is known about the seed microbiome and how these might help in seedling establishment.

Seed borne endophytes have been reported in different crops such as rice, wheat, maize and millets (Verma *et al.*, 2018; Johnston-Monje & Manish Raizada, 2011; Herrera *et al.*, 2016 and Kumar *et al.*, 2021). It is estimated that over 9000 diverse microbial species inhabit the seeds (Shade *et al.*, 2017 and Adam *et al.*, 2018). Principally, seeds acquire their microbiota through multiple paths, including through the xylem or nonvascular tissue of the mother plant, the floral pathway *via* the stigma of the mother or simply by contact in the soil (Barret *et al.*, 2016). While the former two involve mainly vertical transmission from seed to seed, the latter clearly is a case of horizontal transfer. Seed inhabiting endophytes are also known to stimulate the expression of various genes in developing seedling of crop plants related to root



architecture development and defense against biotic and abiotic stresses (Gogna *et al.*, 2015 and Tasmiya *et al.*, 2021).

Against this background, in this study an attempt has been made to explore the possible role of seed inhabited bacterial endophytes in the establishment of finger millet seedlings. We hypothesized that finger millet seeds may carry crucial bacteria that improve seedling growth and development. Our finding suggests that seeds of finger millet harbour important bacterial endophytes, *Paenibacillus quercus* (EC 1) and *Bacillus cereus* (EC 2); enrichment of these bacteria enhanced seedling growth while removal of these bacteria from seeds hampered seedling growth and development.

## MATERIAL AND METHODS

### Seed Material

*Eleusine coracana* (L.) Gaertn, an allotetraploid ( $2n = 4X = 36$ ), is an annual robust grass belonging to family Poaceae, widely grown as a millet in the semi-arid tropics and subtropics of the world under rainfed conditions. *Eleusine coracana* (L.) Gaertn was domesticated around 5000 years ago in western Uganda and the Ethiopian highlands. Later, finger millet was introduced into Western Ghats of India around 3000 BC. Thus, India became the secondary centre of diversity for finger millet. For this study, seeds of *Eleusine coracana*, GPU28 accession was procured from the Department of Crop Physiology, University of Agricultural Sciences, Bangalore, GKVK, Bengaluru - 65, Karnataka and India.

### Isolation of Endophytes from Seeds of *Eleusine coracana*

Seeds of finger millet species, GPU28 were soaked in autoclaved water for 24h. Seeds were thoroughly washed with distilled water, surface sterilized with 70 per cent (v/v) ethanol for 3 min followed by 1 per cent (v/v) sodium hypochlorite for 3 min and 70 per cent (v/v) ethanol for 3min and then washed with sterile double distilled water thrice to remove sterilization solution (Arnold *et al.*, 2000). Surface sterilized seeds were cut into two halves and were

placed onto Potato Dextrose Agar (PDA), Czapek Dok, Malt extract agar amended with streptomycin for fungal colonies emergence while Nutrient Agar (NA) media, LB agar, Trypticase soy agar for bacterial colonies emergence, respectively (Taylor *et al.*, 1999 and Parthasarathy & Sathiyabama (2014). To check for the effectiveness of sterilization, surface impressions of the seed segments were made on media and incubated at  $28 \pm 2$  °C. The plates were monitored for endophytes growth.

### Morphological and Molecular Characterization of Bacterial Endophytes

Purified bacterial isolates were classified into operational taxonomic units (OTUs) based on shape / form, texture, margin, elevation, pigmentation. Voucher numbers were assigned to all the bacterial OTUs and deposited in the library collection at the School of Ecology and Conservation laboratory, University of Agricultural Sciences, GKVK, Bangalore.

For molecular characterization, the genomic DNA of the bacterial endophytes were extracted by alkaline lysis method (Sambrook *et al.*, 1989). The bacteria isolated were cultured individually in NB broth for 24 h at 30 °C. The cells were pelleted by centrifuging at 12,000 rpm for 2 min. The pellet was re-suspended in 650 µl of extraction buffer (100 mM Tris-HCl, 100 mM EDTA, 250 mM NaCl and pH 8.2) and incubated in water bath at 65 °C for 30 min. To the extract, 100 µl of 5 M potassium acetate solution was added and placed on ice for 10 min for precipitation of protein and carbohydrates. The supernatant was collected by centrifuging at 12,000 rpm for 8 min. The DNA was precipitated by adding equal volume of chilled isopropanol for 2 h at -20 °C and then was centrifuged at 12,000 rpm for 5 min. The pellet obtained was washed with 70 per cent chilled ethanol, air dried and dissolved in 20 µl of sterile distilled water. The RNA present in the samples were removed by treating the sample with 3 µl of RNAase at 37 °C for 1h. The DNA was fractionated on 1 per cent (w/v) agarose and visualized following staining with ethidium bromide and quantified using Nano Drop V3.2.1.

Further, the DNA was amplified using 16S rRNA primers (16SF: 5' GTTAGATCTTGGC TCAGGACGAACGC 3') and (16SR: 5' GATCCAGCCGCACCTTCCGATACG 3'). The PCR amplification was carried out by using Eppendorf Master cycler; the total volume of PCR reaction mixtures was 20 µl containing 2.0 µl of 1X PCR Taq. Buffer with MgCl<sub>2</sub> (1.5 mM), 2.0 µl of 10 mM dNTP's mix, 1.0 µl of 16S rRNA primers (forward and reverse, 0.5 µl each), 0.3 µl of Taq DNA Polymerase (1U Genei Bengaluru), 1.0 µl of Template DNA (~50 ng/ µl) and 13.7 µl of sterile distilled water. The PCR conditions used for amplifications were initial denaturation at 94 °C for 5 min followed by 35 cycles of 94 °C for 1 min, 58 °C for 30s, 72 °C for 1 min, and a final extension at 72 °C for 10 min. After PCR amplification, PCR products were visualized on 1 per cent agarose gel with 1X TAE buffer. Purification and sequencing of amplified PCR products were done by Sanger sequencing (AgriGenome Labs Pvt. Ltd., Cochin, Kerala). The sequences were queried in the Basic Local Alignment Search Tool (BLAST) in the NCBI GenBank database (www.ncbi.nlm.nih.gov). The sequence with the highest homology and with a maximum query coverage and maximum score was used to ascertain the identity of the endophytic bacteria. A phylogenetic tree was constructed for each of the isolates following MEGA version X.

### Enrichment of Pre-germinated Seeds with Bacterial Endophytes

Surface sterilized seeds were allowed to pre-germinate (protruding of shoot and root) and uniformly pre-germinated seeds were taken, and treated with 5 ml (10<sup>6</sup> - 10<sup>8</sup> cells/ml) of *Paenibacillus quercus* (EC1) and *Bacillus cereus* (EC2) and consortium of two bacterial endophytes for 3 h. Another set of uniformly pre-germinated seeds were treated with sterile water which served as a control. Both, endophyte enriched and non-enriched pre-germinated seeds were transferred to germination paper. A total of 30 seedlings were used in each treatment, with three replications (each with 10 seedlings). Seedling growth rate was assessed every other day of treatment till 7<sup>th</sup> day.

### Standardization of Antibiotic Concentration

An antibiotic, streptomycin sulphate was used to assess the role of bacterial endophytes in seedling growth and development. Surface sterilized seeds were treated with different concentration of streptomycin sulphate (10, 25 and 50 µg/ml) for 24 h with constant shaking at 120 rpm. Seeds soaked in autoclaved water served as a control. To check the complete disinfection / removal of native bacterial endophytes, streptomycin treated seeds were plotted on nutrient agar plate and observed for 5 days. No bacterial growth around seeds confirmed the complete disinfection.

After 24 h of treatment, seeds were washed thrice with sterile distilled water to remove the traces of streptomycin sulphate. Treated and control seeds were placed on plastic pots (4 inch) containing coir peat and allowed for germination and growth at room temperature for 10 days. After 10 days of treatment seedling height (shoot and root length) was measured. The experiments were conducted in triplicates with 25 seedlings for each replication.

### Statistical Analysis

All experiments were conducted with a minimum of three replications. One way analysis of variance (ANOVA) was performed to statistically validate the results obtained. The means of various treatments were compared using a Duncan's new multiple range test (MRT) test at 95 per cent confidence interval. All statistical analysis was done in MS-Excel and SPSS software.

## RESULTS AND DISCUSSION

### Isolation and Identification of Endophytes from Seeds of Finger Millet

Surface sterilized finger millet seeds were cut into two halves and inoculated onto six different media (Potato Dextrose Agar, Czapek Dok, Malt extract agar amended with streptomycin sulphate for fungal and Nutrient Agar media, LB agar, Trypticase soy agar for bacterial colonies emergence, respectively). Surprisingly, finger millet seeds did not yield any

fungal colonies. These results are intriguing and are indeed supported by an independent study by Mousa *et al.* (2015) on finger millet. The exact reasons for the absence of culturable-fungal endophytes in the seed are not clear and at best can only be conjectured. Among them are a) some structural constraints offered by the seed coat for entry of fungal mycelia into intercellular spaces of seed coat tissue. Indeed, McDonough and Rooney (1986) reviewed the structure of seeds of major millets. He suggested that finger (*Eleusine coracana* (L.) Gaertn.), proso [*Panicum miliaceum* (L.)] and foxtail (*Setaria italica* (L.) P. Beauv.) millets are utricles. In an utricle, the seed is covered by the membranous pericarp, which is not fused with seed coat. The entire utricle is further enclosed by chaff-like bracts called lemma and palea and b) inhibition of fungal entry and growth by endophytic bacteria resident in the seed coat. Further studies would be required to unravel these possibilities.

However, bacterial colonies were obtained from most of the seeds. A total of two bacterial isolates were morphologically grouped as EC 1 and EC 2. Further these bacterial isolates were molecularly identified using 16S rRNA sequencing as *Paenibacillus quercus* (EC 1) and *Bacillus cereus* (EC 2) (Table 1). Phylogenetic relationship of the sequences to closest matches in public database, based on 16S rRNA gene sequences was constructed by using neighbour-joining method in MEGA version X environment. The reliability of the relationships of lineages on the inferred trees was tested by bootstrap analysis for 1000 replicates (Fig. 1). Genera *Paenibacillus* and *Bacillus* belongs to the phylum, firmicutes. Recent past studies showed that *Paenibacillus* and *Bacillus* are the most common

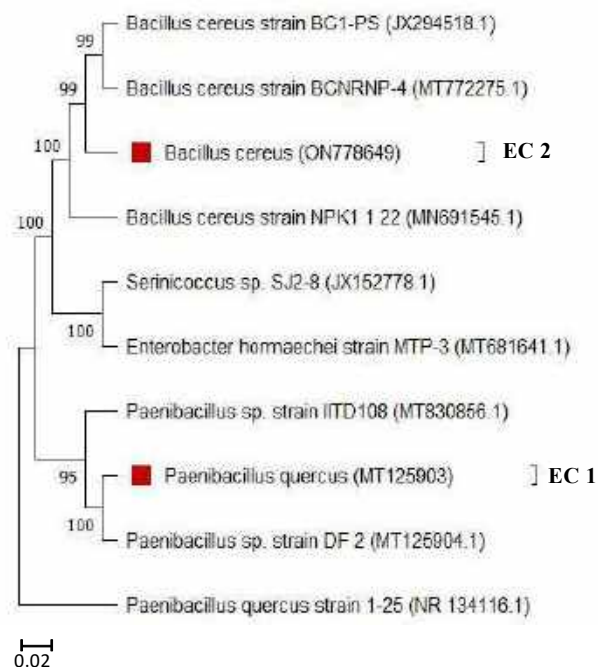


Fig. 1: Neighbor-joining phylogenetic tree showing the relationships of the bacterial isolates from seeds of finger millets. Support bootstrap values are shown in the branches. Scale bar indicates 0.02 substitutions per nucleotide position

seed associated endophytes (Prasannakumar *et al.*, 2020 and Kumar *et al.*, 2020).

### Effect of Seed Bacterial Endophytes on Finger Millet early Seedling Growth and Development

To check the growth promotion ability of seed bacterial endophytes, uniformly pre-germinated finger millet seeds were treated with 5ml of endophytic bacterial suspension and on the other hand, seeds treated with water served as a control. Endophytes enriched seeds showed relatively higher shoot and root length compared to the control at every stage of development (Fig. 2). However, seeds enriched with

TABLE 1  
Molecular identification of seed bacterial endophytes of finger millet using 16S rRNA

Plant species	Bacterial isolate	Sequence Length (bp)	Best Blast search	Query cover (%)	Identity (%)	Gen Bank accession number
<i>E. coracana</i> (GPU28)	EC 1	879	<i>Paenibacillus quercus</i>	100	95.80	MT125903
	EC 2	944	<i>Bacillus cereus</i>	100	100	ON778649

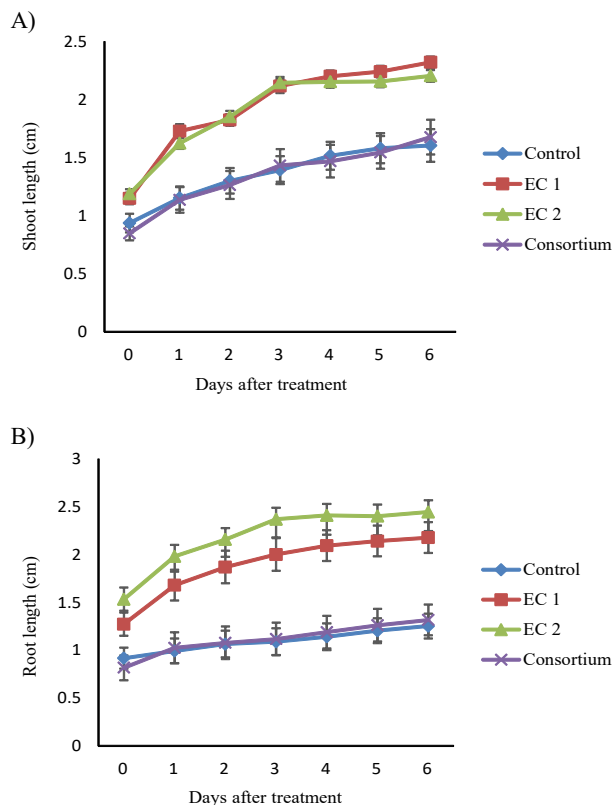


Fig. 2: Effect of bacterial endophytes enrichment in the early seedling growth and development of finger millet. A) shoot and B) root length of finger millet seedling days after treatment. Values represents mean  $\pm$  SE. (EC1 - *Paenibacillus quercus*, EC2 - *Bacillus cereus* and consortium -EC1 + EC2)

consortium of EC 1 and EC 2 showed shoot and root length on par with control (Fig. 2 and Fig. 3). *Paenibacillus* sp. (EC 1) and *Bacillus* sp. (EC 2) are

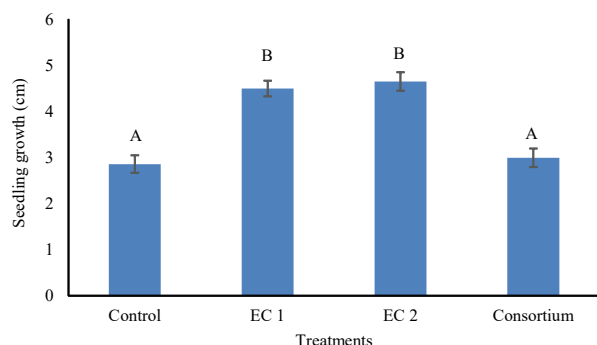


Fig. 3: Growth rate of finger millet seedling after seven days of treatment. Values represents mean  $\pm$  SE and different letters indicate significant differences (P < 0.05) based on Duncan's test. (EC1 - *Paenibacillus quercus*, EC2 - *Bacillus cereus* and consortium -EC1 + EC2)

the most common cultivable bacterial genus and have been frequently recorded as seed associated microbes and they also help in the seedling establishment (Kumar *et al.*, 2020)

### Standardization of Antibiotic Concentration

One of the challenges in analysing the role of microbiome in seedling growth is to obtain seeds that are free of microbiome. Unfortunately, most methods of culture, save aseptic culture of plants, do not preclude microbiome contaminations from the ambient environment. Under these circumstances, use of an antibiotics could help cleanse the tissue of the microbiome. In this study, an antibiotic, streptomycin sulphate was used to cleanse the seed tissues of their bacterial endophytes. Streptomycin sulphate (an amino glycoside antibiotic), a protein synthesis inhibitor, binds to 30S subunit of the bacterial ribosome and interferes with the binding of formyl-methionyl-tRNA to the 30S subunit (Zhu *et al.*, 2001).

To comply to this requirement, an experiment was conducted to evaluate and arrive at an appropriate concentration of streptomycin that could be used to treat the finger millet seeds to cleanse them of the bacterial endophytes. Surface sterilized seeds of finger millet were incubated at different concentrations of streptomycin sulphate (10, 25 and

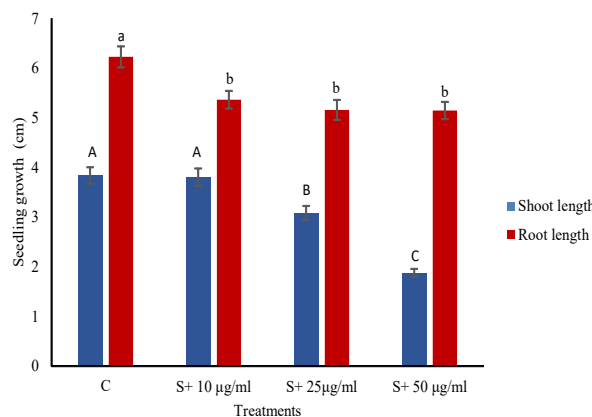


Fig. 4: Shoot & root length of ragi seedling after treating seeds with different concentration of antibiotics for an 24hrs. Values represents mean  $\pm$  SE and different letters indicate significant differences (P < 0.05) based on Duncan's test. (S+ indicate streptomycin sulphate).

50  $\mu\text{g/ml}$ ) for 24 h. The seeds were allowed to grow and their root and shoot lengths (cm) were measured after ten days.

There was a significant difference in the shoot and root length of 50  $\mu\text{g/ml}$  streptomycin sulphate treated seedlings compared to the control (Fig. 4). We found that seed treatment with 50  $\mu\text{g/ml}$  concentration of streptomycin sulphate for 24 h is enough to remove bacterial endophytes, since no bacterial colonies were emerged on Nutrient agar media (Fig. 5). We also observed that seedlings developed from antibiotic treated seedlings were found weak compared to the control as visible in (Fig. 6). Further, the frequency

distribution of shoot and root length of seeds treated with S<sup>+</sup> 50  $\mu\text{g/ml}$  was skewed to shorter length compared to the control (Fig. 7). A recent study by Verma and White (2018) also showed that elimination of bacteria from seeds compromises seedling growth and development.

The present study demonstrates that finger millet seeds harbour functionally important endophytic bacteria, which are crucial for early seedling establishment. This study suggests that the microbiome associated with seeds can be used as a bio-inoculant for sustainable agriculture, but it has to be tested in field conditions. Further research has to

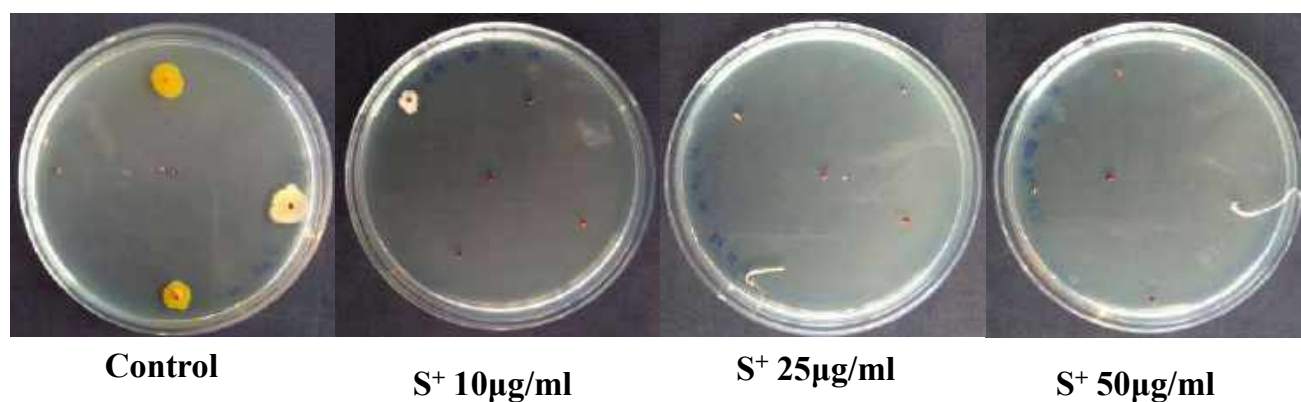


Fig. 5: Absence of bacterial endophytes in the seeds treated with S<sup>+</sup> 50 $\mu\text{g/ml}$ . (S<sup>+</sup> indicate streptomycin sulphate).



Fig. 6: Effect of antibiotic (S<sup>+</sup> indicate streptomycin sulphate) treatment on seedling growth at 10 days

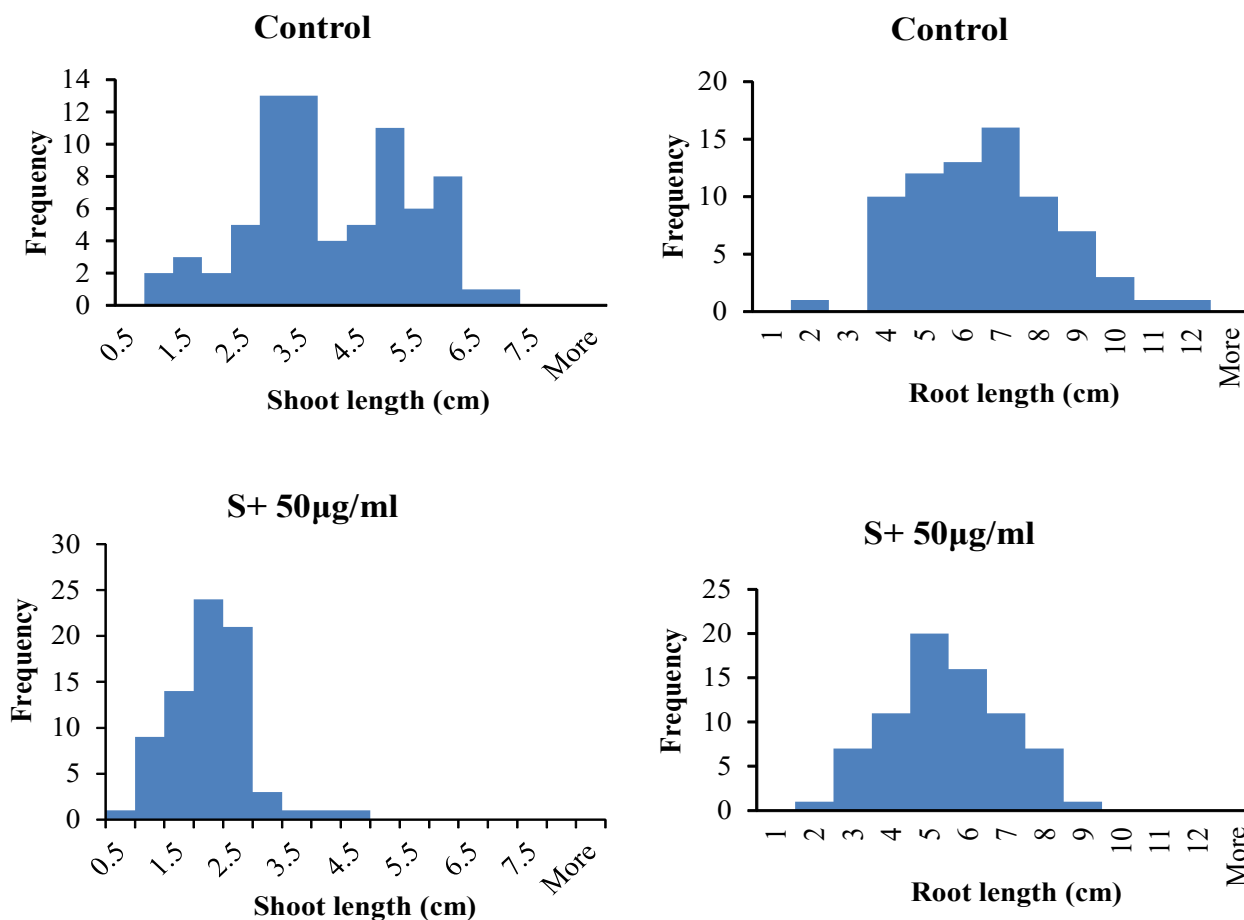


Fig. 7: Frequency distribution of shoot and root length of 10 days old seedlings raised from control and streptomycin sulphate (50µg/ml) treated seeds

be done to find out the possible mechanisms by which endophytes help seedling growth and development.

**Acknowledgments :** The authors thank the Indian Council of Agricultural Research (ICAR) - Centre for Advanced Agricultural Science and Technology (CAAST), Activity 1c - 'Next generation technologies for microbiome enabled seed priming' (ICAR\_NAHEP; F. No./NAHEP/CAAST/2018-19; AB/AC7703) for support. M.H. would like to thank the Council of Scientific and Industrial Research (CSIR), Government of India for providing research fellowship.

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## Standardization of Process for Chia Germination

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### ABSTRACT

Chia seed stands in the front list of super foods, because of its unique nutritional properties and health benefits. Chia seeds are rich in omega 3 fatty acids, protein, fibre and minerals. In the current study, germination of white and black chia seeds was carried out. Raw chia seeds were sprinkled with water, allowed to stand at room temperature, incubated at 24 °C for time duration of 18, 24, 36, 48 and 54 hours with distilled water, warm distilled water at 45 °C and 2 per cent sugar solution with warm distilled water at 45 °C. The highest germination percentage of the treatments were selected for analysis of  $\alpha$  amylase and vitamin C. The highest content of  $\alpha$  amylase, vitamin C of specific time interval and sensory evaluation were carried out. The result revealed that for different time durations, the treatment with distilled water (T1) for white chia and with warm distilled water (T2) for black chia seeds were found to give highest germination percentage and both with T1 were highly accepted for sensory evaluation. Amylase activity was found to be highest at 36 hour's time duration having 1.24 and 1.20 mg/100g for white and black variety, respectively. Vitamin C content was highest for white and black variety at 36 hours having 80.67 and 66.67 mg/100g, respectively. Findings concluded that germination of chia enhanced  $\alpha$  amylase and vitamin C content.

Keywords : Chia seeds, Germination, Vitamin C,  $\alpha$  - amylase

THE search for novel, high quality source of protein, fat, dietary fibre and antioxidant property has been attaining popularity in developing countries for meeting the challenges of malnutrition on one side and prevention and control of non-communicable diseases through diet on the other side. All the super foods are gaining popularity due to their nutraceutical properties (Din *et al.*, 2021). Millet and chia are gluten free, have low glycemic index and chia mucilage can be used as fat replacement (Hiregoudar and Mamatha, 2021). Out of 900 species of genus *Salvia*, only *Salvia hispanica* can be grown domestically (Chaitanya *et al.*, 2022).

Due to increased consumer awareness on healthy food, chia, quinoa and millets are in demand. Quinoa and millets are explored for value added products, however not much of the products available using chia seeds. Chia is having lot of health benefits and

gaining sufficient attention from consumers. To meet the demand, more value-added products from high nutrient rich foods like chia is gaining popularity. Due to climatic change in present days, chia is suitable as it can grow in adverse weather and low nutrient content soils. The use of chia in diet can be considered beneficial in the prevention and treatment of risk factors related to life style diseases such as diabetes and cardiovascular diseases, which are leading cause for fatality. Therefore, crops with multi utility, prominent nutrient composition and user-friendly processing methods are needed (Kilewela *et al.*, 2021).

Germination is an inexpensive and effective method for improving the overall nutritional quality of any grains as it enhances the digestibility and reduces anti nutritional factors (Chavan and Kadam, 1989 and Ghorpade and Kadam, 1989). Germination increases the total phenolic content including  $\gamma$ -aminobutyric acid, the protein and insoluble dietary fibre increase,

whereas soluble dietary fibres and lipid component decreases (Nadtochii *et al.*, 2019). The data available on different processing techniques used for super food is scanty. Keeping this in the background, present study was undertaken with the aim of standardization of process protocol for chia seeds germination.

### MATERIAL AND METHODS

White and black seeded chia were procured from Kilaru Naturals, Hyderabad, Andhra Pradesh, India.

**Germination of chia seeds** : The germination of chia seeds are shown in flow chart (Fig. 1).

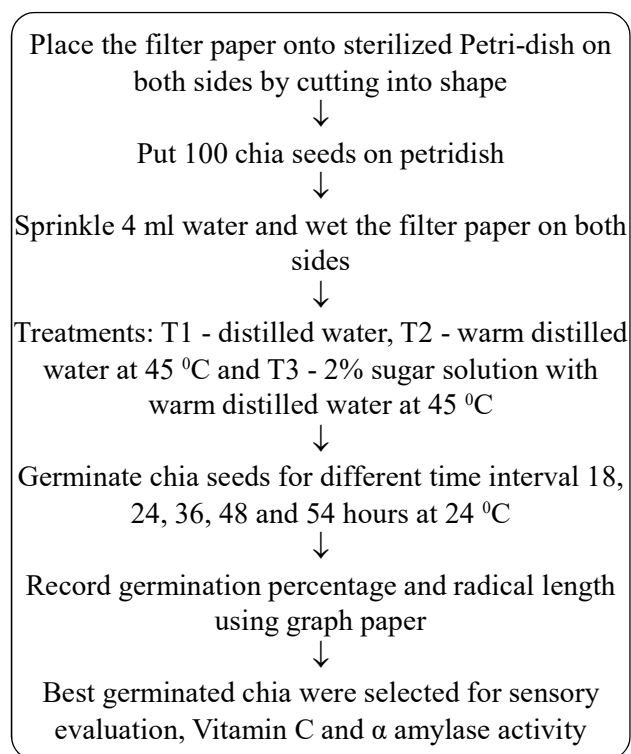


Fig. 1: Flow chart of germination of chia seeds

**Estimation of vitamin C content during germination** : The method for determination of ascorbic acid was used with slight modification given by Harris and Ray (1935) used for germinated chia samples. Ascorbic acid reduced 2, 6-dichlorophenol indophenol dye to a colourless leucobase and gets oxidised to dehydro ascorbic acid changing the dye colour to pink.

**Estimation of  $\alpha$  amylase activity during germination of chia** : The  $\alpha$ -amylase activity was assayed using the method of Bernfeld (1955) for germinated chia.



Plate 1: Germinated white chia



Plate 2: Germinated black chia

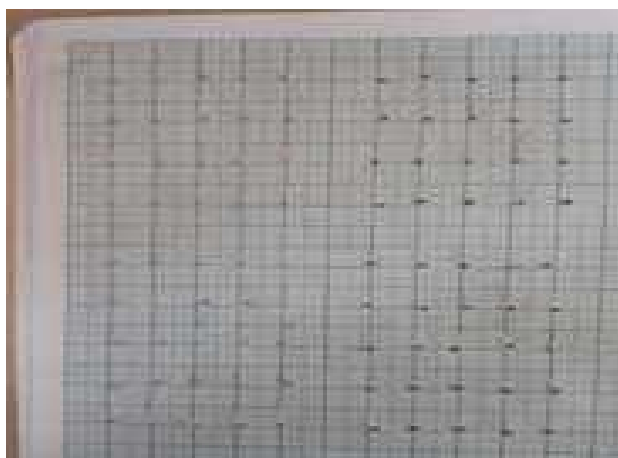


Plate 3: Measurement of length of radical on graph paper

### Sensory Analysis

Sensory analysis of germinated chia was carried out by twenty-one semi-trained panelists using 9-point hedonic scale and scores were recorded for appearance, colour, texture, taste, after-taste, flavour and overall acceptability (Meilgaard *et al.*, 1999).

### Statistical Analysis

The data was subjected to analysis of variance (ANOVA) for testing the significance of variation in



Plate 4: Different treatments of chia germination



Plate 5: Sensory evaluation of germinated chia seeds

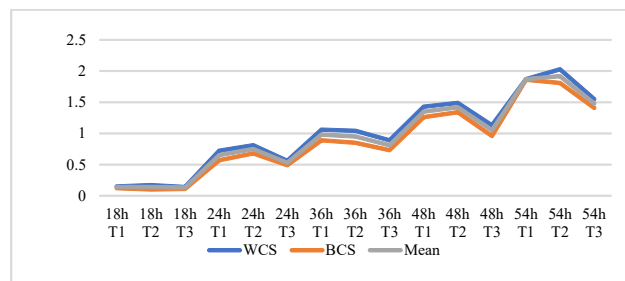


Fig. 2 : Length of radical (cm) of germinated Chia seeds

Germinated at different time interval 18h, 24h, 36h, 48h and 54h (h: hour) and three treatments (T1: Distilled water, T2: Warm distilled water at 45 °C and T3: 2 per cent sugar solution with warm distilled water at 45 °C)

WCS: White chia seeds; BCS: Black chia seeds

germination percentage, vitamin C,  $\alpha$  amylase and sensory evaluation. Mean values were calculated and compared at 5 per cent significance level, one way factor and multiple comparisons were determined using the software OPSTAT (CCS, Haryana Agricultural University, Hisar, India).

## RESULTS AND DISCUSSION

### Germination Percentage of Chia Seeds

Germination is the development of a plant from a seed or spore after a period of dormancy. Among the factors affecting seed germination, substrate and water availability stand out because of their direct influence. In the germination test, an appropriate substrate must be used to provide sufficient amounts of water for soaking the seeds, besides serving as a support for germinated seedlings (Nadtochii *et al.*, 2020). Thus, the choice of the substrate type should consider the size of the seed, its requirement in terms of water, light sensitivity and substrate for the development and evaluation of seedlings (Paiva, 2016). In this present investigation (Table 1), white chia seeds and black chia seeds were germinated at different time intervals - 18h, 24h, 36h, 48h and 54h with three different treatments: T1- distilled water, T2- warm distilled water at 45 °C and T3- 2 per cent sugar solution with warm distilled water at 45 °C. The results from the table 1 revealed that germination percentage of white and black chia seeds at different time interval showed significant difference. 18h, 24h and 54h was found to be significantly higher in

TABLE 1  
Germination percentage of Chia seeds

Treatments	WCS	BCS
18hT1	64.67 ± 0.34 <sup>j</sup>	36.67 ± 0.33 <sup>k</sup>
18hT2	60.67 ± 0.67 <sup>k</sup>	30.33 ± 0.33 <sup>l</sup>
18hT3	41.67 ± 0.89 <sup>l</sup>	27.33 ± 0.33 <sup>m</sup>
24hT1	79.67 ± 0.33 <sup>g</sup>	50.33 ± 0.33 <sup>j</sup>
24hT2	69.33 ± 0.67 <sup>h</sup>	58.67 ± 0.33 <sup>b</sup>
24hT3	67.67 ± 0.33 <sup>i</sup>	53.67 ± 0.33 <sup>i</sup>
36hT1	92.33 ± 0.33 <sup>e</sup>	77.67 ± 0.34 <sup>f</sup>
36hT2	96.67 ± 0.34 <sup>bc</sup>	79.33 ± 0.33 <sup>e</sup>
36hT3	90.00 ± 0.00 <sup>f</sup>	73.33 ± 0.33 <sup>g</sup>
48hT1	96.33 ± 0.33 <sup>bcd</sup>	81.67 ± 0.34 <sup>d</sup>
48hT2	96.67 ± 0.34 <sup>bc</sup>	87.67 ± 0.34 <sup>a</sup>
48hT3	95.00 ± 0.57 <sup>d</sup>	83.33 ± 0.33 <sup>c</sup>
54hT1	98.33 ± 0.33 <sup>a</sup>	87.33 ± 0.33 <sup>a</sup>
54hT2	97.33 ± 03 <sup>ab</sup>	87.33 ± 0.33 <sup>a</sup>
54hT3	95.67 ± 0.67 <sup>cd</sup>	86.33 ± 0.33 <sup>b</sup>
Mean ± SD	82.77 ± 0.69	66.73 ± 0.31
F Test	*	*
C.D. 5%	1.440	0.656
SE(m)	0.494	0.225
C.V.	1.034	0.585

Note: Values are expressed as mean ± SD of three determinations. Germinated at different time interval- 18h, 24h, 36h, 42h and 54h (h: hour)

WCS: White chia seeds; BCS: Black chia seeds; Three treatments; T1: Distilled water; T2: Warm distilled water at 45 °C and T3: 2 per cent sugar solution with warm distilled water at 45 °C.

treatment 1 *i.e.* distilled water followed by T2 *i.e.* warm distilled water for 36 and 48 hours in white chia. With reference to black chia seeds, there was increasing trend of germination percent from 24 to 54 hours in T2 with warm distilled water has given at 24h (58.67), 36h (79.33), 48h (87.67) and 54h (87.33).

The difference in germination percentage between white and black chia seeds may be due to higher percent of anti-nutritional factors in black chia might have hindered the germination at early stage and reached maximum at 54 hours. Therefore, black chia seeds need more time duration to germinate as against to white chia seeds. They also exhibited best at warm distilled water T2. Germination showed

TABLE 2  
Vitamin C (mg/100g) of germinated chia seeds

Sample	0h	18h	24h	36h	48h	54h
GWCS	50.67 ± 0.67	53.33 ± 0.67	66.67 ± 0.67	80.67 ± 0.67	66.67 ± 0.67	52.67 ± 0.67
GBCS	39.33 ± 0.67	40.67 ± 0.67	52.67 ± 0.67	66.67 ± 0.67	52.67 ± 0.67	40.67 ± 0.67
Mean ± SD	45 ± 0.94	47 ± 0.94	59.67 ± 0.94	73.67 ± 0.94	59.67 ± 0.94	46.67 ± 0.94
F Test	*	*	*	*	*	*
C.D. 5%	2.688	2.688	2.688	2.688	2.688	2.688
SE(m)	0.667	0.667	0.667	0.667	0.667	0.667
C.V.	2.566	2.457	1.935	1.567	1.935	2.474

Note: Values are expressed as mean ± SD of three determinations.

GWCS: Germinated white chia seeds; GBCS: Germinated black chia seeds

The highest per cent germination treatment was selected for vitamin C content estimation at different time interval.

more effective result in reducing trypsin inhibitor activity, tannin, polyphenols and phytic acid than other cooking treatments (Ramakrishna *et al.*, 2006).

### Length of Radical

The length of radical was measured with the help of graph paper. The length of radical increased with increase in time interval (Fig. 2).

### Vitamin C Content

Vitamin C is a natural antioxidant, has immense benefits. Germination usually increases the vitamin C content of the grains. The vitamin C content was studied for different time interval of white and black chia (Table 2). The vitamin C content of white chia at 0h, 18h, 24h, 36h, 48h and 54h were 50.67, 53.33, 66.67, 80.67, 66.67 and 52.67 mg/100g respectively. The first three time interval *i.e.* 0h, 18h and 24h, vitamin C content gradually increased. At 36h, it reached the peak point and then decreased at 48 and 54h. Vitamin C content of germinated white chia seeds was found maximum at 36h (80.67 mg/100g). Similarly, for black chia, vitamin C content at different time - 0h, 18h, 24h, 36h, 48h and 54h were 39.33, 40.67, 52.67, 66.67, 52.67 and 40.67 mg/100g respectively. The vitamin C content were maximum for germinated black chia seeds at 36h (66.67 mg/100g). The results of statistical analysis revealed non-significant difference. Srujana *et al.* (2019) studied on germinated quinoa effect on vitamin C

which range from 4.21 to 78.26 mg/100g at different time interval 4 to 60 hours. From the vitamin C content analysis, it is evident that 36h is the best to get maximum vitamin C than the rest of the time duration. Silva *et al.* (2020) studied on germination of soybean and found that vitamin C content of germinated soybean is 61 per cent higher than non germinated soybean.

### $\alpha$ - Amylase Content

The amylases are commonly distributed throughout the plant, but abundantly in the germ and pericarp of the grains. During germination, amylases migrate to regions that are rich in starch, proteins and lipids where they can initiate hydrolytic processes to generate energy (Delcour and Hosney, 2000).

$\alpha$  amylase on germination are activated and start to break down starch into small sugars making it more digestible (Helland *et al.*, 2002). So, the germinated products are suitable for weaning food and for geriatric people. The  $\alpha$ - amylase content was observed at different time interval for white and black chia (Table 3). The white chia contains  $\alpha$ - amylase content at 0h, 18h, 24h, 36h, 48h and 54h were 1.06, 1.08, 1.22, 1.24, 1.20 and 1.19 respectively and for black chia were 1.01, 1.04, 1.19, 1.20, 1.18 and 1.14 mg/100g respectively. The  $\alpha$ -amylase content was highest at 36h for both for germinated

TABLE 3  
 $\alpha$  Amylase (mg/100g) content of germinated chia seeds

Sample	0h	18h	24h	36h	48h	54h
GWCS	1.06	1.08	1.22	1.24	1.20	1.19
GBCS	1.01	1.04	1.19	1.20	1.18	1.14
Mean $\pm$ SD	1.03	1.06	1.20	1.22	1.19	1.16
F Test	*	*	*	*	*	*
C.D. 5%	0.021	0.013	0.013	0.013	0.013	0.013
SE(m)	0.005	0.003	0.003	0.003	0.003	0.003
C.V.	0.882	0.545	0.479	0.472	0.485	0.494

Note: Values are expressed as mean of three determinations  
 GWCS: Germinated white chia seeds; GBCS: Germinated black chia seeds  
 The highest germinated treatment were selected for  $\alpha$  amylase content at different time interval

TABLE 4  
 Sensory evaluation of processed chia seeds (Germination)

Treatment	Appearance	Colour	Texture	Taste	After-Taste	Flavour	Overall-Acceptability
WCS	7.39 $\pm$ 0.19 <sup>b</sup>	7.40 $\pm$ 0.19 <sup>b</sup>	7.14 $\pm$ 0.18 <sup>c</sup>	6.85 $\pm$ 0.19 <sup>c</sup>	6.92 $\pm$ 0.18 <sup>c</sup>	6.96 $\pm$ 0.19 <sup>c</sup>	7.11 $\pm$ 0.16 <sup>b</sup>
BCS	7.19 $\pm$ 0.24 <sup>b</sup>	7.23 $\pm$ 0.27 <sup>b</sup>	6.98 $\pm$ 0.21 <sup>c</sup>	6.92 $\pm$ 0.19 <sup>c</sup>	6.86 $\pm$ 0.19 <sup>c</sup>	7.02 $\pm$ 0.19 <sup>c</sup>	7.03 $\pm$ 0.19 <sup>b</sup>
GWC1	7.92 $\pm$ 0.13 <sup>a</sup>	8.02 $\pm$ 0.12 <sup>a</sup>	7.79 $\pm$ 0.12 <sup>ab</sup>	7.71 $\pm$ 0.16 <sup>ab</sup>	7.59 $\pm$ 0.13 <sup>ab</sup>	7.69 $\pm$ 0.14 <sup>ab</sup>	7.79 $\pm$ 0.09 <sup>a</sup>
GWC2	7.71 $\pm$ 0.12 <sup>ab</sup>	7.64 $\pm$ 0.13 <sup>ab</sup>	7.19 $\pm$ 0.17 <sup>c</sup>	7.29 $\pm$ 0.18 <sup>bc</sup>	7.19 $\pm$ 0.19 <sup>bc</sup>	7.09 $\pm$ 0.17 <sup>c</sup>	7.36 $\pm$ 0.12 <sup>b</sup>
GWC3	7.52 $\pm$ 0.17 <sup>ab</sup>	7.50 $\pm$ 0.17 <sup>ab</sup>	7.00 $\pm$ 0.16 <sup>c</sup>	7.00 $\pm$ 0.17 <sup>c</sup>	6.98 $\pm$ 0.20 <sup>c</sup>	7.23 $\pm$ 0.19 <sup>bc</sup>	7.20 $\pm$ 0.14 <sup>b</sup>
GBC1	7.97 $\pm$ 0.13 <sup>a</sup>	8.02 $\pm$ 0.12 <sup>a</sup>	7.92 $\pm$ 0.12 <sup>a</sup>	7.98 $\pm$ 0.12 <sup>a</sup>	7.96 $\pm$ 0.11 <sup>a</sup>	7.85 $\pm$ 0.12 <sup>a</sup>	7.95 $\pm$ 0.10 <sup>a</sup>
GBC2	7.38 $\pm$ 0.19 <sup>b</sup>	7.33 $\pm$ 0.19 <sup>b</sup>	6.98 $\pm$ 0.18 <sup>c</sup>	7.09 $\pm$ 0.19 <sup>c</sup>	7.07 $\pm$ 0.16 <sup>bc</sup>	7.04 $\pm$ 0.14 <sup>c</sup>	7.15 $\pm$ 0.15 <sup>b</sup>
GBC3	7.28 $\pm$ 0.19 <sup>b</sup>	7.42 $\pm$ 0.20 <sup>b</sup>	7.31 $\pm$ 0.23 <sup>c</sup>	6.98 $\pm$ 0.20 <sup>c</sup>	7.11 $\pm$ 0.21 <sup>bc</sup>	6.905 $\pm$ 0.13 <sup>c</sup>	7.17 $\pm$ 0.17 <sup>b</sup>
Mean $\pm$ SD	7.54 $\pm$ 0.175	7.57 $\pm$ 0.181	7.28 $\pm$ 0.173	7.22 $\pm$ 0.173	7.21 $\pm$ 0.176	7.22 $\pm$ 0.165	7.34 $\pm$ 0.147
F Test	*	*	*	*	*	*	*
C.D. 5%	0.488	0.505	0.485	0.484	0.493	0.461	0.411
SE(m)	0.175	0.181	0.173	0.173	0.176	0.165	0.147
C.V.	10.596	10.921	10.898	10.967	11.209	10.458	9.181

Note: Values are expressed as mean  $\pm$  SD of twenty-one determinations. WCS: White chia seeds; BCS: Black chia seeds; Germinated two varieties of white and black chia seeds keeping temperature and time constant at 24 °C for 36 hours

GWCST1- Germinated white chia seeds with distilled water; GWCST2- Germinated white chia seeds with warm distilled water at 45 °C; GWCST3- Germinated white chia seeds with 2 per cent sugar solution with warm distilled water; GBCST1- Germinated black chia seeds with distilled water; GBCST2- Germinated black chia seeds with warm distilled water at 45 °C; GBCST3- Germinated black chia seeds with 2 per cent sugar solution with warm distilled water

chia seeds. Studies shown by Srujana *et al.* (2019) on germinate quinoa showed similar amylase activity ranging from 0.15 to 1.48 mg/100g at different time interval 4 to 60 hours.

### Sensory Evaluation of Germinated Chia Seeds

Any processing method is acceptable only when it gives a product of acceptable quality by the consumer.

The results obtained revealed (Table 4) that germinated white chia seeds with distilled water GCST1 had the highest mean scores for appearance (7.92), colour (8.02), texture (7.79), taste (7.71), after taste (7.59), flavour (7.69) and overall acceptability (7.79); while germinated black chia seeds with distilled water GBCT1 also scored the highest in all sensory parameters. Germinated white and black chia seeds with distilled water (T1) are highly significant with respect to sensory scores. There were better overall scores for processed germinated chia seeds than the raw chia seeds.

Germination is a simple processing method which enhances the nutrient content and sensory properties. Chia seeds can be germinated using distilled water for white and warm distilled water for black. Highest vitamin C and  $\alpha$  amylase content was reported at 36 hours of germination. Panelist have accepted germinated chia seeds than the raw seeds.

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## AMMI Model and YREM - Based Grain Yield Stability of Horse Gram [*Macrotyloma uniflorum* (Lam.) Verdc.] YMV Disease Resistant Genotypes

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### ABSTRACT

Horse gram yellow mosaic virus (HgYMV) disease is one of the major biotic constraints in horse gram production. Development and deployment of cultivars resistant to HgYMV disease are considered as the most eco-friendly and sustainable approach to mitigate the production losses caused by the disease. However, for easy acceptance of such HgYMV disease-resistant cultivars by farmers, they should be in high yielding background. Under these premises, nine HgYMV disease-resistant genotypes including two checks were field-evaluated in triplicated randomized complete block design (RCBD) to identify those that exhibit stable grain yield plant<sup>-1</sup> across four (two location-two years combination) environments during 2020 and 2021 late rainy seasons. Additive Main effects and Multiplicative Interaction (AMMI) model was used to detect and characterize genotype × environment interaction (GEI). Genotype + Genotype × environment (GGE) bi-plot was used to visually (subjective criterion) interpret GEI patterns of genotypes and identify those that are stable across four environments. AMMI Stability Value (ASV) and Stability Index (SI) were used as objective criteria to assess relative stability of genotypes. A simple statistic, namely, yield relative to environment maximum (YREM) was also used to detect cross-over GEI and to quantify genotypes' attainable grain yield loss attributable to crossover GEI. The genotypes differed significantly and displayed significant GEI for grain yield. GEI<sub>signal</sub> explained over 50 per cent of total SS due to GEI. AMMI 2 model family was adequate to explain detected variation attributable to GEI. One genotype namely, 'Palem 2' was found highly stable across four test environments based on three criteria, namely GGE bi-plot, ASV and SI with high mean grain yield plant<sup>-1</sup>. 'Palem 2' with unit YREM is likely to maintain its high grain yield potential across temporal environments without reduction in grain yield even in the presence of cross over GEI.

**Keywords:** GEI, HgYMV disease, GGE bi-plot, AMMI stability value, Stability index, Resistance, YREM

**H**ORSE GRAM is one of the important climate-resilient indigenous grain legume crops in India. It is the fifth most widely grown legume crop in India (Fuller and Murphy, 2018). It is self-pollinated crop with 2n=20 chromosomes (Halder, 2012). It is one of the good sources of protein to a large number of people, especially those depending on vegetarian diet for source of energy (Morris, 2008). The productivity of horse gram is rather low (Fuller and Murphy, 2018)

as it is grown in marginal soils in rainfed ecosystems by resource-poor farmers. Besides this, its production is constrained by several biotic stresses. Among these, horse gram yellow mosaic virus (HgYMV) disease transmitted by whiteflies (*Bemisia tabaci*) is most devastating (Durga *et al.*, 2014).

Genetic management through the development and deployment of cultivars resistant to HgYMV disease



is considered as the most eco-friendly and sustainable approach to mitigate the production losses caused by the HgYMV disease. Host plant resistance is not only effective, safe, reliable and long-lasting method of control, but also forms an important component of integrated disease management (IDM). However, for easy acceptance of HgYMV disease resistant cultivars by the farmers, they should be in high yielding background. The use of high- yielding HgYMV disease resistant cultivars is expected to contribute to sustainable horse gram production. Identification and deployment of YMV disease resistant genotypes within the working germplasm is a short-term strategy to cater to the immediate YMV disease resistant cultivar requirement of the farmers. Towards this effort, a few genotypes with high levels of resistance to HgYMV disease from among 196 germplasm accessions were selected based on their evaluation in two hotspot locations, namely Main Research Station (MRS), Hebbal, Bengaluru and Krishi Vignana Kendra (KVK), Chamarajnar during 2021 and 2022 summer seasons. We hypothesize that at least one of these HgYMV disease resistant genotypes would serve as potential candidate cultivar if it displays grain productivity better than or at least as good as the check cultivars with high stability. To test this hypothesis, the HgYMV disease resistant genotypes were field evaluated to (i) detect genotype  $\times$  environment interaction (GEI) (if any), (ii) characterize GEI and (iii) identify the genotypes with high grain yield potential and stability across temporal environments.

## MATERIAL AND METHODS

### Experimental Material

The material for the study consisted of seven HgYMV disease resistant genotypes namely, Palem 1, Palem 2, Paiyur 1, Paiyur 2, IC-121640, IC-43516 and IC-392329 and two check varieties *viz.*, PHG 9 and BGM 1 (Table 1). PHG 9 and BGM 1 are high-yielding varieties released by the University of Agricultural Sciences (UAS), Bangalore, India for commercial horse gram production.

TABLE 1  
Details of the HgYMV disease-resistant genotypes used for the study

Genotypes	Source of collection	YMV disease response score
Palem 1	Agriculture Research Station (ARS), Palem, Andhra Pradesh (AP)	2
Palem 2	ARS, Palem, AP	2
Paiyur 1	Regional Research Station (RRS), Paiyur, Tamil Nadu (TN)	2
Paiyur 2	RRS, Paiyur, TN	2
IC-121640	Kerala	2
IC-43516	Karnataka	1
IC-392329	Jharkhand	2
<i>Yield checks</i>		
PHG 9	UAS, GKVK, Bengaluru, Karnataka	2
BGM 1	Karnataka	2

The genotypes were scored for their response to HgYMV disease on 1-6 scale, where, 1=highly resistant and 6=highly susceptible

### Methods

The seeds of 9 HgYMV disease resistant genotypes were sown in randomized complete block design (RCBD) with three replications at two locations, namely Gandhi Krishi Vignana Kendra (GKVK), Bengaluru and Zonal Agricultural Research station (ZARS), VC Farm, Mandya during 2020 and 2021 late rainy seasons. Each accession was sown in a single row of 3m length with a row-to-row spacing of 0.45m. Fifteen-days after sowing, the seedlings were thinned to maintain plant-to-plant spacing of 0.15m. A total of 15 to 16 plants survived to maturity in each genotype. Recommended crop management practices were followed during the crop growth period to raise a healthy crop. Data were recorded on ten randomly chosen plants (avoiding border ones) in each genotype on grain yield plant<sup>-1</sup> (g).

### Statistical Analysis

The replication-wise mean grain yield of HgYMV disease resistant genotypes was used for all statistical analysis as described in following sections.

## ANOVA

Analysis of variance (ANOVA) (Panse and Sukhatme, 1984) was performed to detect significant differences, if any, among the HgYMV disease resistant genotypes.

### Detection and Characterization of Genotype × Environment Interaction (GEI)

For purpose of detection of genotype × environment interaction (GEI), two location-two years combination was considered as four different temporal environments. Replication-wise mean grain yield data recorded from four environments was subjected to Additive main effects and multiplicative interaction (AMMI) model (Gauch and Zobel, 1988). The additive main effects of genotypes and environments were fitted by univariate ANOVA, followed by fitting multiplicative GEI by interaction principal component (IPC) analysis (Gauch and Zobel, 1988). The sum of squares attributable to signal-rich component of GEI ( $GEI_{\text{Signal}}$ ) was computed as  $GEI \text{ SS} - GEI_{\text{Noise}}$ , where,  $GEI_{\text{Noise}} = GEI \text{ degrees of freedom} \times \text{error mean squares}$  from the AMMI ANOVA (Gauch, 2013). The following model was used to estimate main effects of genotypes and environments, and GEI effects.

$$Y_{ij} = \mu + g_i + e_j + \sum_{k=1}^n \lambda_k \alpha_{ik} \gamma_{jk} + \varepsilon_{ij}$$

Where, ' $Y_{ij}$ ' is the mean grain yield of  $i^{\text{th}}$  genotype in the  $j^{\text{th}}$  environment, ' $\mu$ ' is the experimental mean grain yield, ' $g_i$ ' and ' $e_j$ ' are the  $i^{\text{th}}$  genotype and  $j^{\text{th}}$  environment mean deviation from ' $\mu$ ' respectively. ' $\lambda_k$ ' is the square root of eigen value of the  $k^{\text{th}}$  IPC axis, ' $\alpha_{ik}$ ' and ' $\gamma_{jk}$ ' are the IPC scores for  $i^{\text{th}}$  genotype and  $j^{\text{th}}$  environment, respectively and ' $\varepsilon_{ij}$ ' is the residual. All the analyses were implemented using RStudio software v.4.2.1.

### GGE Bi-Plot for Interpretation of GEI

Genotype + Genotype × environment (GGE) bi-plot is a subjective / qualitative means of characterizing GEI patterns and assessment of relative stability of test genotypes. GGE bi-plot utilises a combination of

GGE concepts and AMMI bi-plot (Yan *et al.*, 2000). GGE bi-plot has been suggested for visual interpretation of patterns of GEI, representativeness and discriminating ability of the environments and relative stability of test genotypes. The GGE bi-plot is based on the following model.

$$Y_{ij} - Y_i = \lambda_1 \alpha_{i1} \gamma_{j1} + \lambda_2 \alpha_{i2} \gamma_{j2} + \varepsilon_{ij}$$

Where, ' $Y_{ij}$ ' is the mean grain yield of  $i^{\text{th}}$  genotype in the  $j^{\text{th}}$  environment, ' $Y_i$ ' is mean grain yield of all the test genotypes in the  $j^{\text{th}}$  environment, ' $\lambda_1$ ' and ' $\lambda_2$ ' are square roots of eigen values of first and second IPC axes, 1 and 2, ' $\alpha_{i1}$ ' and ' $\alpha_{i2}$ ' are scores of the first and second IPC, respectively, for the  $i^{\text{th}}$  genotype and ' $\gamma_{j1}$ ' and ' $\gamma_{j2}$ ' are first and second IPC's respectively for  $j^{\text{th}}$  environment.

### AMMI model-based parameters to identify stable genotypes

The relative stability of genotypes can be assessed objectively based on the estimates of AMMI stability value (ASV) (Purchase *et al.*, 2000) and Stability Index (SI) (Farshadfar, 2011). The procedure and formulae for estimating ASV and SI are described in the following sections.

#### AMMI Stability Value (ASV)

ASV was estimated as,

$$ASV = \sqrt{\left[ \frac{SSIPC1}{SSIPC2} (\text{IPC1 score}) \right]^2 + (\text{IPC2 score})^2}$$

Where, SSIPC 1 and SSIPC 2 are sum of squares (SS) attributable to first two IPC's. Conceptually, ASV is the distance from zero in a two-dimensional scatter diagram of IPC 1 vs. IPC 2 scores (Purchase *et al.*, 2000). Since the IPC 1 score generally contributes proportionately more to GEI, it is weighted by the proportional difference between IPC 1 and IPC 2 scores in order to compensate for the relative contribution of IPC 1 and IPC 2 scores to the total

GEI sum of squares. Lower the magnitude of estimates of ASV, greater in the stability of the test genotypes. Higher the magnitude of estimates of ASV, lower is the stability of test genotypes (Purchase *et al.*, 2000).

### Stability Index (SI)

As ASV considers only stability, regardless of grain yield potential of genotypes, SI was estimated to facilitate simultaneous selection of test genotypes with high stability and high mean grain yield. SI was estimated as  $SI = RASV + RY$  where, RASV is rank of the test genotypes based on ASV and RY is the rank of test genotype based on mean grain yield (Farshadfar, 2011) across four environments. The test genotypes with low SI were regarded as those with high mean grain yield and high stability.

### Estimation of Yield Relative to Environment Maximum (YREM)

A simple statistic, namely YREM was used to detect crossover GEI and to quantify reduction in grain yield potential of test genotypes due to crossover GEI. Higher the value of YREM of a genotype, lower is the magnitude of crossover GEI and the lower is the extent of reduction in grain yield potential of that genotype even in the presence of crossover GEI. The grain YREM (Yan, 1999) was estimated as  $Y_{ij} = X_{ij} / MAX_{ij}$ , where, 'Y<sub>ij</sub>' and 'X<sub>ij</sub>' are the YREM and mean grain yield, respectively, of i<sup>th</sup> genotype in j<sup>th</sup> environment. MAX<sub>ij</sub> is the grain yield of highest performer in j<sup>th</sup> environment. The analysis was implemented using statistical analysis option available in Microsoft Excel software.

YREM is a special type of standardized estimate of genotypes' performance, with nullified environment main effect. It is also an intuitive and genotypes' attendance-independent measure of test genotype's performance (Yan, 1999). It is a dynamic measure of genotypes' performance, as it varies with the performance of best genotypes in a given environment and the best genotype also varies with the environment. The performance of best genotype is its potential attainable in a given environment. Hence, YREM is an indicative of magnitude of cross-over GEI. Therefore, in the absence of crossover GEI, the average YREM of a genotype tested across environment must be 1.0. Any departure of a genotype's YREM from 1.0 is interpreted as loss in its attainable grain yield attributable to crossover GEI (Yan, 1999). For example, if a genotype has an across-environments' average YREM = 0.90, then 10 per cent of its attainable grain yield is lost due to crossover GEI.

## RESULTS AND DISCUSSION

### ANOVA

ANOVA is the diagnostic step to detect different sources of variation relevant to the results of field experiments such as those being reported in the present study. Location-wise ANOVA revealed significant mean squares attributable to test genotypes in all four environments for grain yield plant<sup>-1</sup> (Table 2). These results indicated substantial differences among the test genotypes for grain yield plant<sup>-1</sup> and thus provide justification for their use in

TABLE 2  
ANOVA of HgYMV disease-resistant genotypes for grain yield plant<sup>-1</sup> (g) at GKVK, Bengaluru and Zonal Agricultural Research Station, Mandya

Source of variation	Degrees of freedom	GKVK, Bengaluru						ZARS, Mandya					
		2020			2021			2020			2021		
		MSS	'F' Statistic	P≥F	MSS	'F' Statistic	P≥F	MSS	'F' Statistic	P≥F	MSS	'F' Statistic	P≥F
Genotypes	08	2.22	59.92	0.00	2.39	61.18	0.00	2.11	140.07	0.00	2.05	51.16	0.00
Replication	02	0.07	2.02	0.16	0.22	5.72	0.01	0.28	18.79	0.00	0.05	1.28	0.30
Error	16	0.04			0.04			0.01			0.04		

MSS: Mean sum of squares

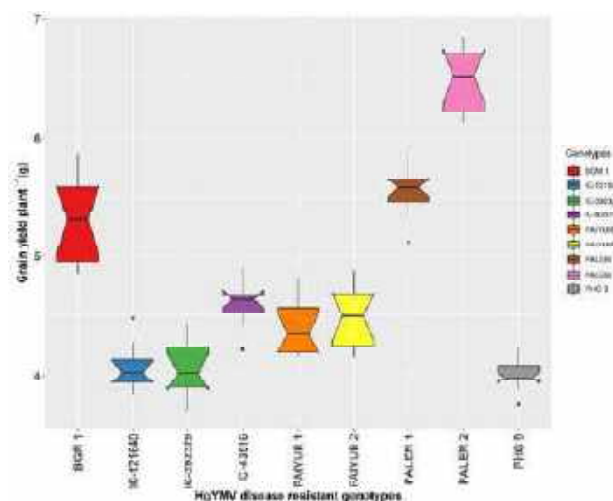


Fig. 1 : Box-Whisker plots showing significant differences among HgYMV disease-resistant genotypes for grain yield plant<sup>-1</sup>

the present study. Box-Whisker plots depicts the range of grain yield plant<sup>-1</sup> of seven test genotypes and two checks across the four environments. The genotype Palem 2 was the highest grain yielder followed by Palem 1 and BGM 1 (Fig. 1).

#### AMMI model - based detection and characterization of GEI effects

Additive ANOVA detects GEI only when the average of all (g-1) (e-1) degrees of freedom (df) contrasts is significant. Classical additive ANOVA indicate a lack of GEI, even when there exists significant GEI for some of the contrasts. Hence, classical additive ANOVA is not a desirable method for detecting GEI. Researchers can declare absence of GEI only if GEI sum of squares of one df is not significant (Gauch, 1988). As an intermediate approach between 1 and (g-1) (e-1) df, AMMI model is widely used to unambiguously detect GEI (Gauch, 1988). AMMI model uses additive ANOVA for detection of main effects of genotypes and environments and multiplicative IPC analysis of GEI effects. The rationale behind the AMMI model is that the observed performance of test genotypes in a particular environment is not the best estimate of true performance of that genotypes in that environment. This is because, most often than not test genotypes interact significantly with test

environment (s) and hence GEI is a rule rather than an exception (Bernardo, 2020). The GEI effects consists of (1) signal / pattern attributable to repeatable and predictable component and (2) noise attributable to non-repeatable and un-predictable component. AMMI model effectively dissects GEI in to 'signal' and 'noise' components using several IPC's. While the first few IPC's tend to capture most of the repeatable and predictable components, later IPC's capture non-repeatable and un-predictable component (Gauch, 2013). AMMI model estimates GEI for i<sup>th</sup> genotype and j<sup>th</sup> environment not only from data pertaining to i<sup>th</sup> genotype and j<sup>th</sup> environment, but also from data of all the genotypes' performance in all the test environments (Bernardo, 2020).

In the present study, sum of squares (SS) attributable to GEI was partitioned into those attributable to  $GSI_{Signal}$  and  $GSI_{Noise}$ . Differences among test genotypes and environments are necessary for existence of GEI effects. In the present study, significant mean squares (Table 3) suggested presence of substantial variability among the test genotypes for grain yield plant<sup>-1</sup>. Significant mean squares attributable to the GEI suggested differential performance of test genotypes across the four environments. However, over 50 per cent of SS due to  $GEI_{signal}$  contributes to SS due to GEI. Thus, a substantial portion of detected GEI effects are repeatable and hence predictable. However, mere detection of GEI does not provide information on the relative performance of genotypes across different test environments. Stability analysis help the researcher to examine the performance of genotypes relative to each other in different environments. Stability analysis requires AMMI model diagnosis, as AMMI constitutes a model family, not a single model. Consequently, model diagnosis is required to determine which member of this model family is best for a given data set and research purpose. The significance of mean squares attributable to IPC's is widely used as a criterion to diagnose the best AMMI model family member for given data set (Gauch, 2013). In the present study, sum of squares (SS) attributable to the first two IPC's explain >99.9 per

TABLE 3  
AMMI ANOVA of HgYMV disease-resistant genotypes for grain yield plant<sup>-1</sup> (g)

Source of variation	Degrees of freedom	Sum of squares	Mean sum of squares	'F' Statistic	P≥F	Proportion
Genotypes	08	68.69	8.58	261.61	0.00	-
Environment	03	0.45	0.15	0.94	0.04	-
G×E interaction	24	1.51	0.06	1.91	0.02	-
PC1	10	1.13	0.11	3.43	0.00	74.70
PC2	08	0.37	0.05	1.41	0.02	24.60
PC3	06	0.01	0.01	0.06	0.99	0.70
Residual	64	2.10	0.03	-	-	-
GEI signal	-	0.79				52.31
GEI noise	-	0.72				47.68

GEI signal SS (0.79) = GEI SS (1.51) - GEI noise SS (0.72), where, GEI noise SS (0.72) = GEI degrees of freedom (24) × AMMI Error MSS (0.03)

cent of SS due to GEI. Further, the significance of mean squares attributable to first two IPC's indicate AMMI 2 is the best model family member that captures predictable component of GEI. Selection of the best AMMI model family member is the key for reliable estimates of genotypes' performance and selection of best genotype (s) with highly predictable performance in future years as well. This argument stems from the fact that it is rather difficult to exploit genotype × temporal environments (such as years) interaction, as breeders cannot establish independent breeding programs for different years. This is because, climate conditions that generate genotype × year interaction variation are not known *a priori*. From grower's point of view, location is a constant-not-variable factor and grain yield consistency over years is the only relevant component of genotypes' performance (Annicchiarico *et al.* 2006). This is because, success of identified best genotype as cultivar in growers production environments depends on the stability of its performance in future years after its release for commercial production (Spoorthi *et al.* 2021b). Several researchers such as Arunkumar and Konda (2014) and Bhardwaj *et al.* (2014) in mungbean, Vaijayanthi *et al.* (2017) in dolichos bean and Khan *et al.* (2021) in bambara groundnut have also detected significant GEI for grain yield and its component

traits. Further, several previous researchers such as Piepho (1994) in fababean, Annicchiarico *et al.* (2006) in wheat, Ebdon and Gauch (2011) in turfgrass, Sadiyah and Hadi (2016) in rice and Spoorthi *et al.* (2021a) in dolichos bean have also reported adequacy of most parsimonious AMMI model family *i.e.* AMMI 2 model to explain the observed variation attributable to GEI.

The significant repeatable component of GEI effects detected in the present study warrants identification of HgYMV disease-resistant genotypes that are specifically suitable to each environment to maximize horse gram production in each environment with/without the presence of YMV infection. The relative stability of test genotypes was assessed based on visual interpretation using GGE bi-plot and stability parameters. While assessment of stability based on GGE bi-plot visualization is a subjective method, that based on stability parameters is an objective method.

#### Assessment of Stability based on GGE Bi-Plot

A major purpose of yield - trial research is the selection of best genotypes for use as a cultivar in target environment. Stability of test genotypes across temporal environments as is the case in the present study is particularly important as it reduces susceptibility to unpredictable component of GEI

effects. The stability of test genotypes across four temporal environments can be qualitatively assessed using the graphical representation of test genotypes based on their first two IPC's in GGE bi-plot (Yan *et al.* 2000). GGE bi-plot is a multivariate analytical tool that graphically displays the interaction between each genotype and each environment. It is a two-dimensional graph and allows visualization of the inter-relationship among environments and test genotypes. There are numerous ways to use and interpret GGE bi-plot. However, four views of the GGE bi-plot are most relevant (Segherloo *et al.*, 2010). These are (i) average environment coordination (AEC) view based on test genotype-focused scaling for ranking of the test genotypes relative to ideal genotype; the ideal genotype is the one whose point is located in the centre of concentric circles in the GGE bi-plot (ii) discriminating and representativeness of test environments view (iii) polygon view based on symmetrical scaling for determining 'which-won-where' pattern of test genotypes in test environments, and (iv) AEC view based on environment-focused scaling for interpreting mean performance of the genotypes vs. their stability patterns (Yan and Kang, 2003). The results of the four views of GGE bi-plot are discussed in the following sections.

### Genotype (s) Relative to Ideal Genotype

An ideal genotype is the one with high mean performance and high stability across the test environments. A single arrowed line passing through the origin in the biplot and center of the circle is referred to as an average environment coordinate (AEC). The average environment is represented by the small circle at the end of the arrow (Yan and Tinker, 2006). An ideal genotype is present at the center of concentric circles with AEC passing through it in positive direction and has a vector length equal to the longest vector of the genotype on the positive side of AEC. Using the ideal genotype as center, several concentric circles are drawn around to help in easy visualization of the distance between each test genotype and ideal genotype. Stable genotypes are the ones which are located closer to the ideal

genotype. The test genotypes namely IC-43516 and Paiyur 2 were identified as near ideal ones on account of being closer to ideal genotype which is located at origin (Fig. 2a).

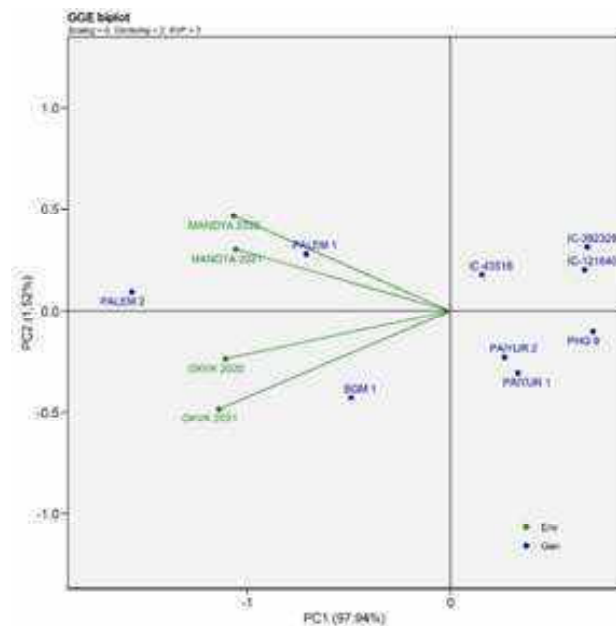


Fig. 2a : Average environment coordination (AEC) view of GGE-biplot for identification of test genotypes relative to ideal genotypes for grain yield plant<sup>1</sup>

### Discriminative ability and representativeness of test environments

Dotted line connecting the test environment pointing to the origin is called environment vector. The length of environment vectors and angle between the respective environment vector with AEC helps in identifying the discriminating ability and representativeness of the test environments. A discriminative environment is the one which has the ability to discriminate between test genotypes while a representative environment should represent average of the four test environments. Shorter and longer environment vectors indicate lower and higher discriminative ability of the environments, respectively. Small and large angle between environment vectors and AEC indicate most and least representativeness of environments, respectively. The acute and obtuse angle between the test environment vectors indicate similarity and dissimilarity between the test environments, respectively. In the present

study, GKVK 2021 late rainy season is discriminative as its environment vector is longer than other environmental vectors. On the other hand GKVK 2020 late rainy season is a representative environment as the vectors of these environments are oriented in acute angle relative to AEC (Fig. 2b).

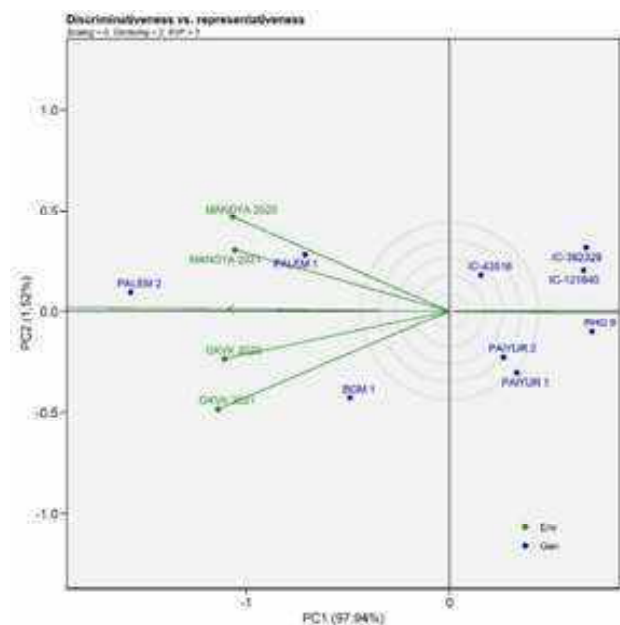


Fig. 2b : Discriminative vs. representativeness view of GGE-biplot for grain yield plant<sup>-1</sup>

### ‘Which-won-where’ View

Polygon view of GGE biplot helps in identifying which won where pattern of genotypes. A polygon is formed by joining all the test genotypes farther from the biplot origin in such a way that all of them fell within the polygon. Perpendicular lines called equality lines, originating from biplot origin are drawn to each side of the polygon. The equality lines divide the biplot into sectors. The vertex genotype in each sector is the winning genotype at environments whose markers (point) fall into the respective sector (Yan *et al.*, 2000). Thus, environments whose markers fall in the sector will have the same winning genotype, while environments of different sectors have different winning genotypes. Thus, polygon view of GGE biplot indicates the presence or absence of crossover GEI. In the present study, test genotypes such as Palem 1 and Palem 2 occupied vertices of the polygon. While

Palem 1 was the winner in ZARS, Mandya during both 2020 and 2021 late rainy seasons, Palem 2 was the winner in GKVK, Bengaluru during both 2020 and 2021 late rainy seasons for grain yield plant<sup>-1</sup> (Fig. 2c).

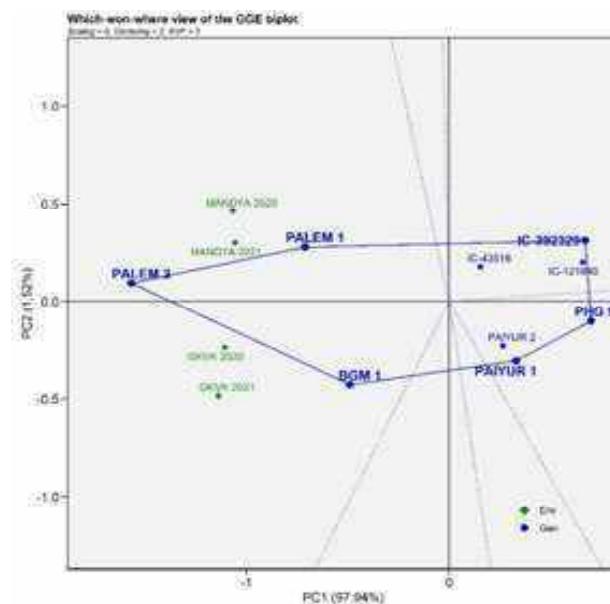


Fig. 2c : Polygon view of GGE-biplot based on the symmetrical scaling for “which won-where” pattern of test genotypes and environments for grain yield plant<sup>-1</sup>

### Mean Performance vs. Stability Patterns

The mean performance and stability could be visualized based on the location of genotypes in relation to AEC using AEC view of GGE bi-plot. The single-arrowed AEC points to higher mean performance of the genotypes across test environments (Yan, 1999). The genotypes with their points located towards AEC arrow are considered to exhibit high mean performance. On the contrary, the genotypes with their points located opposite to AEC arrow are considered to exhibit lower performance. Further, the relative lengths of projections of the genotypes from AEC are indicative of their relative stability. Shorter the length of the projections of genotypes from AEC, greater is the stability of the genotypes. Longer the projections of genotypes, poorer in their stability (Yan and Kang, 2003). In the present study, Palem 2 with shortest vector from the AEC line, was identified as a highly stable genotype across test environments with higher mean grain yield plant<sup>-1</sup> (Fig. 2d).

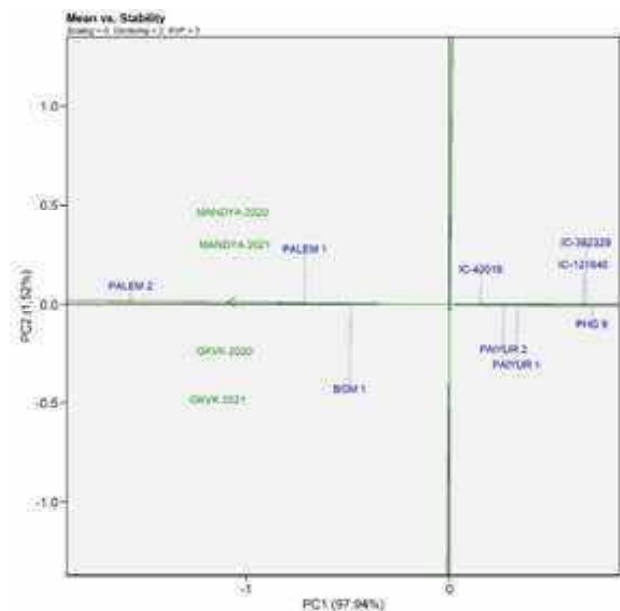


Fig. 2d : Average environment coordination (AEC) view of GGE-biplot based on environment-focused scaling for the mean performance vs. stability of test genotypes for grain yield plant<sup>-1</sup>

**AMMI Model-based Stability Parameters**

**AMMI Stability Value (ASV)**

ASV provides an objective criterion of assessment of stability and hence help to identify test genotypes

stable across the four environments. ASV is the distance from zero in a two-dimensional scatter-plot of IPC 1 scores against IPC 2 scores. In the present study, ASVs were estimated using both IPC 1 and IPC 2, as they significantly contributed towards total GEI variance of grain yield plant<sup>-1</sup> (Table 3). In the present study, Palem 2 and PHG 9 with lower magnitude of the estimates of ASV (Table 4), were adjudged as stable genotypes across the four test environments for grain yield plant<sup>-1</sup>.

**Stability Index (SI)**

SI which takes into account of both mean grain yield and stability in a single criterion helps in simultaneous selection of genotypes with desired performance for mean grain yield coupled with stability. The genotypes with low SI are regarded as those with high grain yield and stability. In the present study, Palem 2 and Palem 1 with lower magnitude of SI (Table 4), were regarded as the best genotypes with high grain yield and stability. Several researchers such as Patel *et al.* (2009), Arunkumar and Konda (2014), Bharadwaj *et al.* (2014), Vijayanthi *et al.* (2016), Vijayanthi *et al.* (2017), Kavya and Rangaiah (2019) have also identified genotypes stable across

TABLE 4

Estimates of AMMI model-based parameters to assess stability of nine HgYMV disease-resistant genotypes for grain yield plant<sup>-1</sup> (g)

Genotypes	Mean	RY	ASV	RASV	SI	Average YREM
Palem 1	5.56	2	0.66	4.5	06.5	0.85
Palem 2	6.49	1	0.15	2.0	03.0	1.00
Paiyur 1	4.41	6	0.84	7.0	13.0	0.68
Paiyur 2	4.48	5	0.66	4.5	09.5	0.69
IC-121640	4.06	7	0.78	6.0	13.0	0.62
IC-43516	4.61	4	0.59	3.0	07.0	0.71
IC-392329	4.04	8	1.20	8.0	16.0	0.62
<i>Yield checks</i>						
PHG 9	4.01	9	0.13	1.0	10.0	0.62
BGM 1	5.30	3	1.42	9.0	12.0	0.82
SEm±	0.28					
CD @P=0.05	0.60					

RY: Rank of the test genotype based on mean grain yield, ASV: AMMI Stability Value, RASV: Rank of the test genotype based on ASV, SI: Stability Index, YREM: Yield relative to environment maximum.



temporal environments based on SI. Of these two genotypes, Palem 2 was found highly stable across four test environments based on three criteria, namely GGE bi-plot, ASV and SI with high mean grain yield plant<sup>-1</sup>.

### YREM

Considering that YREM is a simple statistic which is independent of genotypes' attendance, it could be used as a predictor of genotypes' performance in future years (Yan, 1999). In the present study, unit YREM of Palem 2 (Table 4) indicates that its interaction with the four test environments is of non-crossover type. Unit YREM of Palem 2 also indicates that it remained highest yielder in all the four environments and its grain yield potential as assessed in the present study is attainable in all the test temporal environments without any loss, even if there exists cross-over GEI. Ashwini *et al.* (2021) and Spoorthi *et al.* (2021b) have also used YREM to detect crossover GEI, and to identify stable horse gram and dolichos bean genotypes respectively. Thus, Palem 2 with significantly higher grain yield potential and stability than both the checks, and unit YREM could be used as a cultivar for commercial production in GKVK, Bengaluru and ZARS, Mandya.

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## Differences in Crossbred Cattle Management, Production and Contribution to Livelihood Security in South and North Karnataka

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### ABSTRACT

Crossbred cattle have major contribution in fulfilling the demand of milk by the growing population of India. Among various categories of dairy animals, the productivity was higher in crossbred cattle. The present study was carried out in six districts of Karnataka based on sizable population of crossbred cattle. From each district, two taluks and from each taluk, a cluster of villages were identified based on crossbred cattle population. From each cluster of villages, 20 households owning crossbred cattle were randomly selected, making the total sample size of 240 farm households. The study revealed that the sample households had more number of milking crossbred cattle in south Karnataka as compared to north. Total feed cost and expenditure per animal was more in case of south Karnataka, producing more milk as compared to north. Net return/day per animal and per farm with and without considering cost of fodder were better in south Karnataka as compared to north. Employment generation (mandays/year) per animal was more in case of crossbred cattle of south as compared to north. However, protein, fat and calcium nourishment per animal to the family was better in north Karnataka. Nutrients to farm *i.e.* NPK kg/year/animal was also higher in north Karnataka. Economic and production constraints were expressed by more number of farmers compared to infrastructural and technical constraints. In all the categories, north Karnataka farmers expressed more constraints than in south Karnataka.

*Keywords* : Management, Crossbred cattle, Dairy farming, Livelihood

LIVESTOCK provides livelihood to two-third of rural community and provides employment to about 8.8 per cent of the population in India. India has vast livestock resources. Livestock sector contributes 4.11 per cent GDP and 25.6 per cent of total Agriculture GDP. India is World's highest livestock owner at about 536.76 million. Out of this, 193.47 million is cattle population *i.e.*, 142.11 million indigenous and 51.36 million crossbred cattle (as per 20<sup>th</sup> Livestock Census, 2019). India is the highest milk producer with 198.4 million tons (Pathak *et al.*, 2022). The per capita availability of milk has also increased from 112 grams per day in 1968-69 to 406 gram per day in 2019-20 (Basic Animal Husbandry Statistics, 2021). Crossbred cattle are playing crucial

role in the national economy through supply of milk, dung, fuel etc.

Karnataka stands 11<sup>th</sup> in milk production producing 90.31 lakh tonnes of milk in 2019-20. Hence dairying has become an important source of income for millions of rural families and has assumed an important role in providing employment and income generating opportunities. Karnataka state comprised about 9 per cent of crossbred cow population of the nation contributing more than 80 per cent of milk production in the state. Among cow milk, contribution of crossbred was immense (>75 per cent) than indigenous cows. Among various categories of dairy animals, the productivity was higher in crossbred

cows, followed by buffaloes, non-descript cows and goat. The milk productivity of crossbred cow was slightly lower in Karnataka when compared to the national average (Basic Animal Husbandry Statistics, 2021). Within the state, there were differences in productivity and profitability in different regions. Considering the above facts, present study entitled 'Differences in crossbred cattle management, production and contribution to livelihood security in South and North Karnataka' was undertaken.

### MATERIAL AND METHODS

The present study was carried out purposively in the state of Karnataka. The sampling scheme adopted for this study was three-stage stratified random sampling without replacement. Three districts from south and three from north Karnataka were identified based on sizable population of crossbred cattle. Next, from each district, two taluks and within each taluk, one cluster of village/panchayat were identified based on population density of crossbred cattle. From each selected cluster, 20 households owning crossbred cattle during the survey were selected randomly to serve as the sample. One adult member or head of the household actively engaged in management of crossbred cattle was considered as the respondent. Thus, 20 cattle owners from each cluster, made a total of cattle owners sample size to 240.

The data were collected through semi-structured interview schedule. Information on production and livelihood security parameters was collected and analyzed for estimating the costs, returns from milk production and contribution in livelihood security of farmers from crossbred breeds. The statistical significance of differences in production parameters were tested by using 'z' test with the help of SPSS software. Livelihood security is operationalized as contribution made by crossbred breeds in terms of income generation, nourishment to the family, nutrients to farm, employment generation, security during uncertainties and social status symbol. The index developed by Biradar *et al.* (2013) was used with required modifications as given below:

- *Contribution to the Total Household Income* : The net return was measured by collecting information on different production values of each cow and average values of each parameter were calculated.
- *Nourishment to the Family* : Based on the daily average milk consumed by the family, the nutrients were computed in terms of protein, fat and calcium as suggested by Gopalan *et al.* (1971).
- *Nutrients to the Farm* : The average farm yard manure applied to their respective farm was converted in terms of N, P and K by following the conversion factors suggested by Gautam (2007), that is, one tonne of farm yard manure was equivalent to 8 Kg N, 4 Kg P<sub>2</sub>O<sub>5</sub> and 16 Kg K<sub>2</sub>O.
- *Employment Generation* : Number of hours engaged in crossbred cattle rearing for one year was collected. Total hours spent in a year were divided by 8 hours to convert them in to man-days. Total number of man-days contributed was expressed as mean values.
- *Security during Uncertainties* : Number of households having used crossbred cattle to face the uncertainties in the past two years.
- *Status Symbol* : The number of households who regard keeping crossbred cattle as symbol of social status.

### RESULTS AND DISCUSSION

Data on age, caste, education, family size, landholding, farming experience and income given in Table 1 indicated that majority of cattle owners of both the regions belonged to middle age group and were from general category. Only a smaller portion of the respondents represented SC category. As younger generation is preferring jobs in urban areas, most of the farming practices are shouldered by middle age group. However, sizable population of ST category was involved in dairying in north Karnataka which may be due to accessibility to better resources as compared to south. The majority of cattle owners of both the regions were having high school or intermediate level of education.

TABLE 1  
Socio-economic characteristics of cattle owners of south and north Karnataka

Socio-economic characteristics	Category	South % n=120	North % n=120	P value
Age	Young	17.50	30.00	0.066
	Middle	50.00	45.00	
	Old	32.50	25.00	
Caste	General	50.00	65.83	0.000
	OBC	43.33	20.83	
	SC	6.67	5.83	
	ST	0.00	7.50	
Education	Illiterate	25.00	8.33	0.000
	Primary	4.17	12.50	
	High School/Inter.	65.00	66.67	
	Graduate & above	5.83	12.50	
Family size	Small	65.83	34.17	0.000
	Medium	30.00	49.17	
	Large	4.17	16.67	
Land Holding	Landless	0.00	1.67	0.000
	Marginal	38.33	15.83	
	Small	46.67	20.00	
	Medium	10.83	17.50	
	Large	4.17	45.00	
Experience	Low	23.33	32.50	0.123
	Medium	50.00	37.50	
	High	26.67	30.00	
Annual Income	Low	96.67	64.17	0.000
	Medium	3.33	15.00	
	High	0.00	20.83	

There were illiterate respondents and also graduates, although few in numbers. Majority of cattle owners of south Karnataka lived in small families and owned small land holding, while that of north Karnataka was having medium family size as well as large land holding. The annual income of majority cattle owners was low despite majority farmers had medium to high level of experience in cattle farming. Chi-square test was used to test the association between farmers of different districts and socio-economic characteristics. It was found that

farmer categories of different districts were significantly ( $p < 0.05$ ) associated with socio-economic characteristics such as caste, education, family size, land holding and annual income.

### Management Practices

Stall feeding with hay, green fodder and concentrates was the most common practice followed by open grazing during day time. Sizable households were feeding mineral mixture, but very few adopted the practice of silage feeding. All the crossbred cattle

TABLE 2  
Management practices followed for crossbred cattle in south and north Karnataka

Management practices	South % n=120	North % n=120	P Value
Natural service	0.83	1.67	0.561
Artificial insemination	100.00	100.00	NA
Open grazing	70.00	61.67	0.174
Stall feeding	100.00	100.00	NA
Feeding concentrates	100.00	100.00	NA
Feeding green fodder	100.00	100.00	NA
Mineral mixture feeding	44.17	35.83	0.188
Silage feeding	3.33	0.00	0.044
Hay feeding	100.00	100.00	NA
Closed type of housing	65.00	45.00	0.002
Pucca structure of housing	27.50	36.67	0.128
Location of housing as adjacent of house	44.17	30.83	0.033
Roof of thatch	68.33	23.33	0.000
Roof of asbestos	23.33	70.00	0.000
Open sides ventilation	49.17	50.00	0.897
Stone walls	33.33	40.00	0.284
Brick walls	48.33	27.50	0.001
Plastered wall surface	35.00	45.83	0.087
Concrete floor	36.67	43.33	0.292
Constructed feed manger	42.50	34.17	0.184
Tank watering	27.50	44.17	0.007
Drainage	84.17	87.50	0.459
Twice daily shed cleaning	63.33	60.83	0.690
Day and night confinement	44.17	75.00	0.000
Special protection of newborn calf	75.00	57.50	0.004
Vaccination	81.67	70.00	0.035
Deworming of adult	53.33	45.00	0.197
Deworming of calves	75.00	35.83	0.000
Allowing new born to suckle colostrum within 30 minutes	97.50	99.17	0.313
Disinfection of the naval cord	46.67	22.50	0.000
Proper dispose of dung and urine	90.83	92.50	0.640
Ecto-parasite control measure	73.33	33.33	0.000
Treatment of sick animal by Veterinarians	72.50	60.83	0.055
Trimming of hoof	27.50	17.50	0.064
Horn polishing	11.67	35.00	0.000
Regular cleaning of animal	90.00	84.17	0.178
Cleaning animal shed	99.17	95.00	0.055
Disbudding	33.33	22.50	0.061
Full hand milking method followed	84.17	66.67	0.002
Clean milking method practiced	85.83	35.83	0.000

in the sampling households was provided with Artificial Insemination (AI). This may be due to easy accessibility of AI services through Animal Husbandry Department. Majority adopted closed housing, with either thatch or asbestos roofing. Stone or brick-walls had open sides or windows, mostly without plastering. Concrete floor and feed manger were less common. But majority cattle-sheds had good drainage with shed cleaning done twice daily. In majority cases, animals were confined during night in south and day-night in north which shows that day grazing is a common practice in south. Adoption of health care practices such as vaccination, ecto-parasites control, deworming was better in case of south Karnataka as compared to north. The major reason could be the less awareness among farmers about vaccination and deworming and non access of veterinary services to farmers located in interior and remote areas in case of north. Most newborn were allowed to suckle colostrum within 30 minutes, but disinfection of naval cord was not practiced by majority farmers. Treatment of sick animal was done mostly by veterinarians. Under general practices, majority farmers were regularly cleaning animals and animal shed, but few were following trimming of hoof, disbudding and horn polishing. Majority farmers adopted clean milk production with either full hand milking or full hand

and stripping in south Karnataka, but in north Karnataka clean milk production practices was less adopted.

### Reproductive Parameters

Reproductive parameters of crossbred cattle were ascertained based on the data related to age of puberty, age at first calving, lactation length, dry period, productive life span, inter calving period, conception rate, service period, insemination time and no. of inseminations required to conceive. The average values of the reproduction parameters are presented in Table 3. There was no significant difference with respect to age of puberty, age at first calving, lactation length, dry period, inter calving period, service period and no. of inseminations required to conceive between south and north Karnataka. The productive life span was two years more in north Karnataka (11.76 years) than in south (9.67 years). Conception rate was better in south Karnataka as each animal required 2.66 services per conception compared to north (2.90 services/conception). This could be due to poor quality semen or more reproductive disorders in north Karnataka. Whereas insemination time was better in north Karnataka (9.10 hrs) as compared to south (12.13 hrs). Differences were significantly different in respect of

TABLE 3  
Reproduction parameters of crossbred cattle in south and north Karnataka

Parameter	South		North		P value
	Mean	SD	Mean	SD	
Age of puberty (Yrs)	1.83	0.32	1.87	0.31	0.301
Age at first calving (Yrs)	2.82	0.35	2.90	0.30	0.049
Lactation length (Months)	8.87	0.98	8.61	1.17	0.069
Dry period (Months)	4.09	1.57	3.95	1.06	0.414
Productive life span (Yrs)	9.67	1.35	11.76	1.73	0.000
Inter calving period (Months)	15.68	1.82	15.83	1.55	0.491
Conception rate (No. of service)	2.66	0.52	2.90	0.47	0.000
Service period (Months)	4.33	1.02	4.29	0.90	0.763
Insemination time (hrs)	12.13	2.72	9.10	2.32	0.000
No. of inseminations carried out	1.21	0.44	1.13	0.33	0.116

productive life span, conception rate and insemination time.

### Production Parameters

On the parameters related to dairy production (Table 4), the sample households had more number of milking per day (1.98) in south as compared to north (1.34). Crossbred cattle were producing more milk (7.33 L/anim./day) in south as compared to north (6.33 L/day). Average quantity of dry fodder and concentrates fed per animal was 7.11 & 3.33 kg respectively in south as compared to 7.00 & 2.88 kg respectively of north. But, average quantity of green fodder fed per animal was less (16.50 kg) in south as compared to north (18.44 kg). Thus, total feed cost and expenditure per animal was more in south Karnataka (Rs.106.52 & 140.52, resp.) than in north Karnataka (Rs.101.75 & 135.71, resp.). Crossbred cattle required less expenditure on health per day/animal (Rs.4) but the net return/day

per animal (Rs.64.81) was more in south as compared to north (Rs.41.39). This was because of more productivity of dairy animals, more awareness, less resource constraints including availability of good quality fodder and grazing lands due to high rainfall in south Karnataka as compared to north Karnataka.

Majority of the cattle owners used own farm grown dry and green fodder to feed their cattle or from grazing. Without considering the cost of fodder as shown in Table 4, total feed cost (Rs/anim./day) was more in case of crossbred cattle in south (53.33) as compared to north (46.07). Thus, total expenditure (Rs/anim./day) was more in case of crossbred cattle (87.33) in south as compared to north (80.03). Net return/day per animal (Rs.118) was more in case of crossbred cattle in south as compared to north (Rs.97.07). Dung produced (25.20 to 25.25 Kg/day/animal) was used as manure in own farm. Consumption of milk provided nourishment to family (0.10 & 0.28

TABLE 4  
Production parameters of crossbred cattle in south and north Karnataka

Parameter	South		North		P value
	Mean	SD	Mean	SD	
Total milking animals (no.)	1.98	1.48	1.34	0.97	0.000
Total milk production (L/day)	14.61	13.24	8.50	6.25	0.000
Total milk production (L/anim./day)	7.33	1.37	6.33	0.87	0.000
Total dry fodder fed (kg/anim./day)	7.11	1.74	7.00	2.32	0.694
Total daily green fodder fed (kg/anim./day)	16.50	6.10	18.44	4.37	0.005
Total concentrate fed (kg/anim./day)	3.33	0.72	2.88	0.79	0.000
Total feed cost (Rs/anim.)	106.52	19.23	101.75	17.52	0.046
Labour cost (Rs/anim./day)	30.00	.000a	30.00	.000a	Na
Health cost (Rs/anim./day)	4.00	.000a	4.00	.000a	Na
Total expenditure (Rs/anim./day)	140.52	19.23	135.71	17.48	0.044
Net return/anim. (Rs./day)	64.81	31.17	41.39	20.05	0.000
Milk nourishment to the family (L/day)	0.10	0.43	0.28	0.55	0.005
Employment generation (hrs/day)	1.82	0.32	1.69	0.41	0.008
Dung production (Kg/day/anim.)	25.20	3.37	25.25	3.73	0.906
<b>Without considering cost of fodder</b>					
Total feed cost (Rs/anim./day)	53.33	11.56	46.07	12.66	0.000
Total expenditure (Rs/anim./day)	87.33	11.56	80.03	12.61	0.000
Net return/anim. (Rs./day)	118.00	37.22	97.07	25.26	0.000



L/day in case of south & north). Similar results reported in western Maharashtra (Kolekar *et al.*, 2015). The 'z' test was used to test the difference between the production parameters perceived for crossbred cattle of south and north. Analysis showed that there was a significant difference between majority production parameters of two regions.

Contribution of crossbred cattle to the farmer's livelihood is presented in Table 5. Net return/day per animal (Rs.64.81) and per farm (Rs.127.50) was more in south as compared to north (Rs.41.39 & 55.74, respectively), as majority of the cattle owners used own farm grown dry and green fodder to feed their cattle or from grazing. Without considering the cost of fodder also, net return/day per animal (Rs.118) and per farm (Rs.234.88) was more in crossbred cattle of south as compared to north cattle (Rs.97.07 & 130.08 resp.). Protein, fat and calcium nourishment per animal to the family gm/day was less in case of crossbred cattle of south (3.2, 4.1 & 0.12, respectively) as compared to north (8.96, 11.48 & 0.34,

respectively). Similarly, nutrients supplied to farm *i.e.* NPK kg/year/animal was less in case of crossbred cattle of south (72.96, 36.48 & 145.92, respectively) as compared to north (73.6, 36.8 & 147.2, respectively). Employment generation (Man days/year) per animal was more in case of crossbred cattle of south (83.04) as compared to north (77.11). Security for uncertainties and status symbol was more in case of crossbred cattle of south (90.83% & 84.17%, respectively) as compared to north (71.25% & 75.41%, respectively). The 'F' & 'Chi-square' test was used to test the difference between the types of contribution perceived by farm households in case of crossbred cattle of south and north. Analysis showed that there was a significant difference between majority types of contribution of crossbred cattle of south and north Karnataka.

#### Constraints in Rearing of Cows

As per the data presented in Table 6, economic constraints were perceived by most of the respondents. High cost of treatment, high cost for feeding,

TABLE 5  
Contribution of crossbred cattle to the farmers livelihood in south and north Karnataka

Type of contribution	Unit	Values		P Value
		South	North	
Income from cows	Net return/anim./day (Rs.)	64.81	41.39	0.000
	Net return/farm/day (Rs.)	127.50	55.74	0.000
	Net return/L (Rs.)	5.80	5.58	0.604
Income from cows (Without considering cost of fodder)	Net return/anim./day (Rs.)	118.00	97.07	0.000
	Net return/farm/day (Rs.)	234.88	130.08	0.000
	Net return/L (Rs.)	10.52	13.34	0.000
Nourishment to the Family	Protein (gm/day/family)	3.2	8.96	0.005
	Fat (gm/day/family)	4.1	11.48	0.005
	Calcium (mg/day/family)	120	336	0.005
Nutrients to the Farm	N kg/year	72.96	73.6	0.906
	P kg/year	36.48	36.8	0.906
	K kg/year	145.92	147.2	0.906
Generating Employment	Man days/year	83.04	77.11	0.008
Security for Uncertainties	Percentage	90.83	71.25	0.000
Status Symbol	Percentage	84.17	75.41	0.000

TABLE 6  
Constraints in rearing of crossbred cattle in south and north Karnataka

Constraints	South %n=120	North %n=120	P Value
<i>Economic Constraints</i>			
High cost of treatment	94.17	100.00	0.007
High cost for feeding	92.50	100.00	0.002
Costly wages for workers	90.83	100.00	0.001
No access to credit facility	81.67	99.17	0.000
Poor economic condition	85.83	96.67	0.003
<i>Infrastructural Constraints</i>			
Poor supply of quality semen	34.17	68.33	0.000
Veterinary dispensary located at far away distance	20.83	58.33	0.000
Lack of organized market	32.50	53.33	0.001
Unavailability of veterinary services in time	36.67	47.50	0.089
<i>Technical Constraints</i>			
Poor mass media or extension agency contact	29.17	65.00	0.000
Unavailability of extension advisory services	30.00	63.33	0.000
Unavailability of improved technology	59.17	62.50	0.597
Lack of knowledge on improved practices	49.17	62.50	0.038
Not participated in any training programme	51.67	56.67	0.437
<i>Production Constraints</i>			
Longer inter-calving period	98.33	100.00	0.156
Competition from commercial dairy	97.50	75.83	0.000
Unavailability of grazing land	72.50	90.83	0.000
Uncertain rain fall	89.17	90.83	0.667
Poor milk production	74.17	89.17	0.653
Disease incidence	75.00	85.00	0.053
Longer maturity age	22.50	30.83	0.144
Lack of market demand for cow milk	23.33	25.83	0.003

costly wages for workers were perceived as major constraints by more than 90 per cent farmers of both the regions. Longer inter-calving period, competition from commercial dairy, non-availability of grazing land, uncertain rainfall and disease incidence were perceived as the production constraints by majority farmers. Major infra structural constraint was the unavailability of veterinary services in time and poor supply of quality semen. Poor mass media or extension agency contact, non-participation in training programmes and unavailability of improved technologies were the major technical constraints

cited by the crossbred cattle farmers in both the regions, but more in north Karnataka.

Crossbred cattle were producing more milk (7.33 L/anim./day) in south Karnataka as compared to north (6.33 L/day). As a result, net return/day per animal (Rs.64.81) and per farm (Rs.127.50) was also more in crossbred cattle in south Karnataka as compared to north (Rs.41.39 & 55.74 resp.). Nutrients supply to farm *i.e.* NPK kg/year/animal from cross bred cattle was more in north Karnataka. Longer inter-calving period, high cost of treatment, high cost for feeding and costly wages for workers

were the important constraints perceived by farmers. The potential to enhance the productivity of the crossbred cattle of India through professional farm management and superior nutrition is immense. Therefore, there is need to make efforts for increasing production from crossbred cattle through proper breeding programs, good management practices etc. to hasten the efficiency of milk production and livelihood security of resource poor farmers.

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## Host Plant Traits Impact on the Egg Laying Choice of Female Fruit Borer Moth, *Earias vittella* (Fab.) in Okra

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### ABSTRACT

A comparative study was conducted with different species of *Abelmoschus* to understand the association between the plant traits and egg laying choice of okra shoot and fruit borer, *Earias vittella* (Fab.). The plant traits such as number of branches (NB), stem diameter (SD), leaf length (LL), fruit length (FL), fruit width (FW) and trichomes density (T) significantly influenced the number eggs laid by *E. vittella*. A step-wise multiple regression analysis revealed that the tested plant traits explained 79 per cent of the total variation in number of eggs laid by *E. vittella* ( $Y=17.29-0.61_{NB}-8.17_{SD}+0.48_{LL}+1.16_{FL}-5.73_{FW}+0.11_T$ ,  $R^2 = 0.79$ ) and role of these plant traits which impacts on the egg laying choice of *E. vittella* were discussed in detail.

**Keywords :** Okra shoot and fruit borer, *Abelmoschus* spp., Step-wise regression, Correlation, Path-coefficient analysis, Ovipositional choice

OVER millions of years of evolution, host plants have developed a variety of resistance mechanisms to repel the oviposition and feeding by insect pests. Wild crop relatives in particular are naturally resistant and have been evolved with several morphological, anatomical and physiological traits to circumvent insect pests' attack. Such desirable traits particularly in wild crop relatives are largely untapped. Till to date, Host Plant Resistance (HPR) against phytophagous insect pests is far less commonly exploited by the crop breeders compared to plant pathogens and this may be mainly due to lack of research to identify suitable resistant sources against the former.

Initial orientation of an insect to a potential host plant and subsequent feeding or oviposition is probably evoked by a combination of tactile, chemoreceptive and visual stimuli in many instances (War *et al.*, 2012). Host plant resistance based on morphological traits usually refers to plant traits that interfere with insect movement, feeding or

oviposition. Insect ovipositional behaviour is often influenced by the morphological traits such as size of plant/ plant parts, shape, colour, leaf hairs, cuticle thickness *etc.* that make the plant less attractive or present formidable physical barriers to insect pests (Dhillon & Sharma, 2004 and War *et al.*, 2012). In HPR, the foremost desirable attribute in the insect-plant relationship is resistance to oviposition (ovipositional antixenosis) as these influences 'no-fit' relationship between the necessity of the gravid female egg laying choice and the correlative morphological traits of the plant (Beck, 1965; Kessler & Baldwin, 2002 and Sharma, 2007).

Okra shoot and fruit borer, *Earias vittella* (Fab.) (Lepidoptera: Nolidae) is an economically important oligophagous pest that feeds on numerous host plants in Malvaceae. In okra, gravid female moths of *E. vittella* lays eggs singly on shoot tips, flower buds and tender fruits. The neonate larvae bores into delicate terminal shoots during vegetative phase and during fruit formation, they bore into flower buds and

young fruits (Thippeswamy *et al.*, 1980 and Qasim *et al.*, 2018). Damaged shoots wilt and dry out and infested fruits have a distorted look and contain larval excrement, rendering them unfit for consumption and crop losses often range from 3.5 to 90 per cent (Mandal *et al.*, 2006).

The field of insect-plant interactions has been dominated by studies on complex plant traits that prevent insect pests from approaching, landing, settling, feeding or ovipositing (Painter, 1951). In nutshell, the plant traits that affect the oviposition behaviour of insects (altered number of insects landing or number laying eggs) has been termed as ovipositional antixenosis (non-acceptance or morphological non-preference due to leaf hairiness, stem hardness). Plant traits are known to influence the preference and performance of phytophagous insects. Traits that influence oviposition can differ from traits that favour larval development, but in native hosts the association between traits usually leads to positive preference-performance relationships. However, when herbivores interact with novel hosts, traits that influence oviposition and successful larval development can become decoupled, leading to poor preference-performance relationships. Such host plant-insect relationships have been extensively investigated in several lepidopteran pests namely *Helicoverpa armigera* (Hubner) (Afzal *et al.*, 2012; Thakur *et al.*, 2017 and Ali *et al.*, 2019), *Spodoptera frugiperda* (JE Smith) (da Silva *et al.*, 2021); *Leucinodius orbonalis* (Guenée) (Wagh *et al.*, 2012; Niranjana *et al.*, 2015,); *Trichoplusia ni* (Hubner) (Coapio *et al.*, 2018); *Maruca vitrata* (Geyer) (Jakhar *et al.*, 2017); *Cameraria ohridella* Deschka & Dimic (d'Costa *et al.*, 2014); *Etiella zinckenella* Treitschke (Agus *et al.*, 2012 and Adie & Krisnawati, 2017).

In case of *E. vittella* also, several attempts have been made to understand its host preference (Ernest, 1989; Karban *et al.*, 1997), wild crop relatives (Sankhyan and Verma 1997), antixenosis (Halder *et al.*, 2006; Sharma and Singh, 2010; Aziz *et al.*, 2012; Anitha and Karthika, 2018) through series of varietal screening studies (Kamakshi and Srinivasan, 2008;

Koujalagi *et al.*, 2009; Rajesh and Jat, 2009; Muthukumaran and Ganesan, 2017; Eswaran and Anbanandan, 2018). These studies revealed the importance of plant morphological traits that influence the ovipositional preference of *E. vittella*. Since, detailed studies to explore the association of host-plant morphological traits with the egg laying choice of *E. vittella* are limited particularly in wild crop relatives, the present study was carried out to understand the relationship between host plant morphological traits and oviposition preference of *E. vittella* on selected *Abelmoschus* species.

## MATERIAL AND METHODS

The present study was conducted at Division of Crop Protection, ICAR-Indian Institute of Horticultural Research (ICAR-IIHR), Hesaraghatta, Bengaluru, India (12°58'N, 77°35'E, 890 m. above sea level) during 2021-2022.

### Host Plant Maintenance

Seeds of selected wild species of *Abelmoschus* namely *Abelmoschus tetraphyllus* (Roxb. ex Hornem.) Hochr., *Abelmoschus tuberculatus* Pal & Singh and *Abelmoschus angulosus* Wall. ex Wight & Arn. along with the cultivated species (*Abelmoschus esculentus* L. (Moench)) involving Arka Anamika, IIHR 356, IIHR 358, IIHR 379, IIHR 394, IIHR 402, ACC 1685 were procured from the Division of Vegetable Crops, ICAR-IIHR, Bengaluru. The host plants were grown in polybags (6 x 8") containing standard pot mixture (Red soil - 40%; Coco peat - 30%; Farm yard manure -30%) without any pesticide application (Devakumar, 2018). To avoid insect pest infestation, regular water sprays were given at frequent intervals.

### Insect Culture Maintenance

Okra fruits infested with *E. vittella* larvae were collected from the experimental fields of ICAR-IIHR. The larvae were reared by providing fresh immature fruits of okra in plastic containers until pupation (13.63 x 8.25 x 4.88 cm). The emerged adult moths were collected and released into net cages (1 x 1 x 1 m) for mating. The gravid females were separated and used for ovipositional preference studies.

## Oviposition Assays

Selected host plants were arranged randomly in a net cage and exposed to *E. vittella* @ 20 gravid females/ 10 plants for 48 hrs. Observations were recorded on the number of eggs laid on each host plant. These assays were replicated twenty times simultaneously ( $n = 200$ ). Observations were also recorded on different morphological traits of host plants namely plant traits [plant height (PH, cm), number of branches/ plant (NB) and stem diameter (SD, cm)], leaf traits [number of leaves/ plant (NL), leaf length (LL, cm), petiole diameter (PD, cm)], fruit traits [number of fruits/ plant (NF), fruit length (FL, cm), fruit width (FW, cm), trichomes density on fruits (T,  $\text{cm}^2$ )]. The length and diameter of various plant parts were recorded by using measuring scale. The trichome density was assessed by marking an area of  $10 \text{ mm}^2$  on fruit surface and the total number of trichomes present were counted under an optical stereomicroscope (Leica M205A).

## Statistical Analysis

Data collected on plant traits, *viz.*, Plant height (PH, cm), number of branches/ plant (NB), stem diameter (SD, cm), number of leaves/ plant (NL), leaf length (LL, cm), petiole diameter (PD, cm), number of fruits/ plant (NF), fruit length (FL, cm), fruit width (FW, cm), trichomes density on fruits (T,  $\text{cm}^2$ ) along with number of eggs laid were analysed using one way ANOVA as per Little and Hills (1978). The data were further subjected to correlation analysis and the correlation coefficient values were plotted in corplot using R 4.2.0. Path-coefficient analyses between the plant traits and number of eggs laid were carried out. To get a further insight, a step-wise regression procedure (Ryan, 1997) was employed to select the most crucial plant traits influencing variability in egg laying choice of *E. vittella*. This technique consisted of essentially identifying, stage by stage, trait(s) significantly related to egg laying choice (y). Further, as a measure of goodness-of-fit of the models developed, values pertaining to Co-efficient of Determination ( $R^2$ ) (Agostid'no and Stephens, 1986) were calculated. Variance Inflation Factor

(VIF) value was computed to test the multicollinearity of variables.

## RESULTS AND DISCUSSION

Among all the host plants studied, significantly highest plant height was recorded in ACC 1685 (139.20 cm) and lowest in *A. tuberculatus* (22.30 cm) ( $F_{9,190} = 139.0$ ;  $P < 0.0001$ ). Significantly a greater number of branches per plant were recorded in *A. angulosus* (34.00) and minimum number of branches were recorded in *A. tuberculatus* (1.00) ( $F_{9,190} = 520.5$ ;  $P < 0.0001$ ). Similarly, highest number of leaves per plant was recorded in *A. angulosus* (152.00) and lowest number recorded in *A. tuberculatus* (8.00) ( $F_{9,190} = 368.9$  and  $P < 0.0001$ ). Maximum stem diameter was recorded in ACC 1685 (2.30 cm) and minimum on *A. angulosus* (0.70 cm) ( $F_{9,190} = 140.40$ ;  $P < 0.0001$ ). Significantly maximum length of leaf was observed on ACC 1685 (23.10 cm) and minimum was observed on *A. tuberculatus* (7.80) ( $F_{9,190} = 80.50$  and  $P < 0.0001$ ). Petiole diameter was significantly maximum in ACC 1685 (0.70 cm) and minimum in *A. angulosus* and *A. tetraphyllus* (0.20 cm) ( $F_{9,190} = 39.68$  and  $P < 0.0001$ ). Significantly a greater number of fruits per plant were observed in *A. tetraphyllus* (64.00) and less in *A. tuberculatus* (4.00) ( $F_{9,190} = 165.90$ ;  $P < 0.0001$ ). Highest fruit length was recorded on IIHR 358 (23.90 cm) and lowest was recorded on *A. tuberculatus* (2.10 cm) ( $F_{9,190} = 427.20$  and  $P < 0.0001$ ). Maximum fruit width was observed in ACC 1685 (2.30 cm) and minimum was observed in *A. angulosus* (0.60 cm) ( $F_{9,190} = 218.4$  and  $P < 0.0001$ ). Trichomes density were observed highest on *A. angulosus* (24  $\text{cm}^2$ ) and lowest on *A. tuberculatus* (4  $\text{cm}^2$ ) ( $F_{9,190} = 714.6$  and  $P < 0.0001$ ). The number of eggs laid by *E. vittella* was significantly more on Arka anmaika (45.00) and lowest on *A. tuberculatus* (4.00) ( $F_{9,190} = 96.38$  and  $P < 0.0001$ ) (Table 1).

## Correlation Analysis

Of all the variables analysed, plant traits namely plant height (PH) and stem diameter (SD) showed a significant positive relationship with the number of eggs laid by *E. vittella* (PH:  $r = 0.44$ ,  $P < 0.01$ ;

TABLE 1  
Descriptive statistics of morphological traits of different *Abelmoschus* genotypes and number of eggs laid by *E. vittella* in choice assay

Host plants	Plant traits			Leaf traits			Fruit traits				Eggs laid (No.)
	Plant height (cm)	Branches / Plant (No.)	Stem diameter (cm)	Leaves/ plant (No)	Leaf length (cm)	Petiole diameter (cm)	Fruits/ Plant (No)	Fruit length (cm)	Fruit width (cm)	Trichomes density (cm <sup>2</sup> )	
<i>A. angulosus</i>	59.70±0.70 (44.8 - 66.2)	28.6 ±0.74 (21 - 34)	0.8±0.08 (0.7 - 1.9)	109.25±1.52 (77 - 152)	14.42±0.28 (11.9 - 17.1)	0.3±0.10 (0.2 - 0.5)	42.45±0.85 (32 - 52)	3.72±0.17 (3 - 4.2)	0.86±0.15 (0.6 - 1.1)	148.15±0.75 (128 - 164)	14.6±0.53 (12 - 17)
<i>A. tetraphyllus</i>	112.46±0.99 (99.4 - 132.1)	9.55±0.81 (4 - 14)	1.22±0.07 (1.1 - 1.4)	91.75±1.57 (64 - 120)	12.41±0.43 (8.4 - 14.8)	0.23±0.09 (0.2 - 0.3)	49.95±1.47 (29 - 64)	4.45±0.13 (4 - 4.9)	1.21±0.09 (1.1 - 1.4)	59.25±0.68 (49 - 68)	12.2±0.59 (7 - 14)
<i>A. tuberculatus</i>	31.59±0.85 (22.3 - 39)	1.00±0.00 (1 - 1)	1.64±0.02 (1.5 - 1.8)	11.40±0.53 (8 - 15)	8.96±0.13 (7.8 - 9.8)	0.41±0.02 (0.3 - 0.6)	8.05±0.48 (4 - 9)	2.83±0.09 (2.1 - 3.3)	1.02±0.04 (0.7 - 1.3)	29.05±0.74 (24 - 36)	6.95±0.38 (4 - 9)
<i>Arka anamika</i>	108.71±0.80 (94.3 - 121.3)	3.20±0.23 (3 - 4)	1.47±0.14 (1.1 - 1.8)	26.40±0.50 (23 - 32)	15.75±0.46 (11.4 - 18.2)	0.47±0.07 (0.4 - 0.5)	21.00±0.74 (14 - 26)	18.61±0.29 (16.7 - 21.1)	1.43±0.09 (1.3 - 1.6)	128.95±0.60 (112 - 139)	39.30±0.57 (34 - 45)
ACC 1685	116.06±1.76 (72.2 - 139.2)	3.70±0.42 (2 - 6)	2.07±0.19 (1.4 - 2.3)	26.90±0.66 (22 - 34)	19.90±0.40 (17.3 - 23.1)	0.60±0.09 (0.4 - 0.7)	19.05±0.67 (15 - 24)	14.93±0.47 (11.3 - 17.1)	2.14±0.08 (1.9 - 2.3)	98.25±0.58 (84 - 107)	22.60±1.65 (12 - 32)
IIHR 356	99.14±1.04 (74.3 - 121.2)	3.60±0.36 (3 - 6)	1.63±0.12 (1.3 - 1.9)	28.95±1.05 (21 - 39)	16.16±0.16 (14.3 - 17.1)	0.51±0.15 (0.3 - 0.7)	26.25±0.54 (21 - 32)	13.38±0.24 (11 - 14.5)	1.22±0.07 (1.1 - 1.3)	79.35±0.51 (71 - 86)	28.70±1.19 (17 - 36)
IIHR 358	100.70±1.12 (74.3 - 121.3)	3.75±0.41 (3 - 6)	1.73±0.07 (1.6 - 1.9)	28.05±1.06 (21 - 37)	15.59±0.28 (12.8 - 17.1)	0.53±0.13 (0.3 - 0.7)	19.55±0.74 (15 - 26)	21.20±0.37 (17.2 - 23.9)	1.14±0.06 (1.1 - 1.3)	116.60±0.60 (104 - 129)	37.70±0.54 (32 - 41)
IIHR 379	88.00±0.90 (63.6 - 96.3)	4.35±0.32 (3 - 6)	1.34±0.06 (1.2 - 1.4)	27.35±0.60 (24 - 34)	18.55±0.57 (14.1 - 22.1)	0.46±0.09 (0.4 - 0.6)	20.20±0.81 (13 - 24)	17.15±0.47 (12.3 - 19.3)	1.27±0.07 (1.1 - 1.4)	69.95±0.73 (62 - 79)	30.55±1.27 (22 - 41)
IIHR 394	88.30±0.75 (78.1 - 100.2)	3.35±0.27 (3 - 4)	1.28±0.07 (1.1 - 1.4)	20.10±0.59 (16 - 24)	14.93±0.37 (11.4 - 18.2)	0.46±0.13 (0.3 - 0.6)	12.75±0.64 (9 - 18)	14.83±0.58 (11.6 - 19.3)	1.20±0.06 (1.1 - 1.3)	68.40±0.88 (57 - 81)	27.40±0.90 (19 - 32)
IIHR 402	83.25±0.67 (71.3 - 96.4)	3.45±0.28 (3 - 4)	1.19±0.10 (1 - 1.4)	22.45±0.44 (19 - 26)	15.89±0.42 (12.8 - 18.3)	0.45±0.11 (0.3 - 0.6)	15.95±0.54 (12 - 9)	17.27±0.44 (13.4 - 20.1)	1.10±0.07 (1 - 1.2)	56.35±0.70 (50 - 68)	35.60±1.21 (24 - 44)

Figures in parentheses show the range of values

SD:  $r = 0.11$ ,  $P = 0.05$ ) and the number of branches per plant (NB) recorded a significant negative relationship ( $r = -0.34$ ,  $P < 0.01$ ).

Among the leaf traits studied, the leaf length (LL) ( $r = 0.52$ ,  $P < 0.01$ ) and petiole diameter (PD) ( $r = 0.43$ ,  $P < 0.01$ ) recorded a significant positive relationship with the number of eggs laid by *E. vittella*. However, the number of leaves per plant (NL) exhibited significant negative relationship ( $r = -0.40$ ,  $P < 0.01$ ).

In case of fruit traits studied, fruit length (FL) ( $r = 0.85$ ,  $P < 0.01$ ), fruit width (FW) ( $r = 0.14$ ,  $P < 0.05$ ) and trichomes density (T) ( $r = 0.30$ ,  $P < 0.01$ ) exhibited positive correlation with the number of eggs laid by okra shoot and fruit borer, *E. vittella*. However, the number of fruits per plant (NF) ( $r = -0.29$ ,  $P < 0.01$ ) showed significant negative relationship with number of eggs laid by *E. vittella* (Fig. 1).

## Regression Analysis

Considering the traits under multiple regression analysis, the host plant variables *viz.*, number of branches (NB), stem diameter (SD), leaf length (LL), fruit length (FL), fruit width (FW) and trichomes density (T) were further considered for multiple regression analysis (based on  $r/SE$ , a stringent criterion for identifying significant variables for regression analysis).

## Plant Traits

The simple linear regression analysis involving individual plant traits namely number of branches (NB) and stem diameter (SD) as independent variables, explained the variability in the number of eggs laid by *E. vittella* to the tune of 11 per cent and 1 per cent, respectively. Combining these two plant

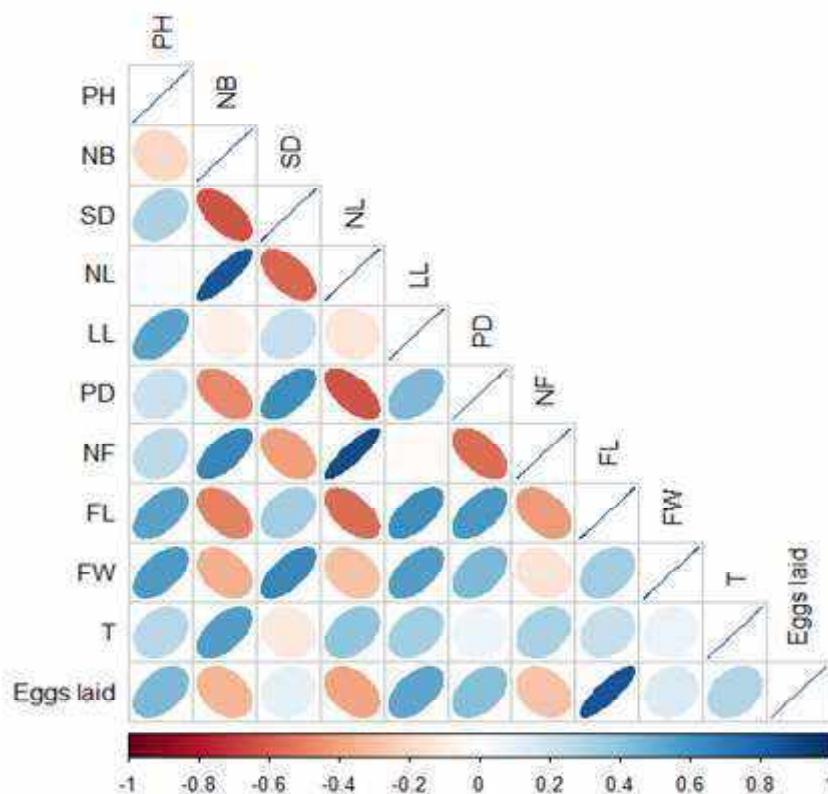


Fig. 1: Correlation analysis of number of eggs laid by *E. vittella* with all host plant morphological traits. The tendency is for the ellipses to be oriented from lower left to upper right indicates positive correlation and ellipse going from lower right to upper left indicates negative correlation. Width of the ellipses indicates strength of the correlation. The flat ellipses indicates strong correlation and blotted ellipses indicates weaker correlation. PH, Plant height (cm); NB, Number of branches/plant; SD, Stem diameter (cm); NL, Number of leaves/ plant; LL, Leaf length (cm); PD, Petiole diameter (cm); NF, Number of fruits/ plant; FL, Fruit length (cm); FW, Fruit width (cm); T, Trichomes density (cm<sup>2</sup>).



traits further explained a maximum of 13 per cent of the variability (Table 2).

**Leaf Traits**

The linear regression considering the leaf length (LL) which was the only significant independent variable

has explained the variability in the number of eggs laid by *E. vittella* to the tune of 27 per cent (Table 2).

**Fruit Traits**

The fruit traits namely fruit length (FL), fruit width (FW) and trichomes density (T) when factored in a

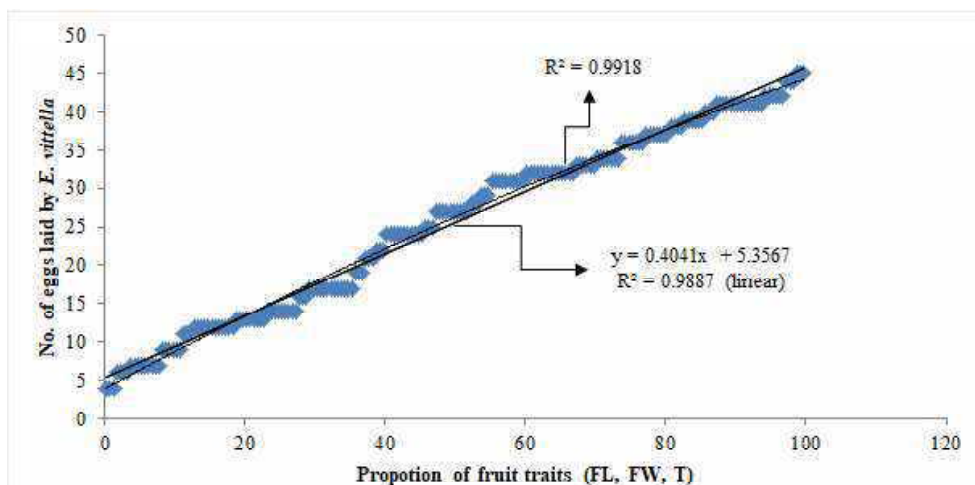


Fig. 2 : Relationship between the proportion fruit traits and number of eggs laid by *E.vittella*

TABLE 2  
Step-wise linear models to estimate *E. vittella* ovipositional preference using grouped plant traits

Variables (r/SE)	Correlation (r) with mean eggs laid	Model	R <sup>2</sup>	VIF
<i>Plant traits</i>				
NB	-0.34 **	Y=28.84-0.51 <sub>NB</sub>	0.11	-
SD	0.11	Y=20.40+3.60 <sub>SD</sub>	0.01	-
NB+SD	-	Y=37.62-0.66 <sub>NB</sub> -5.43 <sub>SD</sub>	0.13	1.15
<i>Leaf traits</i>				
LL	0.52 **	Y=-3.29+1.89 <sub>LL</sub>	0.27	-
<i>Fruit traits</i>				
FL	0.85 **	Y=5.89+1.53 <sub>FL</sub>	0.72	-
FW	0.14 **	Y=19.49+4.83 <sub>FW</sub>	0.02	-
T	0.30 **	Y=17.22+0.98 <sub>T</sub>	0.09	-
FL+FW	-	Y=11.97+1.64 <sub>FL</sub> -5.93 <sub>FW</sub>	0.75	3.98
FL+T	-	Y=3.34+1.49 <sub>FL</sub> +0.37 <sub>T</sub>	0.73	3.77
FW+T	-	Y=12.54+3.95 <sub>FW</sub> +0.09 <sub>T</sub>	0.10	1.11
FL+FW+T	-	Y=9.42+1.60 <sub>FL</sub> -0.59 <sub>FW</sub> +0.04 <sub>T</sub>	0.76	4.19

r- correlation coefficient; SE- standard error; NB - Number of branches/plant; SD - Stem diameter (cm); LL - Leaf length (cm); FL - Fruit length (cm); FW - Fruit width (cm); T - Trichomes density (cm<sup>2</sup>)

TABLE 3  
Step-wise linear regression models to estimate *E. vittella* ovipositional preference using combining of all host-plant morphological traits

Variables (r/SE)	Model	R <sup>2</sup>	VIF
NB+LL	$Y=0.91-0.45_{NB}+1.81_{LL}$	0.36	1.563
NB+FL	$Y=3.66+0.16_{NB}+1.63_{FL}$	0.73	3.726
NB+FW	$Y=27.81-0.5_{NB}+0.76_{FW}$	0.11	1.130
NB+T	$Y=12.74-1.10_{NB}+0.23_T$	0.46	1.841
SD+LL	$Y=-12.74-0.3_{SD}+1.9_{LL}$	0.27	1.372
SD+FL	$Y=14.04-6.84_{SD}+1.66_{FL}$	0.76	4.193
SD+FW	$Y=18.87+1.05_{SD}+4.12_{FW}$	0.02	1.024
SD+T	$Y=9.83+4.81_{SD}+0.1_T$	0.11	1.122
LL+FL	$Y=6.20-0.03_{LL}+1.54_{FL}$	0.72	3.613
LL+FW	$Y=-0.62+2.33_{LL}-7.43_{FW}$	0.30	1.435
LL+T	$Y=-4.46+1.73_{LL}+0.04_T$	0.29	1.400
NB+SD+LL	$Y=15.01-0.75_{NB}-10.75_{SD}+2.02_{LL}$	0.42	1.731
NB+SD+FL	$Y=15.18-0.04_{NB}-7.31_{SD}+1.65_{FL}$	0.76	4.198
NB+SD+FW	$Y=36.09-0.68_{NB}-8.92_{SD}+5.3_{FW}$	0.14	1.169
NB+SD+T	$Y=35.18-1.7_{NB}-16.2_{SD}+0.29_T$	0.59	2.421
NB+LL+FL	$Y=5.46+0.18_{NB}-0.19_{LL}+1.7_{FL}$	0.73	3.747
NB+LL+FW	$Y=7.85-0.65_{NB}+2.6_{LL}-14.2_{FW}$	0.46	1.854
NB+LL+T	$Y=0.52-0.93_{NB}+0.03_{LL}+0.18_T$	0.52	2.076
NB+FL+FW	$Y=10.08+0.1_{NB}+1.69_{FL}-5.45_{FW}$	0.75	4.040
NB+FL+T	$Y=3.24+0.03_{NB}+1.51_{FL}+0.03_T$	0.73	3.772
NB+FW+T	$Y=21.95-1.32_{NB}-8.55_{FW}+0.27_T$	0.50	2.012
SD+LL+FL	$Y=14.15-6.84_{SD}-0.01_{LL}+1.67_{FL}$	0.76	4.191
SD+LL+FW	$Y=-5.32+6.13_{SD}+2.46_{LL}-12.27_{FW}$	0.32	1.476
SD+LL+T	$Y=-5.15+0.62_{SD}+1.71_{LL}+0.04_T$	0.29	1.401
SD+FL+FW	$Y=14.92-5.38_{SD}+1.68_{FL}-2.56_{FW}$	0.76	4.248
SD+FL+T	$Y=11.79-6.33_{SD}+1.62_{FL}+0.02_T$	0.77	4.274
SD+FW+T	$Y=9.60+4.11_{SD}+1.1_{FW}+0.1_T$	0.11	1.122
LL+FL+FW	$Y=8.88+0.41_{LL}+1.54_{FL}-7.45_{FW}$	0.76	4.092
LL+FL+T	$Y=5.09+0.18_{LL}+1.54_{FL}+0.04_T$	0.74	3.789
LL+FW+T	$Y=-1.77+2.16_{LL}-6.88_{FW}+0.04_T$	0.31	1.456
NB+SD+LL+FL	$Y=14.98-0.05_{NB}-7.38_{SD}+0.03_{LL}+1.63_{FL}$	0.76	4.200
NB+SD+LL+FW	$Y=12.46-0.74_{NB}-4.49_{SD}+2.55_{LL}-11.57_{FW}$	0.47	1.880
NB+SD+LL+T	$Y=22.86-1.55_{NB}-17.11_{SD}+1.14_{LL}+0.23_T$	0.66	2.967
NB+SD+FL+FW	$Y=15.56-0.02_{NB}-5.7_{SD}+1.67_{FL}-2.47_{FW}$	0.76	4.250
NB+SD+FL+T	$Y=18.77-0.49_{NB}-10.37_{SD}+1.33_{FL}+0.09_T$	0.78	4.630

Contd....

Variables ( <i>r</i> /SE)	Model	R <sup>2</sup>	VIF
NB+SD+FW+T	$Y=35.61-1.71_{NB}-15.28_{SD}-1.58_{FW}+0.29_T$	0.59	2.428
NB+LL+FL+FW	$Y=8.58+0.04_{NB}+0.35_{LL}+1.57_{FL}-7.05_{FW}$	0.76	4.097
NB+LL+FL+T	$Y=5.09+0.05_{NB}-0.19_{LL}+1.58_{FL}+0.03_T$	0.74	3.794
NB+LL+FW+T	$Y=8.88-1.24_{NB}+1.88_{LL}-17.2_{FW}+0.2_T$	0.66	2.966
NB+FL+FW+T	$Y=10.93-0.18_{NB}+1.47_{FL}-6.91_{FW}+0.07_T$	0.76	4.254
SD+LL+FL+FW	$Y=13.06-4.76_{SD}+0.2_{LL}+1.63_{FL}-3.69_{FW}$	0.77	4.272
SD+LL+FL+T	$Y=12.77-6.26_{SD}-0.11_{LL}+1.65_{FL}+0.03_T$	0.77	4.283
SD+LL+FW+T	$Y=-7.54+7.09_{SD}+2.27_{LL}-12.32_{FW}+0.04_T$	0.34	1.511
SD+FL+FW+T	$Y=12.47-4.38_{SD}+1.64_{FL}-3.22_{FW}+0.03_T$	0.77	4.363
LL+FL+FW+T	$Y=7.80+0.26_{LL}+1.54_{FL}-6.95_{FW}+0.03_T$	0.76	4.234
NB+SD+LL+FL+FW	$Y=14.13-0.03_{NB}-5.40_{SD}+0.27_{LL}+1.58_{FL}-3.86_{FW}$	0.77	4.286
NB+SD+LL+FL+T	$Y=18.13-0.52_{NB}-10.67_{SD}+0.11_{LL}+1.28_{FL}+0.09_T$	0.78	4.640
NB+SD+LL+FW+T	$Y=20.36-1.53_{NB}-11.06_{SD}+1.65_{LL}-11.10_{FW}+0.23_T$	0.71	3.391
NB+SD+FL+FW+T	$Y=19.44-0.49_{NB}-8.43_{SD}+1.34_{FL}-3.21_{FW}+0.1_T$	0.79	4.735
NB+LL+FL+FW+T	$Y=8.82-0.32_{NB}+0.53_{LL}+1.26_{FL}-9.60_{FW}+0.77_T$	0.77	4.394
SD+LL+FL+FW+T	$Y=11.61-4.11_{SD}+0.11_{LL}+1.61_{FL}-3.79_{FW}+0.03_T$	0.77	4.371
NB+SD+LL+FL+FW+T	$Y=17.29-0.61_{NB}-8.17_{SD}+0.48_{LL}+1.16_{FL}-5.73_{FW}+0.11_T$	0.79	4.870

*r*- correlation coefficient; SE- standard error; NB - Number of branches/plant; SD - Stem diameter (cm); LL- Leaf length (cm); FL-Fruit length (cm); FW - Fruit width (cm); T - Trichomes density (cm<sup>2</sup>).

linear equation individually explained 72 per cent, 2 per cent and 9 per cent of the variability in the number of eggs laid by *E. vittella*, respectively.

Of all variables studied, the fruit length (FL) had the highest *R*<sup>2</sup> value of 0.72 ( $Y=5.89 + 1.53_{FL}$ ;  $R^2 = 0.72$ ) among all significant traits studied based on *r*/SE. Meanwhile the combination of these fruit traits enhanced and explained the variability by 76 per cent (Table 2).

### Combining of all Host Plant Morphological Traits

Step-wise regression analysis of all significant host-plant morphological traits (based on *r*/SE) explained the variability in the number of eggs laid in the range of 2-79 per cent with an acceptable VIF values (1.024-4.870, <10.0) indicating lack of multi-collinearity. The regression equation explained maximum of 79 per cent of the variability in the number of eggs laid by *E. vittella* by combining all the host plant morphological traits ( $Y=17.29 - 0.61_{NB} - 8.17_{SD} + 0.48_{LL} + 1.16_{FL} - 5.73_{FW} + 0.11_T$ ,  $R^2 = 0.79$ ).

Thus, combining all the plant variables could enhance the regression coefficient of determination by just 7 per cent (79%) compared to the single independent variable, fruit length (FL) that explained to the tune of 72 per cent. Therefore, the trait fruit length (FL) is quite influencing individual trait compared to other traits (Table 3).

The results of the polynomial models of different orders [(2), (3), (4), (5) and (6)] with all significant fruit traits like fruit length, fruit width and trichomes (based on *r*/SE) increased the coefficient of determination to the maximum of 99 per cent [ $R^2 = 0.9918$ ,  $R^2 = 0.9926$ ,  $R^2 = 0.9933$ ,  $R^2 = 0.9943$ , for polynomial model orders (2), (3), (4) and (5), respectively]. Plotting the residuals observed and estimated number of eggs laid by *E. vittella* using the three fruit traits (FL, FW and T) showed a random dispersal of points across x-axis (Fig.3). Similarly, polynomial models of different orders involving fruit length alone could explain the variability in the egg number to the tune of  $R^2 = 0.7257$ ,  $R^2 = 0.7261$ ,

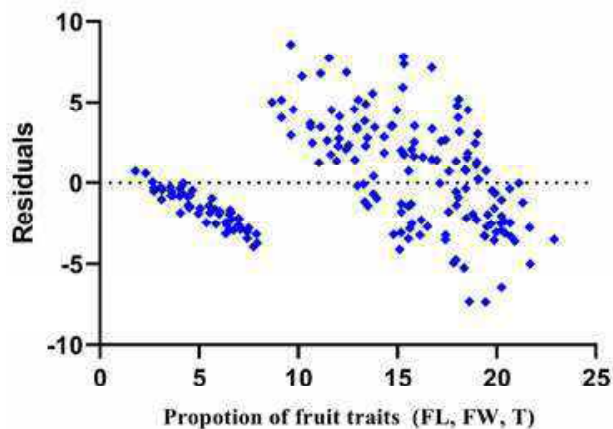


Fig. 3 : Plot of the residuals against the proportion fruit traits

TABLE 4  
Direct and indirect effects of plant traits on eggs laid by *E. vittella*

Pathways of association	Direct effects	Indirect effects	'r'
Branches/ Plant (No.)			-0.34 **
a. Direct effect	-0.38		
b. Indirect effect via			
Stem diameter (cm)		0.15	
Leaf length (cm)		-0.01	
Fruit length (cm)		-0.33	
Fruit width (cm)		0.06	
Trichomes density (cm <sup>2</sup> )		0.17	
Stem diameter (cm)			0.11
a. Direct effect	-0.25		
b. Indirect effect via			
Branches/ Plant (No.)		0.24	
Leaf length (cm)		0.03	
Fruit length (cm)		0.23	
Fruit width (cm)		-0.10	
Trichomes density (cm <sup>2</sup> )		-0.04	
Leaf length (cm)			-0.40 **
a. Direct effect	0.12		
b. Indirect effect via			
Branches/ Plant (No.)		0.03	
Stem diameter (cm)		-0.06	
Fruit length (cm)		0.42	
Fruit width (cm)		-0.09	
Trichomes density (cm <sup>2</sup> )		0.11	

Pathways of association	Direct effects	Indirect effects	'r'
Fruit length (cm)			0.85 **
a. Direct effect	0.68		
b. Indirect effect via			
Branches/ Plant (No.)		0.19	
Stem diameter (cm)		-0.08	
Leaf length (cm)		0.07	
Fruit width (cm)		-0.05	
Trichomes density (cm <sup>2</sup> )		0.07	
Fruit width (cm)			0.14 *
a. Direct effect	-0.16		
b. Indirect effect via			
Branches/ Plant (No.)		0.14	
Stem diameter (cm)		-0.16	
Leaf length (cm)		0.06	
Fruit length (cm)		0.23	
Trichomes density (cm <sup>2</sup> )		0.03	
Trichomes density (cm <sup>2</sup> )			0.30 **
a. Direct effect	0.30		
b. Indirect effect via			
Branches/ Plant (No.)		-0.21	
Stem diameter (cm)		0.03	
Leaf length (cm)		0.04	
Fruit length (cm)		0.15	
Fruit width (cm)		-0.01	

$R^2 = 0.7369$ ,  $R^2 = 0.7408$  and  $R^2 = 0.7419$  by orders (2), (3), (4), (5) and (6), respectively.

**Path Co-efficient Analysis**

To reveal direct and indirect association between significant ( $r/SE$ ) traits and number of eggs laid by *E. vittella* were studied under path-coefficient analysis (Table 4). The results showed that direct effect of number of branches on number of eggs laid by *E. vittella* was highly negative (-0.38). Besides, indirect effects through other traits, viz., stem diameter (0.15), fruit width (0.06) and trichomes density (0.17) was positive and of a reasonable magnitude. However, it exhibited a negative, indirect effect through leaf length (-0.01), fruit length (-0.33). Stem diameter exhibited moderate, negative, direct effect (-0.25) as well as indirect effects via., fruit

width (-0.10), trichomes density (-0.04). However, it exhibited a positive indirect effect through number of branches (0.24), leaf length (0.03) and fruit length (0.23).

Leaf length showed a moderate, positive, direct effect (0.12) besides indirect effects via number of branches (0.03), fruit length (0.42), trichomes density ( $\text{cm}^2$ ) (0.11) were found to be positive. However, it exhibited negative indirect effect through stem diameter (-0.06) and fruit width (-0.09).

Direct effect of the fruit length on number of eggs laid by *E. vittella* was positive and high in magnitude (0.68). The total correlation between fruit length and number of eggs laid by *E. vittella* was highly positive and significant (0.85). Indirect effect of the fruit length via other traits, *viz.*, number of branches (0.19), leaf length (0.07) and trichomes density (0.07) was positive and of a reasonable magnitude, contributing to the total correlation coefficient. However, indirect effect through stem diameter (-0.08) and fruit width (-0.05) was found to be negative.

Fruit width exhibited a negative, direct effect of moderate magnitude (-0.16) but showed a positive indirect effect through number of branches (0.14), leaf length (0.06), fruit length (0.23), trichomes density (0.03). However, stem diameter (-0.16) exhibited a negative indirect effect. Trichomes density showed a positive, direct effect of high magnitude (0.30). Indirect effects *via* stem diameter (0.03), leaf length (0.04) and fruit length (0.15) found to be positive. However, it exhibited a negative indirect effect through number of branches (-0.21) and fruit width (-0.01).

Host plant resistance mechanism defines insect preference and non-preference based on collective heritable plant traits (Bernays and Chapman, 1994). The earliest stage of resistance in insect-plant relationship is resistance to oviposition (= oviposition antixenosis) exhibited by the plants, where insects try to avoid these host plants to lay their eggs. However, oviposition is a sequel of act that heavily depends on variable plant traits (Beck, 1965 and War & Sharma, 2014). The aim of the current study was to determine

how the *Abelmoschus* spp. host plant morphological traits influence the number of eggs laid by okra fruit and shoot borer, *E. vittella*.

Of all traits studied, we found that host plant traits namely number of branches (NB), stem diameter (SD), leaf length (LL), fruit length (FL), fruit width (FW) and trichomes density (T) were found to be most influencing host plant traits that affect the oviposition by *E. vittella* based on  $r/SE$ .

Multiple regression analysis was further employed to find optimized equations for the association between number of eggs laid by *E. vittella* and minimum number of host plant traits with a reasonable  $R^2$  value. Step-wise linear regression equations showed that combination of all significant ( $r/SE$ ) host-plant traits could explain the variability in the number of eggs laid by *E. vittella* to the tune of 79 per cent. However, consideration of only single trait, the fruit length (FL) found to be the potent trait than other host plant traits as it could explain the maximum variability ( $R^2 = 0.72$ ).

Similar findings were observed earlier also where the length of fruits was more crucial for infestation by *E. vittella* as they harboured a greater number of eggs (Halder *et al.*, 2015). Likewise, the pod borer, *Maruca vitrata* infestation level was high with the longest pod length as it harboured a greater number of eggs in cowpea (Halder and Srinivasan, 2011). Step-wise linear equations showed that every equation improved the coefficient of determination ( $R^2$ ) when it has fruit length as one of the variables (Table 2 & 3). The present study also endorses previous findings where it was found that fruit length along with trichomes density as potential host-plant traits which influenced the number of eggs laid by *E. vittella* (Sultani *et al.*, 2011; Muthukumar & Ganesan, 2017 and Anitha & Karthika, 2018).

Several studies were carried out to understand the effect of host plant traits on the oviposition of lepidopteran pests namely leaf miner, *Liriomyza huidobrensis* (Blanchard) on potato (Videla and Valladares, 2007), the gram pod borer, *Helicoverpa armigera* (Hubner) on groundnut (War *et al.*, 2013),

the South American tomato pinworm, *Tuta absoluta* (Meyrick) on tomato (Cherif and Verheggen, 2019) and the pink boll worm, *Pectinophora gossypiella* (Saunders) on Cotton (Madhu and Mohan, 2021).

Brinjal stem borer (*Euzophera perticella* Rag.) had positive significant correlation with stem diameter, number of branches per plant and plant height while negative significant correlation with number of hair/cm<sup>2</sup>. It was observed that with an increase in plant height, stem diameter and number of branches per plant, there was a significant increase in infestation. On the other hand, the cultivars with maximum number of hair/cm<sup>2</sup> showed a decrease in infestation (Javed *et al.*, 2017).

All the host-plant traits that influence the egg laying choice of *E. vittella*, fruit traits must be taken as prime consideration since it is fruit borer. The correlation study between Okra fruit infestation and morphological factors implied that primary branching and trichome length adversely affect the oviposition (Kumar *et al.*, 2021). Another study revealed that the parameters such as fruit length, seed, fruit hairs and diameter were non-significant with *E. vittella* infestation in Okra (Gautam *et al.*, 2013). The density and higher length/ breadth of trichomes adversely affected the *E. vittella* infestation. Similarly, fruit angle to stem also adversely influenced the preference of fruit borer, *E. vittella*. Fruit length found to have positive influence on fruit borer infestation (Muthukumaran and Ganesan, 2017).

In the present study, the host-plant traits namely number of branches, stem diameter, leaf length, fruit length, fruit width and trichomes density which influenced the okra shoot and fruit borer, *E. vittella* egg laying choice upto 79 per cent can be considered as potential traits while searching for ovipositional antixenosis in germplasm. Majorly, fruit traits (fruit length, fruit width and trichomes on fruit) must be taken into prime consideration since *E. vittella* egg laying site is mainly fruit. Further, understanding the olfaction basis of *E. vittella* egg laying choice and identifying potent chemical cues among the selected germplasm apart from host plant traits will aid in greater understanding of potential chemical stimuli

which influences *E. vittella* oviposition and its egg laying choice in toto.

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## Determinants of Preference for Marketing Channels : An Economic Analysis of Vegetable Growers in Chikkaballapur District

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### ABSTRACT

This study has evaluated the factors that influence the farmer's preference of marketing channels for vegetables in Chikkaballapur district, Karnataka. The marketing channels include traditional and modern marketing channels. The primary data were collected from 50 vegetable growing farmers practicing traditional channels using random sampling and 50 vegetable growing farmers who prefer modern marketing channels using snow ball sampling technique. Three major vegetable were chosen for in-depth analysis based on the area dominance. Personal interview method was used to collect the data with the help of semi-structured schedule. Nine variables describing the socio economic characters of the respondents and market factors were considered for the analysis. It was observed during the survey that farmers were practicing many marketing channels. The probit regression was used to identify the factor influencing preference of a particular marketing channel. Empirical findings revealed that factors such as age, farm size, vegetable area, selling price were found to be significantly influencing the choice of channel for all the three selected vegetables. It was interesting to note that the relatively large farm size provided a win-win situation for farmers practicing modern retail marketing channel with planned cultivation and marketing of vegetables, as per the indent received from modern retail outlets. Further, these farmers devoted relatively sizeable area to realise benefits of large economies of scale from flower cultivation than other farmers practicing traditional marketing channel. Distance to the modern market negatively influenced in choosing modern markets by the farmers. There is a need to establish more and more collection centres so as to benefit more number of farmers in the region from these modern retail marketing channel.

*Keywords* : Modern marketing channel, Vegetable growers, Probit model

**A**GRICULTURE is considered as the backbone of Indian economy as it provides food to its million mouths and the raw materials to growing industrial base. The demand for vegetables and fruits is increasing due to awareness about the nutritional and protective nature of these commodities. Vegetables are important constituents of Indian agriculture and nutritional security due to their short duration, high yield and economic viability, generation of on-farm and off-farm employment and nutritional richness.

Our country is blessed with diverse agro-climates with distinct seasons, making it possible to grow wide array of vegetables.

India ranks second in vegetables production in the world, after China with nearly 13 per cent in world's vegetable production. As per FAO statistics, India produced 196.26 million metric tonnes of vegetables from 10.73 million hectares during 2020-21. The production share of vegetables was 59.50 per cent in

total horticultural production during the year 2020-21. Vegetable production was more than doubled in the past decade from about 89 million MT. Amongst vegetables, India is the second largest producer of potato, onion, cauliflower, brinjal, cabbage, *etc.* The vast production base offers India tremendous opportunities for export too.

Marketing of vegetables is particularly important as up to 90-98 per cent of the produce is marketable surplus, except root and tuber crops of which a significant portion is saved for seeds (Singh and Sikka, 1992). The marketing operations of vegetables have a crucial role, due to seasonality of produce, which largely determines the profits of the farmer on one hand and level of availability to consumer on the other hand. The strengthening of Indian agriculture base can be achieved not only by increasing the farm production through new production technologies, but also even to a larger extent through addressing issues in post-harvest handling and distribution by linking farmers to markets. The decision to sell in any channel depends upon many factors than merely price and returns criteria. For instance, selection of the marketing channel depends on information related to product attributes, prices, cost and consumers' demand.

Most of the vegetables are perishable and are highly prone to production and market risks, which may act as deterrent to farmers participation in their cultivation. Local markets for high value commodities are thin, coupled with lower marketable surplus of individual small holder's to be traded remuneratively in distant urban markets due to high transaction costs, need to be addressed though research. The vegetable prices are volatile and fall drastically even with a small change production and market arrivals. Institutional innovations in marketing, enhance farmer's access to quality inputs, improved technology, information and other advisory services which eventually lead to improvement in productivity and reduction in marketing and transaction costs (Hanumanthaiah and Aparna, 2010).

In India the development of organized retail chains had deeply influenced fruit and vegetable marketing system. India is observing growth in organized retailing through the participation of large corporate

firms. Retail industry in India is expected to grow to US\$ 1.3 trillion by 2022, with a Compound Annual Growth Rate of 16.7 per cent over 2015-20, more importantly India is the fifth largest preferred retail destination globally. The country is among the highest in the world in terms of per capita retail store availability. India's retail sector is experiencing exponential growth, with retail development taking place not just in major cities and metros, but also in Tier-II and Tier-III cities. This trend can be closely related with growing urbanization, expanding consumerism and growing number of upper middleclass and high-income households. These retail chains bring in quality culture, instant demands and supply and more commercial nature of production and marketing at the farmer level (Singh and Singh, 2015).

Understanding the factors affecting the market choice (*i.e.* farmer's decision to sell in different marketing channels) is important and can be used to guide farmers, farm investment decision, and market channel development. Further, this also directs in formulating the strategic plans, policies for farmers inclusiveness to the marketing and development of market abilities. In this study, the factors that influence the farmers marketing choices for vegetables in different marketing channels have been evaluated. Co-existence of different marketing channels appears to support producers by providing convenient access to a range of price and quality service combinations. Marketing of vegetables is not a mere selling of the produce; it also includes the value added activities associated with post-harvest quality maintenance, according to the market channel requirement and unit prices for the produce. Market prices vary greatly with produce quality and market location. Markets are chosen not only with price consideration but also on the basis of services offered and their proximity (Roopa *et al.*, 2018). Choice of channel is greatly influenced by various factors such as farmer's socio-characteristics, location-specific attributes such as distance to markets, competition among the trader-buyers, consumer preference and their attributes.

## Description of Vegetable Marketing Channels under Study Area

### Traditional Marketing Channels

1. Farmer - APMC - Wholesaler - Retailer - Consumer
2. Farmer - APMC - Retailer - Consumer
3. Farmer - APMC - Consumer
4. Farmer - Trader - APMC - Retailer - Consumer

### Modern Market Channel

These marketing channels which differ from the traditional channels indicated above and following are the modern retail marketing channels.

1. Farmer - Collection centre - Retail outlet - Consumer
2. Farmer - Trader - Collection centre - Retail outlet - Consumer

## Sources of Data and Analytical Framework

### Conceptual Framework

The conceptual model on marketing explains farmer's marketing behaviour. In marketing of agricultural commodities, selection of marketing channel has bearing on many other decisions, as different channels are characterized by diverse institutional attributes. It is assumed that selection of more than one channel by a farmer maximizes the returns and farmers would also practice it, depending upon the nature of the commodity, availability and accessibility to marketing agency, expected additional benefits and his knowledge. In this study, the farmers are classified into two categories as Farmers participating in Traditional Marketing Channels (FTMC) and Farmers participating in Modern Marketing Channels (FMMC). The FTMC are those who are selling more than 70 per cent of their produce in the traditional marketing channel. The FMMC farmers are those who are selling more than 70 per cent of their produce to the modern marketing channel, as it is needless to mention that we can't get the those category of farmers relying on a single marketing channel. The marketing decision on preference for a particular marketing

agency of a channel depends on many factors like his personal socio-economic characteristics of the farmers, availability of different market choices prices received, degrees of relationship among marketing firms with respect to competition, enforcement mechanisms of the trader including market infrastructure like transportation facility, market lead extension services, government regulations, of the marketing channel.

### Study Area and Sample Design

The study was carried out in Chikkaballapur district, as it was the leading district in vegetable cultivation in Southern part of Karnataka and had greater opportunity to fulfill the large demand for vegetables from nearby huge consumer market and modern retailing prevalent in Bengaluru. To achieve the objective of the study, the required primary data from 50 farmers participating in Traditional Marketing Channels (FTMC), as defined earlier whose produce moves onwards in traditional retail format were selected using random sampling technique, while equal number of 50 farmers participating in Modern Marketing Channels (FMMC), using Snow ball sampling technique whose produce would reach the consumers through the modern retail outlet traced with the help of the organized retail firms. Thus a total sample size for the present study was 100 vegetable growers. The data were collected from the respondents through personal interview method using pre-tested, well-structured schedule. The cluster of villages were chosen randomly which were participant in the modern retail outlet and FTMC respondents were also



Fig. 1: Karnataka State and Chikkaballapur district showing study area

chosen from the same villages so as to ensure homogeneity in the sample except vegetable marketing practice. The required information about socio-economic character of the respondents like age, education level, land holdings, cropping pattern, details of marketing practices pertained to the agricultural year 2021-22.

### Analytical Tools used

The data were analysed using the measure of central tendency and various functional forms as detailed below. The commonly used measures like averages, frequency and per cent were used to synthesize data for functional analysis.

a) *Probit Model* : The empirical specification of market choices can be modelled through probit regression analysis. The probit model is a statistical probability model with two categories in the dependent variable. Probit analysis is based on the cumulative normal probability distribution. The binary dependent variable,  $y$ , takes on the values of zero and one. The outcomes of  $y$  are mutually exclusive and exhaustive. The dependent variable,  $y$ , depends on  $k$  observable variables  $X_k$  where  $k = 1, \dots, K$ . While the values of zero and one were observed for the dependent variable in the probit model, there was a latent, unobserved continuous variable,  $y^*$ .

$$y^* = \sum_{k=1}^K \beta^k X^k + \varepsilon \quad (1)$$

$\varepsilon$  is normally distributed with  $(0, \sigma^2)$

The dummy variable,  $y$ , was observed and was determined through  $y^*$  as explained below :

$$y = \{ 1 \text{ if } y^* > 0, 0 \text{ otherwise} \} \quad (2)$$

The point of interest relates to the probability that  $y$  equals one. From the above equations,

$$\begin{aligned} \text{Prob}(y = 1) &= \text{Prob}\left(\sum_{k=1}^K \beta_x X_k + \varepsilon > 0\right) \\ &= \text{Prob}\left(\varepsilon > -\sum_{k=1}^K \beta_x X_k\right) \\ &= 1 - \Phi\left(-\sum_{k=1}^K \beta_x X_k\right) \end{aligned} \quad (3)$$

Where  $\Phi$  is the cumulative distribution function of  $\varepsilon$

The probit model assumes that the data were generated from a random sample of size  $n$  with a sample observation denoted by  $i$ , for  $i = 1, \dots, N$ . Thus the observations of  $y$  must be statistically independent of each other to rule out possible serial correlation. Additionally, it was assumed that the independent variables chosen were random variables.

The Maximum Likelihood Estimation (MLE) technique was used to estimate probit model parameters. MLE focused on choosing parameter estimates that gives the highest probability or likelihood of obtaining the observed sample  $y$ . The main principle of MLE was to choose as an estimate of  $\beta$  for set of  $K$  numbers that would maximize the likelihood of having observed this particular  $y$  (Aldrich and Nelson, 1984).

The probit model for the study was specified as below

$$Y_{ki}^* = \beta_{k0} + \beta_{k1} X_1 + \beta_{k2} X_2 + \beta_{k3} X_3 + \beta_{k4} X_4 + \beta_{k5} X_5 + \beta_{k6} X_6 + \beta_{k7} X_7 + \beta_{k8} X_8 + \beta_{k9} X_9 + \beta_{k10} X_{10} + \beta_{k11} X_{11} + \varepsilon$$

Where,

$Y$  = Dependent variable (Channel Chosen), Binary variable with 1 for FMMC and 0 for TMMC

$X_1$  = Age of vegetable grower (years)

$X_2$  = Level of education (No. of formal years of education)

$X_3$  = household size (No.)

$X_4$  = Social participation (0-No participation, 1-Panchayat, 2-Co-operative, 3-SHG, 4-Others)

$X_5$  = Average farm size (acres)

$X_6$  = Area under the selected vegetable (acres)

$X_7$  = Credit facility by the members of channel (1 = Yes, otherwise 0)

$X_8$  = Access to extension service (1 = Yes, otherwise 0)

$X_9$  = Quality checking (1 = Yes, otherwise 0)

$X_{10}$  = Price realized in each channel (In Rs. / qtl)

$X_{11}$  = Distance to market (Kms)

In equation (4)  $Y_{ki}^*$  is a variable reflecting choice of a marketing channel by the  $i^{\text{th}}$  farmer with  $k$  denoting the market choice ( $k = 0, 1$ ), if  $k$  is 0 then farmer is

selling more than 70 per cent of the produce through the traditional market channel otherwise to modern marketing channel.

The probit model was used to estimate the impact of the independent variables on consumer behaviour regarding the sale of vegetables and to predict probabilities of change in producer’s channel choice under several simulated variable levels.

**b. Output-elasticities**

Marginal effects of the explanatory variables at the mean could be obtained by:

$$\text{Marginal effect of } X_i = \frac{dy}{dX_i} * \frac{\bar{X}_i}{\bar{Y}} \text{ (or) } b_i * \frac{\bar{X}_i}{\bar{Y}} \tag{5}$$

Where,

B = Parameter estimate (partial elasticity associated with each independent variable)

x = Mean of independent variable

y = Mean of dependent variable

**RESULTS AND DISCUSSION**

The descriptive statistics and the description of variables are reported in Table 1 and 2. The two choices available for the marketing include traditional marketing channel (FTMC) and modern marketing channel (FMMC) and were expressed in dummy variable. As mentioned earlier, value 1 was used if farmer sold through modern marketing channel otherwise zero. From the conceptual model, it is hypothesized that the decision of choosing a marketing channel choice depends on characteristics of respondents like socio-economic and market attributes like age (AGE), education (EDU), Household size (HHSIZE), Social participation (SOCP), farm size (FAMSIZE), area under vegetables (VEGAREA), Credit facility provided by the channel member (CREDFAC) Extension services provided by the personnel in the channel (EXTSERV), Quality checking at market place (QULICHECK),

TABLE 1  
Summary statistics of variables used in probit model- Modern market channel

Variable	Mean	Standard deviation	Maximum value	Minimum value	Expected sign
AGE - Age of the respondents(Years)	44.94	13.46	53.00	29.00	-
EDU -Education level (years of formal education)	11.00	1.33	18.00	7.00	+
HHSIZE - Size of the household (No.)	5.00	2.89	8.00	3.00	-
SOCP - Social participation (No.)					
No participation (8.00)					
Panchayat member (3.00)		1.23	4.00	0.00	+
Co-operative member (25.00)					
SHG member (10.00)					
Others (15.00)					
FARMSIZE -Size of farm (Acres)	4.40	3.08	18.00	1.00	+
VEGAREA -Area under the vegetable (Acres)	1.44	0.83	3.50	0.50	-
CREDFAC-Credit facility provided by the channel member (1=Yes, 0=otherwise)	0.26	0.44	1.00	0.00	-
EXTSERV- Extension service by the personnel in the channelb (1=Yes, 0=otherwise)	0.82	0.39	1.00	0.00	+
QULICHECK -Practice of quality checking (1=Yes, 0=otherwise)	1.00	1.23	1.00	0.00	+
PRICE -Price realized in each channel (Rs/Qtl)	2291.76	1586.57	4850.00	2140.53	+
DISTMRKT -Distance to market (Kms)	4.24	4.55	25.00	2.00	-

TABLE 2  
Summary of statistics of variables used in probit model- Traditional market channel

Variable description	Mean	Standard deviation	Maximum value	Minimum value	Expected sign
AGE - Age of the respondents (Years)	48.64	14.34	85.00	27.00	-
EDU -Education level (years of formal education)	8.07	0.98	13.00	3.00	+
HHSIZE - Size of the household (No.)	6.00	55.00	12.00	4.00	+
SOCP - Social participation (No.)					
No participation (30.00)					
Panchayat member (0.00)					
Co-operative member (10.00)		1.15	4.00	0.00	-
SHG member (5.00)					
Others (10.00)					
FARMSIZE - Size of farm (Acres)	2.73	3.30	16.70	1.50	-
VEGAREA -Area under the vegetable (Acres)	2.04	1.32	6.50	3.00	+
CREDFAC-Credit facility provided by the channel member (1=Yes, 0=otherwise)	0.62	0.49	1.00	0.00	+
EXTSERV- Extension service by the personnel in the channel (1=Yes, 0=otherwise)	0.40	0.69	1.00	0.00	-
QULICHECK -Practice of quality checking (1=Yes, 0=otherwise)	0.00	0.89	1.00	0.00	-
PRICE -Price realized in each channel (Rs/Qtl)	1613.907	12.04	3250.00	1520.00	-
DISTMRKT -Distance to market (Kms)	8.37	6.98	22.00	4.00	+

Price realised in each channel (PRICE), Distance to the market (DISTMRKT).

It could be observed from summary characteristics of the variables used in the probit model for modern and traditional farmer (Table 1 and Table 2) that the age was an important factor in choice of market channel. Younger farmer are more likely to choose modern marketing channel to dispose their output as average age of respondents in the modern marketing channel was 45 years for respondents in FMMC category which is less than age of the farmers in traditional marketing channel *i.e.*, 49 years.

Thus age was found to be inversely related to the choice of modern marketing channel. Education (Edu) level could be an important determinant as it would help to gather and take benefit of information flow through different means and aids in rational decisions. Literacy is also an indicator of managerial decisions

and marketing practices. The education level of the farmer measured in terms of total number of years of formal schooling. Descriptive statistics revealed that on an average, farmers in the category of FMMC have completed the higher secondary school whereas traditional retail farmers on an average have possessed primary education.

Thus possession of higher education ensured the farmers to take advantage of information, better managerial decisions and ultimately facilitated better marketing practices. Social participation by the farmers is also an important factor, which was observed with most of sample farmers in modern marketing channel like membership in co-operatives whereas the level of participation in such organisation with respect to traditional retail farmers was poor. Participation as member or office bearers in various social organisations will strengthen their social network and enabled them to timely access to information and better managerial decisions.

Farm size also turned out to be prominent factor in the literature of the marketing choice decisions. Farm size is used as a proxy for wealth of farmers. The literature revealed that in Indian context, wealth has an effect on the farmer's choice of place as wealthier farmers can take advantage of low transportation cost due to economies of large scale benefits both in input sourcing and output marketing, in addition they can have or wider social network helping in better market access and market facilities. Average land holding of traditional farmers (2.73 acres) was less compared to the modern farmers (4.40 acres). This indicates that the modern farmers are better-off than traditional farmers in reaping benefits of economies of scale. Further, vegetable farm size is also equally important, as it is an important determinant of vegetable production, investment and marketing decisions. Results presented in table also revealed average area under vegetable cultivation of 1.44 acres in the case of FMCC farmers and 2.04 acres for FTMC farmers. The reason behind this is that the FMCC practicing farmers with large holdings allocated higher area for cultivation of flowers like rose, chrysanthemum, marigold *etc.* which provide better year round income and savings. It was observed during the survey that respondents reported floriculture was more beneficial enterprise than vegetable cultivation. As vegetables generally associated with lump-sum mostly one time returns, which is not useful in managing recurring expenses. Another reason could be that due to very close and intimate contact of farmers practicing in FMCC retail format with personnel in retail outlet and collection centre, succeeded in getting more frequent, regular indents for the vegetables they produce. Credit facility (CREDFAC) is also important factor in choice of channel, as the agents in traditional market channels like commission agent, wholesaler and retailer provided credit facility in advance of crop season to meet expenses of farmers starting from sowing to various peak crop season, which might have compelled to some extent to sell through that agency. If the farmer is availing credit facility in the particular channel then value of dummy variable is one otherwise zero.

With advent of more number of modern retail outlet in metropolitan cities, farmers are getting relatively better marketing option for their produce in general and vegetables in particular. Strict quality checking and frequent lot rejection due to the poor quality in the modern marketing channel hinders the farmers to choose this channel. This variable (QUALICHECK) is taken as dummy variable with value one for prevalence of quality checking and zero otherwise. These channels also provide extension service in term of advisory role by the personnel in the channel. If any farmers availing advisory services from particular channel then the value is one otherwise zero. The average value of this is near to one in modern channel, it is due to the reason every modern outlet will be having a person placed may be horticulture specialist or agricultural graduates, who is assisting farmers in observing good agricultural practices (GAP) right from selection of variety, planting time, spraying schedule, harvesting schedule, *etc.* Price is also another major factor in choice of marketing channels and obviously, the channel with better price realisation would be preferred. Average price realised by the farmer in modern channel (Rs.2291.76/ qtl) was found to be comparatively higher (42%) compared to the traditional channel (Rs.1613.91/ qtl). Distance to the market also a factor which helps the farmers to choose between channels. Shorter the distance to collection centre higher the probability of farmers to for with and prefer modern channel and visa-versa. The average distance between farm and the collection centre in the case of modern format of retail market channel is 4.24 km and is 8.37 km in the case of FTMC, so the distance had positive relation with probability to choose modern market channel.

The estimates of the probit model about factors influencing the preference for marketing channel are presented in Table 3. The coefficients of these parameters indicate direction of association with dependent variable. Majority of the parameters listed in the model have shown positive influence of choice of channel. The pseudo  $r^2$  indicated that the model chosen for estimating choice of channel and its determinants was found to be good fit to the data as it worked to be very high per cent of explanation of

TABLE 3  
Estimates of probit model on factor affecting choice of channel between traditional and modern marketing channel for major vegetables

Variables	Parameters	Tomato		Potato		Cabbage	
		Co - efficient	P - Value	Co - efficient	P - Value	Co - efficient	P - Value
INTERCEPT	$\beta_0$	10.619 (22.585)	1.523	26.33 (12.22)	2.589	2.234 (2.619)	5.265
AGE	$\beta_1$	-3.138 * (1.440)	0.012	-2.633 ** (1.026)	0.022	-1.094 ** (0.015)	0.005
EDU	$\beta_2$	0.208 * (1.002)	0.045	1.88 (1.023)	1.253	4.706 * (1.206)	0.025
HHSIZE	$\beta_3$	-1.998 (0.671)	1.352	1.835 (5.41)	2.142	-4.024 * (-1.689)	0.012
SOCP	$\beta_4$	3.406 * (0.098)	0.042	-3.063 (1.079)	1.025	2.472 * (0.008)	0.032
FARMSIZE	$\beta_5$	1.294 ** (0.572)	0.001	3.198 ** (0.496)	0.007	1.194 (1.975)	0.125
VEGAREA	$\beta_6$	-2.812 ** (0.811)	0.007	-2.228 ** (2.035)	0.031	-4.115 * (-1.541)	0.032
CREFAC	$\beta_7$	-1.083 (2.969)	0.552	-1.070 * (0.034)	0.025	-2.806 * (-1.057)	0.039
EXTSERV	$\beta_8$	6.789 (10.828)	1.985	2.412 (3.523)	1.253	1.795 * (0.245)	0.009
QULITY	$\beta_9$	3.136 ** (1.181)	0.045	1.112 (2.740)	2.021	1.342 (1.957)	0.108
PRICE	$\beta_{10}$	2.016 ** (1.006)	0.003	3.094 ** (1.033)	0.015	3.098 ** (1.058)	0.024
DISTMARKT	$\beta_{11}$	-2.980 (4.403)	7.252	-1.062 (0.002)	2.562	-2.109 (0.059)	0.014
Pseudo r <sup>2</sup>			0.80		0.69		0.73

Figures in parenthesis indicate standard error; \*\* and \* indicates level of significance at one and five per cent level of probability

80 per cent, 69 per cent and 73 per cent in the case of tomato, potato and cabbage, respectively. The estimated coefficient for age and area under vegetables revealed negative and significant influences on the choice of channel for all selected vegetables. The farm size and price realised in each channels have positive and significant influence on choice of channel for all the three vegetables. Social participation by the farmer and prevalence of practice of quality checking found to have positive and significant influence in choosing marketing channel for tomato. Credit facility by the

agencies in traditional channel has also showed negative and significant impact on choosing the modern market channel for potato. House hold size has negative and significant influence on choice of channel for cabbage farmers. Whereas the extension services and social participation have positive and significant impact in the case of marketing practice of choosing MMC for cabbage. Although distance to the market exhibit negative relation with choice of channel but fail to emerge as significantly influencing factor.



The estimated coefficient for age and area under vegetables revealed negative and significant influences on the choice of channel for all selected vegetables. The farm size and price realised in each channels have positive and significant influence on choice of channel for all the three vegetables. Social participation by the farmer and prevalence of practice of quality checking found to have positive and significant influence in choosing marketing channel for tomato. Credit facility by the agencies in traditional channel has also showed negative and significant impact on choosing the modern market channel for potato. House hold size has negative and significant influence on choice of channel for cabbage farmers. Whereas the extension services and social participation have positive and significant impact in the case of marketing practice of choosing MMC for cabbage (Pavithra and Gaddi, 2022). Although distance to the market exhibit negative relation with choice of channel but fail to emerge as significantly influencing factor.

The chosen independent variables were regressed against the choice of the channel as dependent variable. Table 4 indicate the marginal efficiency of the factor influencing choice of channel. It could be inferred based on coefficient for age of respondents

TABLE 4  
Marginal efficiency of factors affecting choice of the channel

Variable	Marginal efficiency		
	Tomato	Potato	Cabbage
AGE	-2.199	-2.821	-5.819
EDU	1.386	1.706	1.015
HHSIZE	-1.647	-0.857	-3.438
SOC	2.227	0.203	3.537
FARMSIZE	1.394	0.306	1.193
VEGAREA	-2.006	-1.054	-1.119
CREDFAC	-4.211	-2.745	-2.542
EXTSERV	4.527	0.187	1.417
QULITY	1.054	1.110	2.012
PRICE	1.049	1.094	2.104
DISTMARKT	-1.984	-0.940	-5.819

that younger farmer tend to choose the modern channel even though, there exist a risk of rejection of produce was high based on quality specifications set. As one per cent increase in the age of the respondents will reduce 2.199 per cent of farmers to choose modern marketing channel in case of tomato, 2.821 per cent in case of potato and 5.819 per cent in case of cabbage. These results are consistent with the findings reported by Bongiwe and Micah (2013) while studying Factors affecting the choice of marketing channel by vegetable farmers in Swaziland and reported negative relation with age and choice of NAM Board market channel. As expected literacy level of respondents has positive and significant influence on choice of market channel in case of tomato and cabbage. These results are in line with Gilbert and Adam (2017). House hold size of cabbage farmers has negative and significant influence of choice of MMC. As one per cent increase in the household size reduces 3.438 per cent of farmers to choose modern channel. Social participation has positive and significant impact on choice of channel in case of tomato and cabbage farmers. As one per cent increase in the social participation increases 2.227 and 3.537 per cent of farmers to switch to modern marketing channel due to expanded social network and timely information enable farmers to take better marketing decisions. Farm size has positive and significant impact on choice of channel for all selected vegetables.

Farm size is taken as proxy for wealth of the individual. Wealthier farmers due to their vast social network, market access and market facility, probability to choose modern market channel was high. One per cent increase in farm size, access to modern markets would rise marginally by 1.394, 0.306 and 1.193 per cent for tomato, potato and cabbage respectively. Whereas area under selected vegetable has significant and negative impact on dependent variable. Large area under the vegetable means large produce which cannot be absorbed by the modern market channel, so farmers choose traditional channel and also there is problem of quality maintenance when cultivated vegetables on larger area. To take the advantage of better price realisation in modern retail marketing channel, the

large farmers are much planned and allocate manageable area for vegetable cultivation and flower cultivation was even better preference enterprise in the study area. The flower cultivation provides them regular and continuous returns to farmers and help meet day to day operational expenses in farming. As one per cent increase in vegetable area decreases the farmer's probability to choose modern channel by 2.006 per cent in case of tomato growers and about one per cent in the case of both potato, cabbage cultivation. Thus findings of the present study are in line with that of Rajanna *et al.* (2017) who reported that, small farmers with lesser vegetable area reaped more benefit from modern marketing channel.

Credit facility extended by the commission agents in APMC restrict farmers to go for modern channel. As one per cent increase in the credit facility, increase the farmers to choose the traditional channel by 2.745 per cent and 2.542 per cent in the case of potato and cabbage respectively. Price is positively and significantly influencing factor in choice of a particular retail format of marketing channel. As price increases, the farmer's choice to go for modern market channel also increases. One per cent increase in price would results in increase in the preference for modern retail format by 1.049 per cent, 1.094 per cent and 2.104 per cent in the case of tomato, potato and cabbage farmers, respectively.

Marketing decision is more important in farming since, whatever farmers gained through modern production technologies are being lost in the process of marketing. In the democratic counties like India, even with multiple number of policies, programs and interventions to improve the conditions for agriculture marketing, the glitches and hitches found continuing. However, the reasons are obvious, as are inherent in the characters of farming, farm produce and Indian farmers. However, leaving greatly to the free play of market forces, do some time help in finding better and acceptable prices for both the producer-seller and consumer-buyer. In this direction, with the emergence of modern format of retail marketing, largely located in consumption centres are playing some encouraging role of providing better prices to

producer and fresh and relatively cheaper and variety of consumer requirements at one place. Hence in the present study which was attempt to quantify the extent of association between producer's characters and choice of marketing channel brought out very interesting facts using probit regression analysis. It was in line with the approri that age and vegetable area were negatively influencing the choice of FMMC, while literacy and farm size were positively and significantly influencing preference for FMMC channel. Large farmers with more average land holding allocate less area to the vegetable cultivation took advantage of better price from FMMC and revealed win-win situation with planned and regulated production of vegetables compared to FTMC participants. Small farmers generally having financial commitment with commission agents and problem in maintaining quality of the produce, who faced frequent rejection from modern retail outlets preferred to go with APMC and other traditional mode of marketing than with collection centres of FMMC. Therefore, popularising and with expanded absorption capacity of higher quantity coupled with more number of collection centres from FMMC retail formats would help more number of farmers to get better income and improve their living standard.

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## Study on Comparative Diversity of Bacterial Endosymbionts in Invasive Rugose Spiralling Whitefly, *Aleurodicus rugioperculatus* Martin on Different Hosts

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### ABSTRACT

The contest of the study is to know the influence of host crops on the diversity of bacterial endosymbionts of rugose spiralling whitefly (RSW) *Aleurodicus rugioperculatus* Martin. RSW samples were collected from four different hosts (coconut, banana, maize and pongamia) and locations (Bangalore, Doddaballapura, Kolar, Shimoga, Davanagere, Gadag, Tumkur and Mandya). The bacterial symbionts were isolated on nutrient agar media and bacteria were identified by molecular method. RSW found on coconut were recorded highest bacterial species (19) followed by banana (14). The Shannon diversity index and Margleaf richness index bacterial endosymbionts were maximum in coconut (1.73: 0.75) followed by banana (1.47: 0.67). Evenness index of bacterial endosymbionts was maximum in pongamia (0.70) followed by coconut (0.58). Among the bacterial phyla of RSW, Firmicutes were found dominant (69.49%) followed by Proteobacteria (23.72%). Class Bacilli (68.8%) and genus Bacillus (44.26%) were found major in RSW among all the four hosts, followed by g-proteobacteria (16%) and staphylococcus (13.11%). The bacterial endosymbiont diversity of RSW showed a significant difference ( $Z=2.57$ ) between the four hosts and showed that the host influences the bacterial diversity of RSW.

Keywords : Rugose spiralling whitefly, Endosymbionts, Diversity indices

THE recent invasive insect pest, rugose spiraling whitefly (RSW), *Aleurodicus rugioperculatus* Martin (Hemiptera: Sternorrhyncha: Aleyrodidae) was described by Martin from Belize in Central America in 2004 based on puparium collected under the leaves of coconut. In India, the RSW was first observed in Pollachi area in Coimbatore district of Tamil Nadu on coconut and first reported in Kottayam from Kerala during July - August 2016 (Sundararaj and Selvaraj, 2017). Presently, the infestation spread over to Karnataka, Kerala and Andhra Pradesh and North India. RSW is morphologically distinguishable from the other whitefly species by its large size (2-3 mm in length) with irregular brown bands on wings and the male possesses a pincer-like structure at the tip of the abdomen. The most preferred hosts are coconut, banana, custard apple, guava and *Canna indica* L.

Besides this, interestingly some plant species are preferred for only some life stages of RSW i.e., arecanut, neem, *Parthenium*, mango, tapioca, pepper, ornamental creeper and sapota recorded only the egg stages of the pest (Alagar *et al.*, 2020).

The quick adaptation trait of the RSW to changing climate and to the new host is the key factor in their spatiotemporal distribution. The reason behind this adaptation is still unknown. Several research workers worked on the effect of different hosts on the lifecycle of RSW (Pradhan *et al.*, 2021) but the works on influence of hosts on its microbial diversity is scarce. The bacterial endosymbionts play an important role in host nutrition, development, fitness, survival, modulation of immune responses and communication

(Devaiah *et al.*, 2022) Therefore the present study focusing on microbial diversity of RSW was carried out on four different hosts namely, coconut, banana, maize and pongamia with the hypothesis that host influences the microbial diversity of the RSW.

## MATERIAL AND METHODS

### Sample Collection

Live RSW adults were collected along with the leaves in a polythene bag with micro holes for aeration, from four different hosts (coconut, banana, maize and pongamia) during 2021-2022 in different locations of Karnataka (Bangalore, Doddaballapura, Kolar, Shimoga, Davanagere, Gadag, Tumkur and Mandya). RSW adults were maintained in the laboratory on the same host for further experimental studies.

### Isolation of Gut Bacteria

The collected adults of RSW on different crops and locations were surface sterilized with 70 per cent ethanol for one minute followed by 0.1 per cent sodium hypochlorite for one minute, then rinsed with sterile distilled water 2 to 3 times to remove the external microbes.

The surface sterilized adults were crushed in a microcentrifuge tube using a micro pestle with 1 ml Phosphate Buffer Saline (PBS) solution (pH 7.4). The homogenized samples were centrifuged at 2000 rpm for 10 minutes. Serial dilution of samples was made up of 10<sup>-7</sup> dilutions. The aliquot of 1 µl of all the dilutions was plated on Nutrient Agar (NA) for isolating the symbiotic bacteria. An aliquot was spread using a sterilized spreader. The plating was done by the spread plate technique. The plates were incubated at 28 °C for 48 hours. After 24 hours, plates were observed for microbial growth.

### DNA Isolation from Gut Bacterial Isolates

Representative colony from colonies showing similar morphology were selected and pure cultured by sub culturing the same media. The pure culture was added to nutrient broth for the multiplication of bacterial cells. Total DNA was extracted from bacterial colonies by inoculating the single colony of bacterial culture

in nutrient broth and incubated at 37 °C for 24 hrs. Later, transferred 1.5 ml culture to micro centrifuge tube and centrifuged at 10000 rpm for 3 minutes and collected pellets. Resuspended the pellets on 400 µL sucrose buffer and vortex. Added 32 µL lysozyme, incubated for 10 minutes at 60 °C. Then added 45 µL 10 per cent SDS and 5 µL proteinase, mix well and incubate again in water bath for 10mins at 60 °C. Added 240 µL NaCl and 140 µL freshly prepared 10 per cent CTAB and kept in water bath for 10 minutes. Added 500 µL Chloroform : Isoamyl alcohol (24:1), mixed well and centrifuged at 12000 rpm for 10 minutes. Transferred the upper aqueous phase into new tube and added 50 µL 3M sodium acetate and 300 µL of isopropanol, mixed gently and incubated overnight at -20 °C. Spun at 12000 rpm for 15 min to pellet down the DNA. Added one ml 70 per cent ethanol and spun at 12000 rpm for 10 min (twice). Discarded the supernatant and allowed drying. Resuspended the DNA in 40 µL TE Buffer, added 2 µL RNase and incubated at 37 °C for 30 min (Swathi *et al.*, 2015 and Devaiah *et al.*, 2022).

### Amplification of 16s rRNA Gene

The 16SrRNA gene was amplified from bacterial colonies by PCR, using universal eubacterial primer pairs eu27. F (5' AGAGTTTGATCCTGGCTCAG-3') and eu1495.R (5'- ACGGCTACCTTGTTA CGACTT3'). PCRs were carried out in 30 µL reactions with each reaction tube containing 0.5 ml of each primer, ~ 15ng of template DNA, 3 µL Taq buffer, 1.5 µL Taq Polymerase. The following condition was used for the PCR reactions: 98 °C for 1 min, 59 °C for 30 seconds and 72 °C for 1 min for 30 cycles and a final extension of 72 °C for 10 min. PCR products were subsequently subjected to Agarose gel electrophoresis. Aliquots (2L) of each PCR product were resolved electrophoretically on one per cent agarose gel using 10X TAE buffer. The PCR products visualized with an UV transilluminator and photographed with a gel documentation system (Gel Doc 200, BIO-RAD, USA) after staining the gel with ethidium bromide (0.5mg mL<sup>-1</sup>) (Promega), the DNA molecular weight marker, a 1-kbp DNA ladder (Promega) was used to determine the size of the amplified fragments.

### 16S rRNA Sequencing Analysis

The purified PCR products were sent for sequencing. The nucleotide sequencing of the PCR fragments was performed. The DNA sequences corresponding to 16SrRNA gene, obtained from individual bacteria was reverse complemented using software Bioedit. The obtained sequences were analysed along with the sequences retrieved from the NCBI (National Centre for Biotechnology Information) GenBank using bioinformatics software and bacterial isolates were identified.

Agarose gel electrophoresis was performed and the aliquots of each PCR product was resolved electrophoretically on 1-1.5 per cent agarose gel using 0.5' TAE buffer. The PCR products were visualized with a UV transilluminator and photographed with a gel documentation system after staining the gel with ethidium bromide ( $0.5 \mu\text{g mL}^{-1}$ ), the DNA molecular weight marker, a 1-kbp DNA ladder, was used to determine the size of the amplified fragments. The amplicons eluted and sent for sequencing. The obtained sequences were analyzed along with the 16sRNA gene sequences retrieved from the NCBI GenBank and sequences were obtained and accession ID were obtained.

### Diversity of Bacterial Endosymbionts

The microbial diversity of RSW on different hosts and between hosts was calculated by using Shanon diversity index (H) and species richness was calculated by using Margalef index of richness (K) species evenness was calculated by using Shanon evenness ( $E_H$ ) index by using following formulae.

#### Shanon Diversity Index ( $H$ ) = $-\sum \pi_i \times \ln(\pi_i)$

$\Sigma$  : A Greek symbol that means ; 'sum'

$\ln$  : Natural log

$\pi_i$  : The proportion of the entire community made up of species i

The higher the value of  $H$ , the higher the diversity of species in a particular community. The lower the value of  $H$ , the lower the diversity. A value of  $H=0$  indicates a community that only has one species

#### Evenness Index ( $E_H$ ) = $H / \ln(S)$

H : The Shannon Diversity Index

S : The total number of unique species

#### Margalef Index of Richness (K)

$K = \log S / \log N$

S indicates the number of species and N indicates the total number of individuals in the sample

The significance test for diversity of endosymbionts of RSW in different hosts was computed by subjecting the data to Wilcoxon Signed Ranks Test for by using the software SPSS 16.0.

### RESULTS AND DISCUSSION

Samples of RSW collected on different crops from different locations revealed the diversity of gut bacteria varied with hosts and locations (Table 1). Among the hosts, coconut showed more number of bacterial species (19) followed by banana (14) and the least was found in pongamia with three bacterial symbionts. A significant difference ( $Z=2.5$ ) in microbial diversity of RSW on all four hosts (Table 2). The results were found similar to the work of Saranya *et al.* (2022), recorded 17 gut bacterial isolates of RSW from coconut and 32 isolates from banana. Similarly, 11 bacterial genera were isolated from sweet potato whitefly, *Bemisia tabaci*, which included *Pseudomonas*, *Deinococcus*, *Sphingomonas*, *Acinetobacter*, *Staphylococcus*, *Modestobacter*, *Micrococcus*, *Bacillus*, *Kocuria*, *Microbacterium*, *Erwinia*, *Brevibacterium*, *Exiguobacterium* and *Moraxella* (Ateyyat *et al.*, 2010; Indiragandhi *et al.*, 2010 and Visotto *et al.*, 2009).

The majority of the bacterial species belonging to the phylum Firmicutes in all the hosts except the pongamia where Actinobacteria was the dominant phylum (Table 3). Among the three phylums, Firmicutes (69.49%) was found to be dominant followed by Proteobacteria (23.72%) (Fig. 1). Of the different classes, Bacilli (68.8%) was the major class in all the hosts followed by g-proteobacteria (16%). Among the different genera, *Bacillus* constitutes 44.26 per cent of the bacterial species followed by *Staphylococcus* (13.11%). Similarly, Saranya

TABLE 1  
Diversity of bacterial endosymbionts of RSW in four different hosts

Coconut	Banana	Maize	Pongamia
<i>Bacillus</i> sp.	<i>Bacillus cereus</i>	<i>Bacillus</i> sp.	<i>Curtobacterium citreum</i>
<i>Bacillus cereus</i>	<i>Staphylococcus saprophyticus</i>	<i>Terribacillus</i> sp.	<i>Metabacillus indicus</i>
<i>Kocuria palustris</i>	<i>Bacillus pumilus</i>	<i>Priestia aryabhatai</i>	<i>Brachybacterium</i> sp.
<i>Micrococcus aloeverae</i>	<i>Bacillus</i> sp.	<i>Bacillus</i> sp.	
<i>Brevundimonas</i> sp.	<i>Staphylococcus</i> sp.	<i>Bacillus aeriis</i>	
<i>Staphylococcus haemolyticus</i>	<i>Klebsiella variicola</i>	<i>Bacillus xiamenensis</i>	
<i>Serratia nematodiphila</i>	<i>Staphylococcus</i> sp.	<i>Priestia megaterium</i>	
<i>Bacillus subtilis</i>	<i>Lactococcus lactis</i>	<i>Bacillus pumilus</i>	
<i>Cytobacillus kochii</i>	<i>Bacillus</i> sp.		
<i>Acinetobacter pittii</i>	<i>Acinetobacter soli</i>		
<i>Pseudomonas monteilii</i>	<i>Staphylococcus</i> sp.		
<i>Pseudomonas plecoglossicida</i>	<i>Enterobacter hormaechei</i>		
<i>Bacillus pumilus</i>	<i>Bacillus</i> sp.		
<i>Bacillus albus</i>	<i>Bacillus subtilis</i>		
<i>Bacillus subtilis</i>			
<i>Bacillus licheniformis</i>			
<i>Bacillus</i> sp.			
<i>Pseudomonas</i> sp.			
<i>Bacillus haynesii</i>			

TABLE 2

Number of isolates from the different hosts

Host	No. of microbes	Z Value
Coconut	19	2.570 **
Banana	14	
Maize	8	
Pongamia	3	

\*\* Significant at 1% level

*et al.* (2022) observed *Bacillus* (80-100%) was the most abundant bacterial genus in the RSW reared on all tested host plants. The genera *Bacillus* (30%), *Acinetobacter* (10%) and *Exiguobacterium* (10%) were observed in the RSW reared on coconut plants.

### Diversity Indices

The Shannon diversity index computed showed that, highest bacterial diversity was observed in coconut

(1.73) followed by banana (1.47). Interestingly, the Evenness index for pongamia found high (since, number of species is are less) followed by coconut (0.58). Margalef index of richness (K) for coconut was maximum (0.75) followed by banana (0.67) and least was found in pongamia (0.28) (Table 4). Saranya *et al.* (2022) found maximum bacterial diversity and species richness for the isolates of RSW from coconut (2.20 : 3.64), followed by those in the isolates of RSW from banana (2.20: 3.64) as indicated by the Shannon and Margalef diversity indices.

The present study revealed that, host plants influence the gut microbial diversity in host insects. This observation was supported by the findings of Jones *et al.* (2019), who showed that maize and soybean altered the microbial communities in the fall armyworm, *Spodoptera frugiperda*. The gut bacterial populations of *Henosepilachna vigintioctopunctata*

TABLE 3  
Bacterial endosymbionts diversity of RSW in four different hosts

Phylum	Class	Species
Firmicutes	Bacilli (42)	<i>Bacillus</i> sp.(27) <i>Staphylococcus</i> sp. (8) <i>Terribacillus</i> sp.(1) <i>Priestia</i> (2) <i>Niallia nealsonii</i> <i>Brevibacillus brevis</i> <i>Cytobacillus kochii</i> <i>Lactococcus lactis</i>
Proteobacteria	a- proteobacteria (3)	<i>Sphingobium yanoikuyae</i> <i>Brevundimonas</i> (2)
	b-proteobacteria (2)	<i>Chromobacterium haemolyticum</i> <i>Acidovorax</i> sp.
	g-proteobacteria (10)	<i>Acinetobacter</i> (4) <i>Serratia nematodiphila</i> <i>Enterobacter hormaechei</i> <i>Klebsiella variicola</i> <i>Stenotrophomonas maltophilia</i>
Actinobacteria	Actinomycetia (4)	<i>Kocuria palustris</i> <i>Micrococcus aloeverae</i> <i>Curtobacterium</i> (2)

Note: Number present in parenthesis indicate total number of species

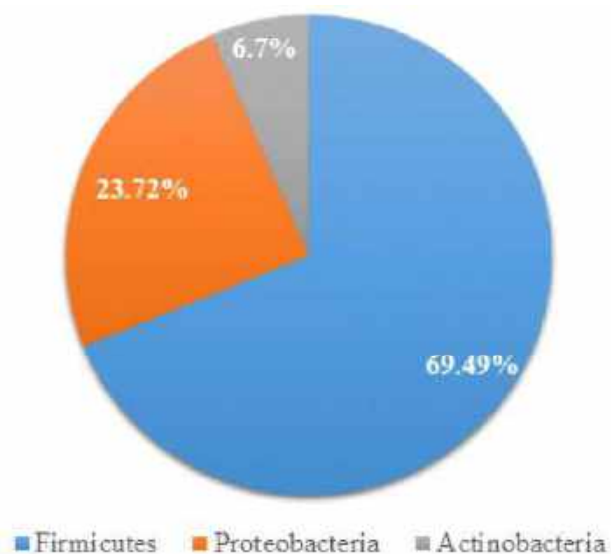


Fig. 1 : Diversity of bacterial endosymbionts at phylum level in RSW on four different hosts

TABLE 4  
Diversity indices of bacterial endosymbionts isolated from RSW

Host	H	E <sub>H</sub>	K
Coconut	1.73	0.58	0.75
Banana	1.47	0.55	0.67
Maize	0.90	0.43	0.53
Pongamia	0.77	0.70	0.28

H= Shannon diversity index, E<sub>H</sub> = Evenness index, K= Margalef index of richness

were influenced by the host plants, *Solanum melongena* (QZ) and *Solanum nigrum* (LK). LK is associated with phylum Cyanobacteria, class Alphaproteobacteria and genus *Ochrobactrum*, while QZ supports *Bacillus* and *Lactococcus*.



Host plants have a positive impact on the shaping of microbial communities associated with *Spodoptera littoralis* (Tang *et al.* 2012), *Helicoverpa* spp. (Priya *et al.*, 2012; Tang *et al.*, 2012; Xiang *et al.*, 2006), *Lymantria dispar* (Broderick *et al.*, 2004; Mason and Raffa 2014) and *Leptinotarsa decemlineata* (Chung *et al.*, 2017). Plant characters such as leaf surface, wax composition and the availability of sugars in plants might influence bacterial community composition in the host insect (Lindow and Brandl 2003).

This study enables with deep understanding of the bacterial endosymbionts associated with the RSW on different host crops. This experiment revealed that, RSW collected on the coconut and banana has harboured more diversity of endosymbionts. The change in bacterial diversity in different host crops have made a channel to study about how the endosymbionts are obtained at different crops and eliminated from their body. Understanding the specific functions of each endosymbionts and the transmission patterns will be a suitable area for future research..

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## Standardization of Herbal Enriched Finger Millet Based Composite Flour Mix

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### ABSTRACT

Composite flour is a mixture of flours from cereals, pulses, oilseeds and tubers with or without addition of wheat flour. Medicinal herbs own many health benefits because of its, therapeutic or curative aids. Incorporation of herbs to food products helps to manage many diseases. Finger millet based composite flour (FBCF) had been developed with the incorporation of black rice, black soya bean, barnyard millet and pumpkin seeds. The present research investigation proceeded by standardizing the composite flour mix by using finger millet and other ingredients in the ratio of 70:30, 60:40, 50:50 and 40:60. The developed mixes were subjected to functional properties and sensory evaluation to select the best combination. Best accepted combination of FBCF analyzed for nutrient composition. Addition of 50 per cent finger millet and 50 per cent in combination of other ingredients had better functional properties and sensory scores compared to other treatments. Finger millet based composite mix had significantly higher protein (15.93 g/100 g) and carbohydrate (63.26 g/ 100 g) compared to control. Considering the literature studies herbs which are rich in antioxidant activity *viz.*, *Amrutha balli*, Indian borage, *honagone* leaves, dried ginger, turmeric and clove had been selected to develop herbal mix. Herbal mix was standardized by considering the sensory evaluation scores of *kashaya*. Herbal mix treatment 3 (HMT3) had good sensory scores and antioxidant activity (83.33%) compared to other treatments. Further best accepted composite flour was used for the preparation of herbal enriched composite flour by incorporating the developed herbal mix at different composition. It can be concluded that finger millet based herbal enriched dosa mix treatment 3 (HFCFT3) had better sensory scores and also the developed mix had good nutritional composition and antioxidant activity.

**Keywords :** Composite flour, Herbal mix, Finger millet, Sensory scores

COMPOSITE flour (CF) is a mixture of different flours from cereal, legume or root crops with or without addition of wheat flour which helps to satisfy specific functional characteristics and nutrient composition [Bolarinwa *et al.*, 2015]. CF provides essential amino acid balance, dietary fibre, antioxidants and high mineral content as compared to wheat flour, which may help to overcome the problem of protein energy malnutrition and other diseases [Tangariya *et al.*, 2018]. The use of composite flour based on wheat and other cereals including minor millets in traditional and bakery products is becoming popular because of

the economic and nutritional advantages of composite flour [Bolarinwa *et al.*, 2015]. Composite flour technology has been widely adopted round the globe for development of functional foods with the desired therapeutic value [Raihan and Saini, 2017]. Composite flour had a few advantages for developing countries because it reduces the import of finger millet flour and encourages the use of domestic agricultural products as flour.

Finger millet is the most nutritious among all major cereals and it has been perceived as 'super cereal' by

the United States National Academies [Ranganatha *et al.*, 2022]. Herbs are plants with savory or aromatic properties that are used for flavoring, garnishing food, medicinal purposes and for fragrances. The use of medicinal plants has attained an important role in health system all over the world. This involves use of medicinal plants not only for the treatment of disease but also as a potential material for maintaining good health [Yadav *et al.*, 2020]. Herbs are beneficial for human health because as it contains significant amount of micronutrients, vitamins, antioxidants, phytochemicals and fiber content that may help protect against degenerative diseases and micronutrient malnutrition [Gupta *et al.*, 2012]. A combination of composite flour along with herbal mix is rare and so this will help to achieve the expected outcome by balancing the unavailable nutrients.

Commercially many composite flour mixes are available in the market but ready to prepare herbal enriched mixes are less available. As the herbs own many health benefits, it's incorporation in food product helps to manage many diseases. Hence, considering the research need present quest has been put forth with the objectives:

1. To standardize the herbal enriched finger millet based composite flour mix
2. To analyze the functional properties and nutrient composition of developed mix

## MATERIAL AND METHODS

*Development of Finger Millet Based Composite Flour Mix* : For the preparation of finger millet based composite flour mix, finger millet was substituted with other ingredients such as black rice, black soya bean, barnyard millet and pumpkin seeds. Each ingredient was cleaned, dried and powdered separately to prepare composite flour mix. The powdered ingredients were weighed at different levels and used for the preparation of finger millet based composite flour mix. Composite flour was developed by incorporating other ingredients at different levels (Table 1) by substituting finger millet flour at different

TABLE 1  
Formulation of composite flour mix

Ingredients	CFT1 (70:30)	CFT2 (60:40)	CFT3 (50:50)	CFT4 (40:60)
Finger millet	70	60	50	40
Black rice	15	15	15	15
Barnyard millet	5	10	15	20
Black soybean	5	10	15	20
Pumpkin seeds	5	5	5	5

CFT1 - Finger millet based composite flour treatment 1 (70:30); CFT2 - Finger millet based composite flour treatment 2 (60:40); CFT3 - Finger millet based composite flour treatment 3 (50:50); CFT4 - Finger millet based composite flour treatment 4 (40:60)

ratios (70:30, 60:40, 50:50 and 40:60). Developed Composite flour mixes were analyzed for the functional properties and gruel was prepared and subjected for sensory evaluation. Nutritive value of



Fig. 1: Finger millet based composite flour gruel

best accepted flour mix was calculated and used for the enrichment with herbal mix.

## Functional Properties of Composite Flour

Functional properties such as water absorption capacity, oil absorption capacity, swelling power, swelling capacity was analyzed to finger millet based and wheat based composite flour to select the best combination.

### Water Absorption Capacity (WAC)

A suspension of 1.0 g of sample in 10 ml distilled water was agitated 4 times allowing 10 min. resting periods between each mixing and centrifuged at 3250 rpm for 25 min. The supernatant was decanted and tubes were air-dried and then weighed (Sindhu and Khatkar, 2016).

WAC (ml/g) = Volume of water/weight of sample absorbed

### Oil Absorption Capacity (OAC)

The 3 ml refined groundnut oil was added to 0.5 g of sample and stirred for 1 minute. After 30 min. at room temperature, the tubes were centrifuged at 3200 rpm for 25 minutes. The volume of unabsorbed oil was determined (Sindhu and Khatkar, 2016).

OAC (ml/g) = Volume of fat/weight of sample

### Swelling Capacity

The swelling capacity was determined by the method described by Potter and Hotchkiss (2012) 100 ml graduated cylinder was filled with the sample to 10 ml mark. The distilled water was added to give a total volume of 50 ml. The top of the graduated cylinder was tightly covered and mixed by inverting the cylinder. The suspension was inverted again after 2 min and left to stand for a further 8 min and the volume occupied by the sample was taken after the 8<sup>th</sup> min.

### Foam Capacity

The foam capacity (FC) were determined as described by Hasnadi *et al.*, 2020 with slight modification. One gram of flour sample was added to 50 mL distilled water at  $30 \pm 2$  °C in a graduated cylinder. The suspension was mixed and shaken for 5 min to foam. The volume of foam at 30 sec after whipping was expressed as foam capacity using the formula,

Where, AW = after whipping, BW = before whipping. The volume of foam was recorded one hour after whipping to determine foam stability as per percent of initial foam volume.

### Flour Dispersibility

Dispersibility is an index that measures how well flour blends can be rehydrated with water without formation of lumps. The flour dispersions of 2, 4, 6, 8, 10, 12, 14, 16, 18, 20, 22 and 30 per cent (w/v) prepared in 5 ml distilled water was heated at 90 °C for 1 hr in water bath. The contents were cooled under

tap water and kept for 2 hr at  $10 \pm 2$  °C (Baranwal and Sankhla, 2019).

### Bulk Density

The volume of 100 g of the flour was measured in a measuring cylinder (250 ml) after tapping the cylinder on a wooden plank until no visible decrease in volume was noticed and based on the weight and volume, the apparent (bulk) density was calculated (Jones *et al.*, 2000).

$$\text{Bulk density (g/ml)} = \frac{\text{Seed weight (g)}}{\text{Seed volume (ml)}}$$

*Proximate Principles* : Proximate principles of herbal enriched wheat based composite flour mix was analyzed. Proximate principles (PC) *viz.*, moisture, fat, crude protein, crude fiber and ash by standard methods (AOAC, 2005). Difference method was used to calculate carbohydrate and energy value by computation method.

*Development of Herbal mix* : Different herbs such as *Amrutha balli*, Clove, *Honagone* leaves, Indian borage, turmeric, dried ginger were procured. Herbs were cleaned and dried to remove moisture content. Further the herbs made into fine powder by using mixer and sieved with mesh size (212 µm). Herbal mix was standardized by adding selected herbs with different composition (Table 2) and subjected for sensory evaluation by a group of 21 trained panel members in the form of *kashaya*. The product was evaluated for sensory evaluation by using nine point hedonic scale (Amerine *et al.*, 1965). Further best

TABLE 2

Formulation of herbal mix

Herbs	HMT1	HMT2	HMT3	HMT4
<i>Amruthaballi</i>	1.75	1.5	1.0	0.5
Clove	0.25	0.5	1.0	1.5
<i>Honagone</i> leaves	2	2	2	2
Dried ginger	2	2	2	2
Indian borage	2	2	2	2
Turmeric	2	2	2	2

HMT1- Herbal mix treatment 1  
 HMT2- Herbal mix treatment 2  
 HMT3- Herbal mix treatment 3  
 HMT4- Herbal mix treatment 4



Fig. 2: Herbal mix kashaya drink

accepted combination was analyzed for antioxidant activity by three different methods such as DPPH (2, 2-diphenyl-1-picryl-hydrazyl-hydrate), FRAP (Ferric ion reducing antioxidant power) and ABTS (2, 2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)).

**Extraction of Sample :** The sample (2g) was weighed accurately and extracted at room temperature with 85 per cent aqueous methanol under agitation using a magnetic stirrer for 30 min. The extracts were soaked overnight and mixtures were centrifuged at 2500 rpm for 10 min and the supernatants were collected and dried in hot air oven at 27-35 °C, known amount of solvent added to dried supernatant. Extract kept in refrigerator for further antioxidant analysis.

#### Scavenging Ability toward DPPH Radical

DPPH method was used in the determination of the antioxidant activity, which is based on the quantification of free radical scavenging with modifications. This method depends on the reduction of DPPH• radical (purple) to a yellow colored diphenyl picrylhydrazine. A decrease in the DPPH absorbance indicates an increase of the DPPH• radical scavenging activity. A methanolic solution containing 0.06 mM of the DPPH• radical was prepared daily and protected from light. 0.1 mL of extract was added to 3.9 mL of DPPH• methanolic solution. The decrease in absorbance at 515 nm using a UV-Vis spectrophotometer was measured at 1 min intervals for the first 10 min and then at 5 min intervals until stabilization. All measurements were performed in triplicate (Lemine *et al.*, 2014)..

#### ABTS + Assay

The ABTS+ assay was performed according to method established previously with modifications. The

pre-formed radical monocation (ABTS+•) was produced by oxidation of 7 mM ABTS stock solution with 145 mM potassium persulfate and then incubated in the dark for 16 h at room temperature before use. The ABTS + working solution was prepared by diluting the stock solution with ethanol until reach an absorbance of  $0.70 \pm 0.02$  (at 734 nm). All samples were diluted approximately to provide 20-80 per cent inhibition of the blank absorbance. 30  $\mu$ L of the extract was mixed with 3.0 mL ABTS + working solution. The absorbance of the mixture was measured at 734 nm after 6 min of incubation at room temperature. The ABTS scavenging capacity was expressed as  $\mu$ M Trolox/g mix (Xiao *et al.*, 2020).

#### Ferric Reducing Antioxidant Power (FRAP)

Ferric reducing antioxidant power assay was measured according to the procedure with some modifications. The FRAP reagent contained 2.5 mL of a 10 mM TPTZ (2,4,6-Tripyridyl-s-Triazine) solution in 40 mM HCl, 2.5 mL of 10 mM  $\text{FeCl}_3 \cdot 6\text{H}_2\text{O}$  and 25 mL of 300 mM acetate buffer (pH 3.6). It was freshly prepared and warmed at 37 °C. A 900  $\mu$ L FRAP reagent was mixed with 90  $\mu$ L water and 30  $\mu$ L of the extract. The reaction mixture was incubated at 37 °C for 10 min and the absorbance was measured at 593 nm. FRAP was expressed as  $\mu$ M de  $\text{F}_e\text{SO}_4/\text{g}$  of dry sample (Xiao *et al.*, 2020).

**Development of Herbal Enriched Finger Millet Based Composite Flour Mix :** Best combination of composite flour mix and herbal mix was used for the development of herbal enriched finger millet based composite flour mix. Standardized composite flour mix was substituted with herbs in the ratio of 85:15,

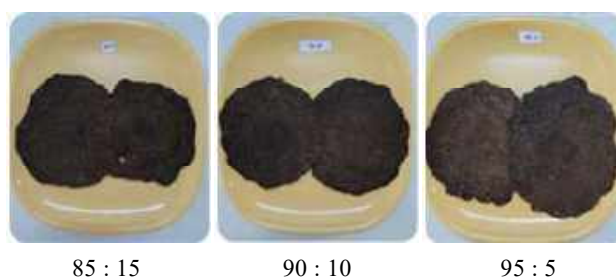


Fig. 3: Herbal enriched finger millet based dosa

90:10, 95:5 had been added to formulate herbal enriched composite mix. The developed mixes were used for the preparation of dosa and subjected for sensory evaluation to know the acceptability.

### Statistical Analysis

The SPSS version 16 software programme was used to estimate the mean, standard deviation, standard error mean, 'S.E. diff', 'CD' and 'F' value. One - way ANOVA was employed to know the difference between the products (Fisher and Yuest, 1963). The data thus obtained from nutrient contents were statistically analyzed by applying 't' test. The critical difference between the products was tested at 5 per cent significance.

### RESULTS AND DISCUSSION

Table 3 depicts the functional properties of finger millet based composite flour mix. The functional properties of food proteins are important in food processing and food product formulation (Alvarez-Jubetea *et al.*, 2010). As the addition of other ingredients in composite flour increases up to 50 per cent water absorption capacity, oil absorption capacity and bulk density increased in the treatments whereas, swelling power, per cent

solubility, flour dispersibility and foaming capacity decreased. This might be due to the addition of soy bean in the composite flour mix, soy protein had the highest water-binding capacity and also it contain polysaccharides, which absorb a significant amount of water. Chandra *et al.* (2015) found higher results compared to present study, this is due to the addition of soy bean at higher level as compared to the present study.

Table 4 represents the nutrient composition of finger millet based composite flour mix. Finger millet based composite mix had significantly higher protein and carbohydrate compared to control sample. Higher protein content due to the higher amount of protein in the soy bean and pumpkin seeds. Bolarinwa *et al.* (2015) observed lesser results compared to present study this might be due to lesser quantity of soy bean used and addition of other ingredients which are not rich in macronutrients.

Fig. 4 and 5 depicts the sensory evaluation of finger millet based composite flour gruel and herbal kashaya respectively. Sensory scores of up to 50 per cent addition composite flour to finger millet flour (CFT3) was accepted by the panel members after that the scores were slightly decreased. Significant difference was found in flavor and taste among the different

TABLE 3  
Functional properties of finger millet based composite flour mix

Treatments	Water absorption capacity (%)	Oil absorption capacity (%)	Swelling power (%)	Per cent solubility (%)	Flour dispersibility (%)	Foaming capacity (%)	Bulk density (g/ml)
CFT1	100±0.01 <sup>b</sup>	103±0.05 <sup>a</sup>	3.53±0.03 <sup>d</sup>	0.76±0.05 <sup>a</sup>	71.33±0.57 <sup>a</sup>	6.64±0.05 <sup>a</sup>	0.65±0.01 <sup>a</sup>
CFT2	110±0.01 <sup>b</sup>	103±0.11 <sup>a</sup>	3.59±0.01 <sup>c</sup>	0.56±0.05 <sup>a</sup>	70.00±1.00 <sup>a</sup>	6.62±0.15 <sup>a</sup>	0.61±0.01 <sup>b</sup>
CFT3	153±0.05 <sup>a</sup>	106±0.05 <sup>a</sup>	3.98±0.01 <sup>a</sup>	0.46±0.05 <sup>b</sup>	67.33±0.28 <sup>b</sup>	5.43±0.05 <sup>b</sup>	0.57±0.01 <sup>c</sup>
CFT4	156±0.29 <sup>a</sup>	101±0.01 <sup>b</sup>	3.71±0.01 <sup>b</sup>	0.40±0.01 <sup>b</sup>	62.41±0.14 <sup>b</sup>	5.13±0.05 <sup>b</sup>	0.56±0.01 <sup>c</sup>
F value	72.73 **	9.11 *	346.48 **	40.33 **	191.93 **	927.98 **	98.83 **
S. Em±	0.031	0.035	0.016	0.027	0.299	0.022	0.015
CD@ 5 %	0.103	0.115	0.051	0.089	0.976	0.072	0.051

CFT1- Finger millet based composite flour treatment 1 (70:30); CFT2- Finger millet based composite flour treatment 2 (60:40); CFT3- Finger millet based composite flour treatment 3 (50:50); CFT4- Finger millet based composite flour treatment 4 (40:60)

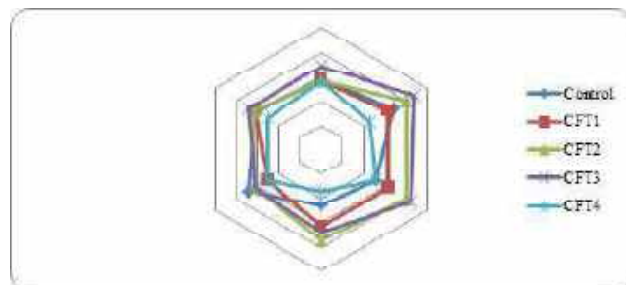
Note: S.Em: Standard Error of mean, C.D: Critical Difference, \*\* - Significant at 0.01 per cent level,

\*- Significant at 0.05 per cent Different super scripts within a column indicate significant difference at 0.05 level by DMRT

TABLE 4  
Nutrient composition of finger millet based composite flour mix

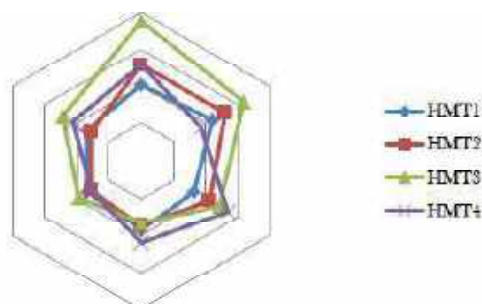
Treatments	Moisture (g/100 g)	Protein (g/100 g)	Fat (g/100 g)	Crude fiber (g/100 g)	Ash (g/100 g)	Carbohydrate (g/100 g)	Energy Kcal
Control	5.58 ± 0.04	8.25 ± 0.31	7.82 ± 0.16	2.40 ± 0.07	2.35 ± 0.05	73.10 ± 0.19	395
CFT3	7.50 ± 0.39	15.93 ± 0.85	8.54 ± 0.27	2.70 ± 0.10	2.53 ± 0.02	63.26 ± 0.70	287
t value	8.49 <sup>NS</sup>	3.84 <sup>*</sup>	14.66 <sup>NS</sup>	5.54 <sup>NS</sup>	4.14 <sup>NS</sup>	23.16 <sup>*</sup>	27.37 <sup>*</sup>

\*\* - Significant at 0.01 per cent level, \* - Significant at 0.05 per cent, NS-Non Significant, # computed nutritive value



CFT1- Finger millet based composite flour treatment 1 (70:30)  
CFT2- Finger millet based composite flour treatment 2 (60:40)  
CFT3- Finger millet based composite flour treatment 3 (50:50)  
CFT4- Finger millet based composite flour treatment 4 (40:60)

Fig. 4: Sensory scores of Finger millet based composite flour mix



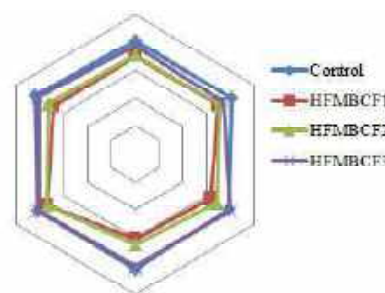
HMT1- Herbal mix treatment 1  
HMT2- Herbal mix treatment 2  
HMT3- Herbal mix treatment 3  
HMT4- Herbal mix treatment 4

Fig. 5: Sensory scores of herbal kashaya drink

treatment. This might be due to soy bean proteins might modify flavor by binding flavors and off-flavors to generate flavors on cooking and to release reactants that may produce flavors, especially in hydrolysis or proteolysis. (Pawar *et al.*, 2020) observed higher sensory scores for porridge mix compared to present investigation. This might be due to amount of soy bean

addition difference. HMT3 had better sensory scores with respect to appearance (8.38), color (8.09), flavor (7.71), taste (7.33), texture (7.47) and overall acceptability (7.71) compared to other treatment. Fig. 6 depicts the sensory scores of herbal enriched finger millet based composite flour dosa. HFMBCF3 treatment ranked first with overall acceptability score 8.25 followed by HFMBCF 2 with score of 7.28. Highly significant difference was observed in sensory parameters among the three treatments. This difference was found due to addition of herbal mix gives bitter flavour in dosa, bitterness might be due to higher amount of polyphenols and flavonoid content in herbs (Kumar *et al.*, 2021).

Table 5 showed the antioxidant activity of herbal mix by different method. As the concentration increases, antioxidant activity increased in the herbal mix. Among the three method ABTS method showed higher antioxidant activity 83.33 per cent at the



HFCE1- Herbal enriched finger millet based composite flour treatment 1 (FCF: HM 85:15)  
HFCE2- Herbal enriched finger millet based composite flour treatment 2 (FCF: HM 90:10)  
HFCE3- Herbal enriched finger millet based composite flour treatment 3 (FCF: HM 95:5)

Fig. 6 : Sensory scores of herbal enriched finger millet composite flour based dosa



TABLE 5  
Antioxidant activity of herbal mix

Concentration (µg/ml)	% Radical scavenging activity		
	DPPH	FRAP	ABTS
100	40.92	32.13	50.66
150	47.54	38.13	58.66
200	59.13	46.46	69.33
250	64.93	50.94	81.41
300	76.52	66.69	83.33

concentration of 300 (µg/ml). Natarajan *et al.* (2006) observed higher results compared to the present study. This difference was observed due to different herbs addition and the level of addition of herbs variation in the herbal mix.

Addition of finger millet with other ingredients rich in protein and micronutrients plays an important role in maintaining overall health. Results concluded that 50 per cent of finger millet and 50 per cent of other ingredients had better sensory scores compared to other treatments. Best accepted composite flour mix had good nutrient composition compared to control. Addition of five per cent of herbal mix to finger millet based composite flour dosa had better sensory scores by panel members. To increase the consumption of macronutrients there is a need for fortification to the staple food because the single cereal foodies lacking in protein and fatty acids. Hence, finger millet based composite flour enriched with other cereal, millet, oilseed and herbs fortification to the finger millet helps to improve nutritional status of the population.

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## Comparative *in silico* Analysis of Coat Protein (CP) of *Tomato Leaf Curl Virus* (ToLCV) and *Tomato Yellow Leaf Curl Virus* (TYLCV) and their Molecular Docking with GroEL Protein of *Hamiltonella* an Endosymbiont of their Vector *Bemisia Tabaci*

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preparation;  
ANITHA PETER :  
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### ABSTRACT

Begomoviruses belonging to the family *Geminiviridae* are one of the devastating group of DNA viruses causing huge losses to agricultural crop production. Begomoviruses are characterized by the presence of monopartite or bipartite genome and genetic material being the single stranded circular DNA with overlapping open reading frames (ORFs). *Tomato leaf curl virus* (ToLCV) and *tomato yellow leaf curl virus* (TYLCV) are the major begomoviruses that infect tomato crop. These viruses are transmitted by whitefly, *Bemisia tabaci* in a circulative and persistent manner. The successful infection of these viruses on healthy plant mainly rely on the interaction between the viral proteins and whitefly proteins. Coat Protein (CP) of both the viruses are crucial for the successful transmission of viruses from infected plant to a healthy plant. During the process of transmission, CP interacts with the whitefly proteins in the digestive tract, midgut, haemolymph and salivary glands. The amino acid sequences of TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL are retrieved from NCBI database and subjected to pair wise alignment. The alignment results of TYLCV-CP and ToLCV-CP revealed that the proteins have very few differences. Homology modelling was carried out using SWISS-MODEL and the obtained models are subjected to validation on PDB sum web server. Ramachandran plot and Ramachandran plot statistics confirmed that the modelled structures are reliable. The modelled structures are used for protein-protein docking studies on H-dock online tool. Docking between TYLCV-CP and *Hamiltonella* GroEL and ToLCV-CP and *Hamiltonella* GroEL showed that the interaction between CP of TYLCV and *Hamiltonella* GroEL is stronger compared to the interaction of ToLCV-CP and *Hamiltonella* GroEL. Although, the number of amino acids of TYLCV-CP involved in interaction with *Hamiltonella* GroEL are lesser, the frequency at which amino acids of TYLCV-CP involved in interaction is higher than that of ToLCV-CP, indicating that CP of TYLCV interacts strongly with *Hamiltonella* GroEL in comparison with CP of ToLCV.

**Keywords :** Tomato leaf curl virus, Tomato yellow leaf curl virus, *Hamiltonella* GroEL

**B**EGOMOVIRUS belongs to the family *Geminiviridae* and is the largest genus among viruses (Gutierrez, 1999). They majorly infect dicotyledonous crops causing huge crop losses in the tropical and sub-tropical region (Rana *et al.*, 2012). The group of virus has led to the severe outbreak of many diseases

such as cassava mosaic disease in Africa, cotton leaf curl disease in India, tomato leaf curl disease (ToLCD), tomato yellow leaf curl disease (TYLCD), yellow vein disease of okra, papaya leaf curl disease and mung bean yellow mosaic disease (Varma and Malathi, 2003). In addition to these crops,

begomoviruses also infect chilly, beans, cucurbits, cabbage and potato (Inoue-Nagata *et al.*, 2016; Kumar *et al.*, 2011 and Leke *et al.*, 2015). Begomoviruses either have a monopartite genome (DNA-A) or a bipartite genomes (DNA-A and DNA-B) with circular single stranded DNA (ssDNA) as the genetic material that encode the proteins that are required for replication, movement (intracellular and intercellular), transmission and pathogenesis (Hanley-Bowdoin *et al.*, 2013). Bipartite begomoviruses possess two genomes, *i.e.*, ~2.7 kb of DNA-A and ~2.6 kb of DNA-B. Monopartite begomoviruses are characterized by the presence of only DNA-A component of ~2.7 kb. DNA-A and DNA-B components have partially overlapping open reading frames (ORFs) which are reported to be present in a bidirectional manner (Fontenelle *et al.*, 2007 and Kheyir-Pour *et al.*, 2000).

Sweet potato white fly, *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) transmits begomoviruses from infected plant to a healthy plant when it feeds on the phloem sap of the healthy plant. The vector, *B. tabaci* transmits begomoviruses in a persistent and circulative manner (Hogenhout *et al.*, 2008). When *B. tabaci* feeds on the begomovirus infected plant, the insect ingests the begomoviral particles into the alimentary canal while ingesting the phloem sap. This is followed by subsequent movement of begomoviral particles into the filter chamber and midgut region. In the midgut region, begomovirus particles cross the gut membrane and enter into the haemolymph. Further, virion particles reach the salivary glands, pierce them and settle in salivary duct. Upon feeding on a healthy plant in the following feeding cycle, the acquired begomoviral particles are egested out into the healthy plant along with the saliva (Czosnek *et al.*, 2002 and Ghanim *et al.*, 2001).

During the movement of begomovirus in the whitefly vector, the viral particles interact with whitefly proteins facilitating the transport from the digestive tract to the haemolymph followed by movement from haemolymph to salivary glands. (Rana *et al.*, 2016). Once the viral particles are in the salivary

glands of whitefly, they are translocated into the salivary duct which are egested into the healthy plants when the whitefly feeds on them (Gray, 1996). Proteomics and transcriptomics studies have revealed the interaction of various whitefly proteins with the coat protein (CP) of the begomovirus. According to Briddon *et al.* (1990) and Noris *et al.* (1998), CP of the begomovirus is the only viral protein required for insect mediated transmission of the virus. Earlier studies by Noris *et al.* (1998) has shown that altering the amino acid sequence of the CP results in change in vector specificity and ability of the vector to transmit the virus.

Rana *et al.* (2016) have reported the role of *B. tabaci* midgut protein (MGP) in transmission of tomato leaf curl virus (ToLCV) by carrying out *in vitro* pull down assay, dot blot assay and yeast two hybrid assay using ToLCV-CP as bait. Some of the interactions between the virus and the whitefly reduces the spread of viral transmission. *B. tabaci* heat shock protein 70 (HSP70) interacts with CP of *tomato yellow leaf curl virus* (TYLCV) leading to the inhibition of virus transmission (Gotz *et al.*, 2012). Saurav *et al.* (2019) have confirmed the interaction of *B. tabaci* thioredoxin-like protein (TLP) with the CP of ToLCV by carrying out *in vitro* pull down experiments and dot blot assays. However, the exact role of TLP to be involved in virus transmission or inhibition is yet to be investigated. Similarly, many of the whitefly proteins either promote begomoviral transmission or reduce the viral transmission such as peptidoglycan recognition protein (PGRP) (Wang *et al.*, 2016), cyclophilin B (Kanakala & Ghanim, 2016), GroEL of the endosymbionts *Hamiltonella* and *Arseonophonus* (Morin *et al.*, 1999; Rana *et al.*, 2012), tumorous imaginal disc (TID) (Zhao *et al.*, 2020) interact with the CP of begomoviruses. All these reports show that CP of the begomovirus is responsible for circulative and persistent transmission of the virus via its vector.

Considering the importance of the CP from the earlier studies, we have carried out *in silico* comparison of the CP of TYLCV and ToLCV. The amino acid sequences of the CP of both TYLCV

and ToLCV are collected, followed by homology modelling for the prediction of 3D structure of the protein. The predicted 3D structure of the protein was used for molecular docking with GroEL of the whitefly endosymbiont.

## MATERIAL AND METHODS

### Collection of Amino Acid Sequences of TYLCV-CP, ToLCV-CP and the *Hamiltonella* GroEL Proteins

The amino acid sequences of CP of both TYLCV and ToLCV are collected from NCBI (<https://www.ncbi.nlm.nih.gov/>). The CP of the TYLCV carrying the accession number AXR75906.1 was retrieved. Similarly, the CP of ToLCV carrying the accession number BAP27993.1 was also retrieved. The amino acid sequence of the *Hamiltonella* GroEL was also retrieved from NCBI database with the accession number AFQ62604.1.

### Pair Wise Alignment of TYLCV-CP and ToLCV-CP

The collected sequences are subjected to pair wise alignment using Needleman-Wunsch algorithm ([https://www.ebi.ac.uk/Tools/psa/emboss\\_needle/](https://www.ebi.ac.uk/Tools/psa/emboss_needle/)).

### Homology Modelling of TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL Proteins

The retrieved amino acid sequences of TYLCV-CP and ToLCV-CP are used for the prediction of the three dimensional structure of the proteins on swiss-model (<https://swissmodel.expasy.org/>). CP of the geminivirus determined by electron cryo-microscopy with global model quality estimation (GMQE) score of 0.55 and identity *per cent* of 74.51 was used as the template for the prediction of 3D structure of ToLCV-CP. GMQE score ranges from 0 to 1 and higher the value better the reliability of the predicted structure. To predict the 3D structure of TYLCV-CP, near atomic resolution structure of a plant geminivirus determined by electron cryo-microscopy with the GMQE score of 0.54 and identity *per cent* of 81.46 was used as

template. 3D structure of *Hamiltonella* GroEL was also predicted in a similar way using GroEL of *Xanthomonas oryzae pv. oryzae* as template, which have GMQE value of 0.83 and the identity *per cent* of 73.90. All the structures are saved in PDB format.

### Validation of Predicted Protein Structure

Based on the geometry, geometry and the solvent potential of the protein model, the SWISS-MODEL web server automatically calculates the Qualitative Model Energy Analysis (QMEAN) score. The SWISS-MODEL also provides the Z-score which are compared with the expected value for any structure. The 3D structure of the proteins generated by SWISS-MODEL was checked for the quality using PROCHECK. This is done by uploading the .pdb file format obtained from SWISS-MODEL to PDBsum webserver. This provides the Ramachandran plot as well as the Ramachandran plot statistics. Ramachandran plot is used to assess the quality of the modelled protein and the Ramachandran plot statistics shows the total number of amino acid residues present in favourable, allowed and disallowed regions.

### Molecular Docking

The molecular docking studies were carried out using an online tool, H-dock (Yan *et al.*, 2020). The 3D structures generated by homology modelling on SWISS-MODEL were used for docking studies. TYLCV-CP was docked with *Hamiltonella* GroEL, similarly, ToLCV-CP was also docked with *Hamiltonella* GroEL. 10 different conformation were generated from docking tool and were ranked according to their binding energies.

## RESULTS AND DISCUSSION

### Pair Wise Alignment of TYLCV-CP and ToLCV-CP

The amino acid sequences of TYLCV-CP and ToLCV-CP retrieved from NCBI were used for pair wise alignment using Needleman-Wunsch algorithm. The alignment data is represented in Fig. 1.

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MSKRPGDIIISTPVSKVRRRLNFDSPYSTRAAVPIVQGTNKRRSWYRPM
|:| | | . | | | | . | . | | | | | | | : | . | | . | | | . : . | | . : : | . | | |
MAKRPADIIISIPASKVRRRLNFDNPNYGARAVVPIARVT-KAKAWTNRPM

YRKPRIYRMYRSPDVPRGCEGPKVQSYEQRDDIKHTGIVRCVS-----
. | | | | : | | | | | | | | | | | | | : | . | | | | | |
NRKPRMYRMYRSPDVPRGCEGPKVQSFESR-----ARCVSYWQSHV

---DVTRGSGITHRVGKRFCVKSIFYFLGKVVMDENIKKQNHNTNQVMFFLV
| | | | | : | : | | | | | | | | | | | | . | | | : | | | | | . : | | | | |
VLVDVTRGTGLTHRVGKRFCVKSIVYVLGKIWMDENIKTKNHNTNSVMFFLV

RDRRPGYSSPMDFGQVFNMFNEPSTATVKNDLRDRFQVMRKPFHATVIGG
| | | | . | | | . | | | : | | | | | | | | | | | | . . | | : | : | | : | | | . | |
RDRRPTG-SFQDFGEVFNMFNEPSTATVKNMHRDRYQVLRKWHATVTGG

PSGMKEQALVKRFFKINSHVVTYNHQEAQKYENHTENALLLYMACHASNP
. . . . : | | | | | : : | . : : : : . | | . | | . | | | | | | | | | | : | | | | | | | | |
TYASREQALVKRFVRVNNYVVYNQOEAGKYENHTENALMLYMACHASNP

VYATMKIRIYFYDSISN
| | | | : | | | | | | | : : |
VYATLKRIRYFYDSVTN

```

Fig. 1 : Pair wise alignment of amino acid sequences of TYLCV-CP and ToLCV-CP.  
 “|” indicates conserved residue; “-” indicates gap; “.” Indicates mismatch; “:” indicates substitutions

## Homology Modelling

3D structures of TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL were built using SWISS-MODEL. The GMQE score and QMEAN values of TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL are 0.74 and -2.66, 0.71 and -2.40 and 0.82 and -0.90, respectively. The GMQE score and QMEAN values of all the modelled structures indicate that the modelled structures are reliable and have good quality (Benkert *et al.*, 2009).

## Structure Validation of Modelled Protein

The predicted local similarity to target against the predicted 3D structure of TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL is represented in the Fig. 2A, 2B and 2C, respectively. The value of most of the residues is more than 0.6, indicating that the local quality estimates of the residues of the predicted models are good. According to the reports of Odukelu

*et al.* (2019), the residues having the value of less than 0.6 are considered to be of poor quality. The modelled structures of TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL lie within the range of other protein structures in protein data bank (PDB) indicating the reliability of the predicted models (Fig. 2D, 2E and 2F).

Further validation was carried out by uploading the obtained models (.pdb) to PDbsum web server. Ramachandran plot (Fig. 3A, B, C) and Ramachandran plot statistics were obtained from the server. According to Ramachandran plot statistics, the model predicted for TYLCV-CP had 88.7 per cent in the most favoured regions, 10.8 per cent in the additional allowed regions, 0.5 per cent in the generously allowed region and 0 per cent in disallowed region (Fig. 3D). The model predicted for ToLCV-CP had 91.4 per cent of residues in the most favoured regions, 7.6 per cent in the additional allowed

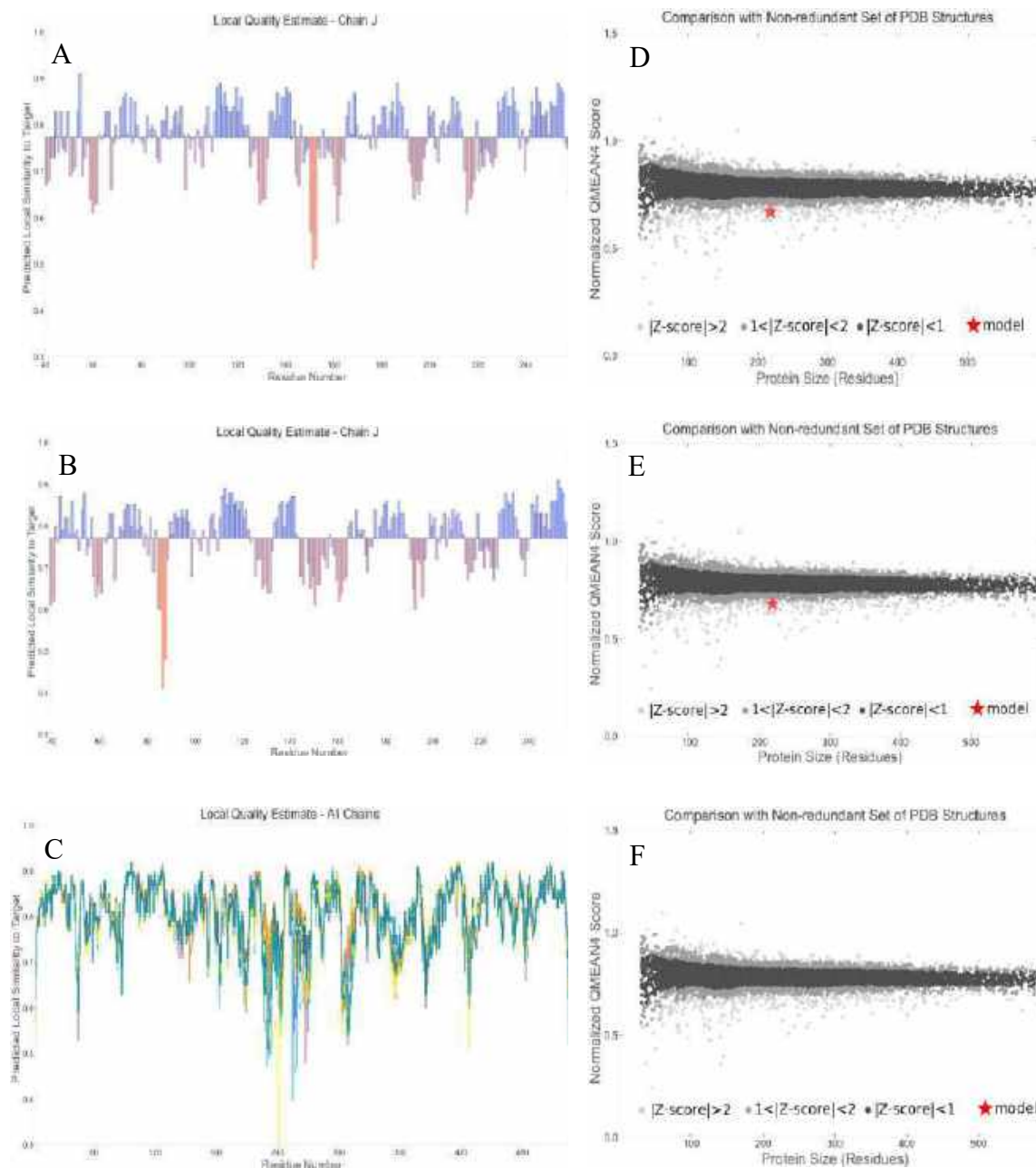


Fig. 2 : Structure validation of modelled TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL. A, B, C – local quality estimate of the residues of the predicted TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL; D, E, F - comparison of the predicted structures of TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL with nonredundant set of PDB structures

regions, 1 per cent in the generously allowed region and 0 per cent in disallowed region (Fig. 3E). For the model predicted for *Hamiltonella* GroEL, 91.2 per cent of residues in the most favoured regions, 6.6 per cent in the additional allowed regions, 1.1 per cent in the generously allowed region and 1 per cent in disallowed region (Fig. 3F). These results validate

that the predicted structures are good models (Oduselu *et al.*, 2019).

### Docking of TYLCV-CP and ToLCV-CP with *Hamiltonella* GroEL

The validated models of TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL obtained from SWISS-MODEL

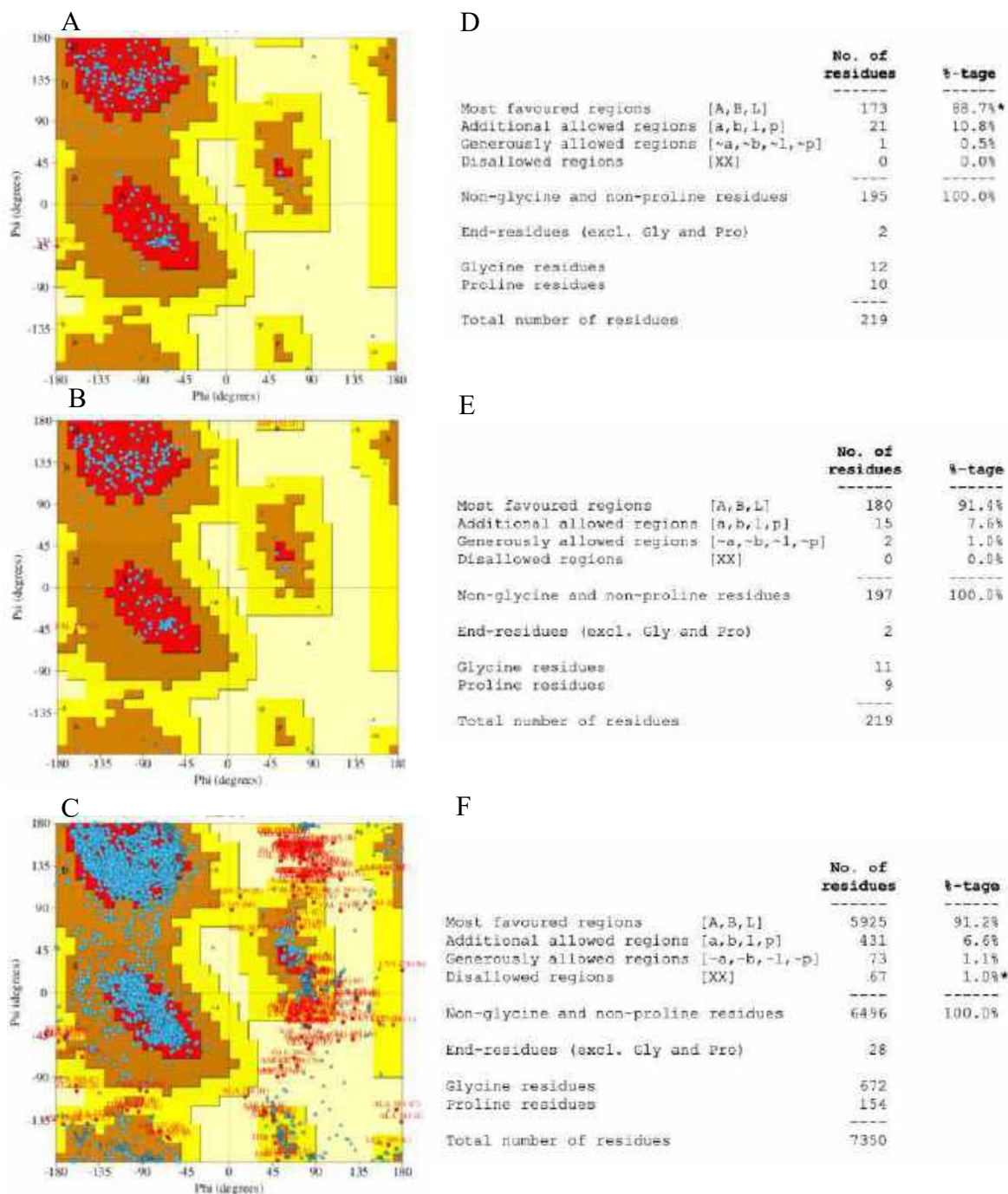


Fig 3. Structure validation using Ramachandran plot and Ramachandran plot statistics.

A, B, C – Ramachandran plots of TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL; D, E, F – Ramachandran plot statistics of TYLCV-CP, ToLCV-CP and *Hamiltonella* GroEL

were used for docking. TYLCV-CP and ToLCV-CP were docked with *Hamiltonella* GroEL, separately, using an online tool, H-dock. The ToLCV-CP and TYLCV-CP docked with *Hamiltonella* GroEL are

represented in the Fig. 4A and Fig. 4B. H-dock tool also lists the amino acid residues that are involved in protein-protein interaction. Table 1 and 2 lists out the amino acids involved during the interaction of



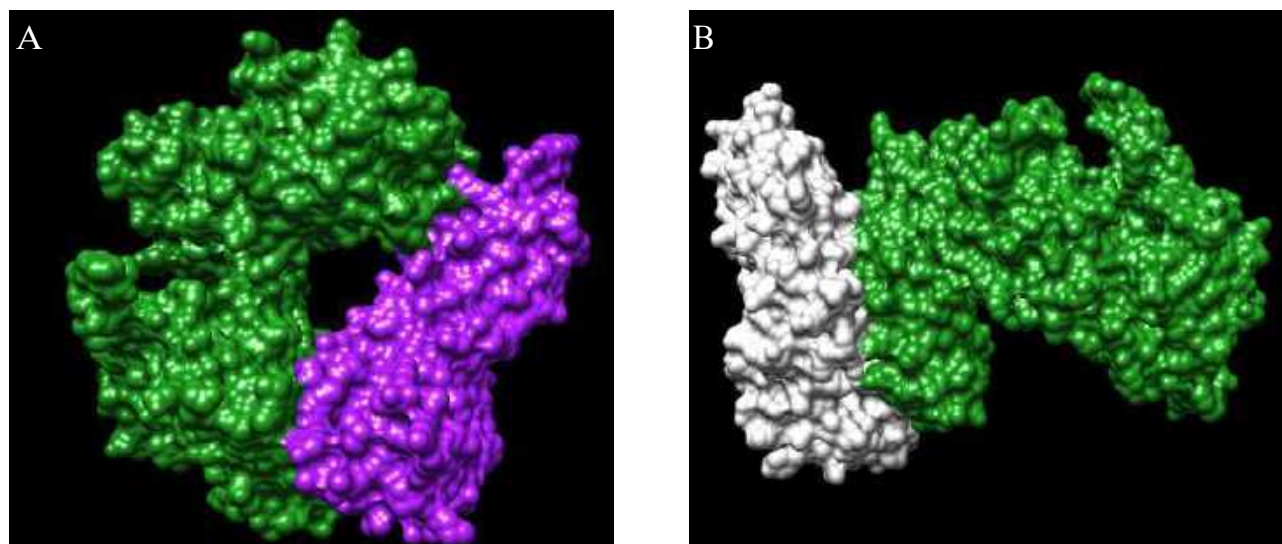


Fig. 4 : Docking of ToLCV-CP with *Hamiltonella* GroEL (A) and TYLCV-CP with *Hamiltonella* GroEL (B). Green colour structure - *Hamiltonella* GroEL; Purple colour structure - ToLCV-CP; grey colour structure - TYLCV-CP

TABLE 1  
Amino acids involved in interaction between ToLCV-CP and *Hamiltonella* GroEL

Amino acids of TYLCV-CP	Position of TYLCV-CP amino acids	<i>Hamiltonella</i> GroEL amino acids	Position of <i>Hamiltonella</i> GroEL amino acids
Pro	63	Asp	283, 316, 361
Asp	64	Lys	286, 297, 311, 343,
Arg	67, 142, 144, 182, 203	Ala	287, 293, 299, 356
Gly	68	Gln	290, 348, 351, 352
Tyr	116	Ile	294, 301, 305, 342
Gln	179	Gly	298, 306
Val	180	Val	300
Met	181	Ser	302, 340, 344, 358
Lys	183, 202, 206	Met	307
Phe	184, 204, 205	Glu	308, 315, 338, 347,355
His	185, 210	Thr	313, 341, 357
Ala	186	Arg	345, 353, 362
Thr	187	Leu	365
Leu	200		
Ile	207		
Asn	208		
Ser	209		
Glu	226		

TABLE 2  
Amino acids involved in interaction between TYLCV-CP and *Hamiltonella* GroEL

ToLCV-CP amino acids	Position of ToLCV-CP amino acids	<i>Hamiltonella</i> GroEL amino acids	Position of <i>Hamiltonella</i> GroEL amino acids
Pro	63	Asp	283, 316, 361
Asp	64	Lys	286, 297, 311, 343
Arg	67, 203	Ala	287, 293, 299
Gly	68	Gln	290, 348, 351, 352
Tyr	116	Ile	294, 301, 305, 342,
Gln	179	Gly	298, 306
Val	180	Val	300
Met	181	Ser	302, 340, 344, 358
Lys	183	Met	307
Phe	184	Glu	308, 315, 338
His	185	Thr	313, 341, 357
Ala	186	Arg	345, 353, 362
Thr	187	Glu	347, 355
Leu	200	Ala	356
Phe	204, 205	Leu	365
Ile	207		
Asn	208		
Ser	209		
His	210		
Glu	226		

ToLCV-CP with *Hamiltonella* GroEL and TYLCV-CP with *Hamiltonella* GroEL, respectively. The interaction between TYLCV-CP protein and *Hamiltonella* GroEL is stronger in comparison with the interaction between ToLCV-CP and *Hamiltonella* GroEL. The number of amino acids of TYLCV-CP that are involved in interaction with *Hamiltonella* GroEL are more than that of ToLCV-CP. Earlier reports by Czosnek *et al.* (2017) supports our results that TYLCV-CP interacts with GroEL of *Hamiltonella*.

This study shows that there are similarities among the CP of TYLCV and ToLCV as shown by Needleman-Wunsch based pair wise alignment. Though there are similarities, the interaction of TYLCV-CP and ToLCV-CP with *Hamiltonella* GroEL studied by protein-protein docking shows differences

in the way they interact with GroEL protein. GroEL proteins of many other whitefly endosymbionts such as, *Rickettsia* and *Arsenophonus* are reported to interact poorly with TYLCV-CP (Gottlieb *et al.*, 2010). This ability of the begomoviruses, especially TYLCV-CP, to interact with various proteins of whitefly and its endosymbionts has made it one of the devastating disease on tomato leading to huge crop loss.

Molecular docking studies can be used to study the protein-ligand as well as protein-protein interactions. These studies can be performed to know how many/ what are the amino acids that are involved in interaction. Also, docking studies reveals how strongly the two molecules are interacting with each other. Further, docking studies can be exploited to know what are the whitefly proteins interacting with CP of

other group of viruses. This help us to understand the pathway that virus follows within a whitefly and will probably be helpful in understanding to prevent plant viral infection, leading to improved crop yield.

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## Profile Characteristics, Constraints and Suggestions of Farm Youth Practicing Family Farming in Parbhani District of Maharashtra

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### ABSTRACT

Family farming plays an important role in Indian agriculture with its potential in eradication of poverty with various cost effective, resource sufficient aspects along with income gaining, socio-economic and psychological aspects. Therefore, studying the comprehensive nature of the farm youth through their profile characteristics, make a significant contribution in this field. The systematic study and analysis of the profile characteristics of farm youth provide an idea on the extension activities to be conducted to improve the knowledge, skill and attitude of the young farmers of Parbhani district. Two taluks were selected and from each taluk four villages were selected based on the maximum number of farm youth involved in family farming. In each village the list of farm youth practicing family farming was prepared in consultation with extension personnel and 20 respondents were selected from each village by using snowball technique, thus, making total sample of 160. The study revealed that majority of the respondents belonged to middle age category, farming experience at medium level with the education level up to pre-university level. More than half of the respondents belonged to medium level of psychological characteristics. Majority of farm youth families belonged to small size of land holding (60.62%), medium level of livestock units (46.25%) and materials (55.00%). The major constraint faced by the farm youth lack of infrastructure facilities (Transportation, Electricity, Storage *etc.*) and they have suggested to provide proper infrastructure facilities (Transportation, electricity, storage *etc.*).

**Keywords :** Constraints, Family farming behaviour, Farm youth, Profile characteristics

INDIA is predominantly an agriculture dependent nation where 54.60 per cent of its population is engaged in agriculture (Anonymous, 2021). Our country is the youngest nation in the world with 40.00 per cent of the population falling in the youth category and 67-68 per cent of them live in rural areas (Rajendran and Paul, 2020). Agriculture sector being a largest employer for rural youth, many young farmers engage in high-tech, high risk and high-returns agri-ventures. More than 85 per cent of farmers belonged to small (1.00 to 2.00 ha) and marginal farmers (< 1.00 ha), majority of them are practicing family farming for their livelihood in India (Anonymous, 2020). In relation to this, family farming

is a means an agricultural holding which is managed and operated by a household and where farm labour is largely supplied by that household. Family farming behaviour is the totality of behaviour of a farm youth in relation to his farming activities. Family farms contribute majorly to economy of the nation as it constitutes 85 per cent of total agricultural holdings in the country and 60 per cent of the production comes from these family farms (Bitan *et al.*, 2016). A family farm is 'managed and operated by a family and predominantly reliant on family labour, including that of both women and men'. The family farming assumes the greater importance for sound management of farm resources to enhance the farm productivity

and reduce the environmental degradation; improve the quality of life of resource poor farmers and to maintain sustainability.

In India, youth are major producer of food in terms of value, volume and number of hours worked because agriculture is largely house hold enterprise. Farm youth has either direct or indirect effect on the family farming behaviour. The constraints and suggestions with respect to farm youth practicing family farming helps in understanding predisposition of farm youth to participating in family farming activities and making it more economical and profitable.

### METHODOLOGY

The study was conducted in purposively selected Parbhani district of Maharashtra state during 2022-23. Out of nine taluks, two taluks namely,

Parbhani and Jintur were selected based on the maximum number of farm youth involved in Family Farming, in consultation with extension personnel of development departments. Further, from each taluk, four villages were selected. From each village, the list of farm youth practicing family farming was prepared in consultation with extension personnel and 20 farm youth from each village were selected by using snowball technique, thus, making a total sample 160 respondents. Data were gathered through personal interview method with the help of structured pre-tested interview schedule. The collected data were quantified and analysed using frequencies, percentages, mean and standard deviation. The personal, psychological and socio-economic characteristics were categorised as low, medium and high based on the mean and standard deviation. Constraints and suggestions were ranked based on mean scores.

TABLE 1  
Personal characteristics of the respondents

Characteristics	Mean	SD	Level	f	%
Age (Farm Youth)	-	-	Young (18-25 yrs.)	39	24.37
			Middle (26-30yrs.)	89	55.63
			Adult (31-35yrs)	32	20.00
Education	-	-	Illiterate	00	00.00
			Read & write	00	00.00
			Primary school	17	10.62
			Middle school	23	14.38
			High school	39	24.38
			PUC	52	32.50
			Diploma	00	00.00
			Degree	29	18.12
			PG	00	00.00
Family size	-	-	Small (<5 members)	39	24.38
			Medium (5-8 members)	74	46.24
			Large (>8 members)	47	29.38
Farming experience (years)	9.86	1.62	Low (< 9.05)	25	15.63
			Medium (9.05- 10.67)	98	61.25
			High (>10.67)	37	23.12
Livestock Rearing Experience (years)	7.73	1.40	Low (<7.03)	51	31.88
			Medium (7.03-8.43)	73	45.62
			High (>8.43)	36	22.50

## RESULTS AND DISCUSSION

### Profile Characteristics of the Farm Youth practicing Family Farming

The results of personal characteristics of the farm youth were presented in Table 1. The results revealed that more than half (55.63%) of the respondents belonged to middle aged, followed by young (24.37%) and adult (20.00%) aged youth categories. Nearly 32.50 per cent of the farm youth had completed their education up to pre-university level, followed by high school (24.38%), middle school (14.38%), 18.12 per cent were graduates and 10.62 per cent of them completed primary schooling. Majority of farm youth belonged to medium family size (46.24%) followed by large (29.38%) and small (24.38%) sized family. Majority of the farm youth had medium level of farming experience (61.25%), followed by high (23.12%) and low (15.63%) level. The 45.62 per cent of farm youth were experienced at medium level followed by low (31.88%) and high (22.50%) level in livestock farming.

Majority of the enthusiastic and efficient middle aged youth belonged to farming background which made them to build their experience in farming including livestock rearing. They were able complete their education from high school to pre-university level. The reason might be due to functioning of government and private aided schools, few of the respondents would have studied in colleges situated in nearby towns. As majority of them belonged to medium sized family and all their efforts were concentrated towards farming and family welfare at early age rather than higher education. Hence, the farm youth (at the age of 26 to 30 years) were experienced in farming, livestock rearing at medium level with their affordable education level. Similar findings were reported by Dhanashree *et al.* (2014), Harshitha (2018) and Sampraja (2022).

The result of psychological characteristics of the farm youth were presented in Table 2 and it is revealed that more than half (60.00%) of the respondents belonged to medium level of extension orientation, followed by low (26.25%) and high (13.75%) levels

of extension orientation. Majority of the respondents belonged to medium level (45.63%) mass media use, followed by 35.00 per cent and 19.37 per cent of high and low level, respectively. More than half of the farm youth had medium level marketing orientation (60.62%), followed by 30.00 per cent and 9.38 per cent of them belonged to low and high level, respectively. About 46.24 per cent, 29.38 per cent and 24.38 per cent of them had medium, high and low level of scientific orientation, respectively. About half of the farm youth (50.00%) had high level of achievement motivation followed by low (35.00%) and medium (15.00%) level. Majority (68.12%) of respondents had medium level of credit orientation followed by 28.76 per cent and 03.12 per cent of them had low and high level, respectively. Deferred gratification of respondents ranged from medium level (50.62%) to low level (29.38%) and 20.00 per cent of high level.

The reason for the medium level of psychological characteristics of farm youth might be enthusiasm, experience and interest of the farm youth in finding new things through extension personnel contact, participating in various social and extension activities and use of different mass media. Education level and need for the modern technologies attract and motivate the farm youth towards building up of regular extension contacts. Lack of awareness on extension services or dearth of the interest in consulting the extension officers/ agents created a number of the farm youth to lower extension orientation. Due to the knowledge and awareness about credit institutions (like FPOs, SHGs, Government Schemes *etc.*) and acceptance of financial assistance by farm youth had progressed compared to age-old farmers. Similar findings were reported by Gopala (2006), Ereneus (2010), Yashodhara (2015) and Harshitha (2018).

The result of socio-economic characteristics of the farm youth were presented in Table 3. About 39.38 per cent of farm youth annual income belonged to medium level of annual income, whereas 31.24 per cent belonged to high level and 29.38 per cent belonged to low level of annual income. About 60.62 per cent of respondent families were belonged small

TABLE 2  
Psychological characteristics of the respondents

Characteristics	Mean	SD	Level	f	%
Extension Orientation	5.79	1.44	Low (<5.07)	42	26.25
			Medium (5.07- 6.51)	96	60.00
			High (>6.51)	22	13.75
Mass Media Use	18.76	1.37	Low (<18.07)	31	19.37
			Medium (18.07-19.44)	73	45.63
			High (>19.44)	56	35.00
Marketing Orientation	15.72	1.60	Low (<14.92)	48	30.00
			Medium (14.92-16.52)	97	60.62
			High (>16.52)	15	09.38
Scientific Orientation	16.66	1.15	Low (<16.08)	39	24.38
			Medium (16.08-17.23)	74	46.24
			High (>17.23)	47	29.38
Achievement Motivation	27.40	1.66	Low (<26.57)	56	35.00
			Medium (26.57-28.23)	24	15.00
			High (>28.23)	80	50.00
Credit Orientation	1.75	0.52	Low (<1.49)	46	28.76
			Medium (1.49-2.01)	109	68.12
			High (>2.01)	05	03.12
Deferred Gratification	38.90	2.50	Low (<37.65)	47	29.38
			Medium (37.65-40.15)	81	50.62
			High (>40.15)	32	20.00

TABLE 3  
Socio- economic characteristics of the respondents

Characteristics	Mean	SD	Level	f	%
Annual Income	68793.75	16497.03	Low (<60545.24)	47	29.38
			Medium (60545.24-77042.26)	63	39.38
			High (>77042.26)	50	31.24
Land Holding	-	-	Marginal (<2.5 acres)	35	21.88
			Small (2.5-5 acres)	97	60.62
			Big (>5 acres)	28	17.50
Livestock Possession	11.66	5.46	Low (<8.93)	55	34.38
			Medium (8.93-14.39)	74	46.25
			High (>14.39)	31	19.37
Material Possession	11.75	1.22	Low (<11.14)	32	20.00
			Medium (11.14 -12.36)	88	55.00
			High (>12.36)	40	25.00



land holder's category followed by 21.88 per cent marginal and 17.50 per cent big holding categories. About 46.25 per cent of farm youth possessed medium level of livestock, 34.38 per cent possessed low and 19.37 per cent high level. About 55.00 per cent of farm youth possessed medium, 25.00 per cent possessed high and 20.00 low level of materials.

The probable reason for the above findings might be attributed for diverse annual income groups of respondents, due to the size of the land holding, subsidiary occupations like dairy farm, poultry and fishery by the respondents. Another reason might be majority of them were educated and can assume economic aspects of various units. The fragmentation of their ancestral land might have resulted to smaller size of land holdings. To carry out the farming efficiently respondents owned several materials like plough, tractor, pump set etc. Similar findings were reported by Malik (2010); Jyoti (2012) and Harshitha (2018).

### Constraints Faced by Farm Youth and their Suggestions with Respect to Family Farming Behaviour

Constraints faced by farm youth with respect to family farming behaviour are represented in Table 4. The major constraints faced by the farm youth were lack of infrastructural facilities (Transportation, Electricity,

Storage etc.) (rank I), followed by non-availability of quality inputs (like seeds and fertilizers) in time (rank II) and lack of market facilities (rank III).

Suggestions given by farm youth to improve family farming behaviour are represented in Table 5. The major suggestions given by the farm youth were provide proper infrastructure facilities (Transportation, Electricity, Storage etc.) (Rank I), timely supply of necessary inputs (seeds/ planting material/ breeds/species/fertilizers) (Rank II) and provide timely market information and facilities at local level (Rank III).

The reasons might be due to lack of sufficient facilities to transport the products, irregular electricity supply which pose difficulties in irrigation, storing of products resulting in wastage of products and bring less or no profit to farm youth. Non-availability of timely and quality seeds and fertilizers make the farm youth difficult in farming. Further, to get better prices for their produce they have suggested to provide the marketing facilities at local level which include storage and transportation facilities. Supply of necessary inputs in time to take up activities. Further, organizing need based training programmes to increase knowledge and skills to solve the field problems. Similar findings were reported by Madhu (2010) and Saha & Bahal (2010).

TABLE 4  
Constraints faced by farm youth with respect to family farming behaviour

Constraints	Number	Percentage	Rank
Lack of infrastructure facilities (Transportation, Electricity, Storage etc.)	148	92.50	I
Non availability of quality inputs (like seeds and fertilizers) in time	143	89.38	II
Lack of market facilities	135	84.37	III
Complicated procedure to get loan	128	80.00	IV
Poor accessibility of extension agencies for technical guidance	121	75.63	V
Failure of crops (Hailstorm, Pest and Diseases etc.)	118	73.75	VI
Poor water management	111	69.38	VII
Inadequate support from officials of agriculture & other departments	108	67.50	VIII
Lack of credit to invest on other income generating activities	101	63.13	IX
Skill requirement of farm family	97	60.63	X
High cost of production and lower returns	89	55.62	XI

TABLE 5  
Suggestions of farm youth to improve family farming behaviour (n=160)

Suggestion	Number	Percentage	Rank
Provide proper infrastructure facilities (Transportation, Electricity, Storage etc.)	152	95.00	I
Timely supply of necessary inputs (seeds/ planting material/ breeds/ species/fertilizers)	147	91.88	II
Provide timely market information and facilities at local level	145	90.63	III
Provision of easy, timely and adequate credit at lower interest rate	138	86.25	IV
Extension personnel should visit their farm once in a week during crop season	131	81.88	V
A comprehensive crop insurance scheme to protect farmers not only from vagaries of nature but also from market fluctuations	124	77.50	VI
There is need for fixing minimum support price for all the crops	119	74.38	VII
Provide adequate support from the department officials	110	68.75	VIII
Provide credit timely with subsidy (financial support)	102	67.50	IX
Effective extension methods (field days, result demonstration, method demonstration etc.) should be conducted periodically	98	61.25	X

The involvement of youth in agriculture plays major role to bring potential change as they are more productive and receptive to new ideas and advanced technologies. Their risk taking ability and their inclination towards modernization might help to make farming as profitable enterprise. The study revealed that 55.63 per cent of the respondents belonged to middle age category, 32.50 per cent completed their education up to pre-university level, 61.25 per cent had medium level farming experience. More than half of the respondents belonged medium level of extension orientation (60.00%), credit orientation (68.12%) and deferred gratification (50.62%). Farm youth owned small land holding (60.62%), medium level of livestock units (46.25%) and materials (55.00%). The major constraint faced by farm youth lack of infrastructure facilities (Transportation, Electricity, Storage etc.) was (rank I) and they have suggested to provide proper infrastructure facilities (Transportation, Electricity, Storage etc.) (rank I). The results of the study implied the need for planning and organizing the extension education programmes and activities for farm youth by selecting them considering their major profile

characteristics, constraints and suggestions to increase the family farming behaviour.

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## Studies on the Effect of White Muscardine Infection on Growth and Yield Performance of Bivoltine Silkworm, *Bombyx mori* Breeds

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### ABSTRACT

To know the possibilities of tolerance for both abiotic and biotic stresses in silkworm, *Bombyx mori*, an investigation was conducted to assess the growth and yield performance of thermotolerant bivoltine silkworm breeds inoculated with *Beauveria bassiana*. Three muscardine resistant thermotolerant bivoltine breeds viz., B1, B4, B8 and muscardine susceptible but productive breed CSR<sub>4</sub> were selected and topically inoculated with the fungal spore suspension of *Beauveria bassiana* ( $6.86 \times 10^4$  spores/ml). The results revealed that the thermotolerant bivoltine silkworm breed B1 appeared to be more tolerant to fungal infection followed by B4 breed. The quantitative traits in B1 thermotolerant bivoltine silkworm breed under muscardine inoculation treatment revealed significantly longer larval duration (7.44 days) and highest fifth instar larval weight (37.28 g), pupation rate (68.12%) and cocoon yield by weight (14,731.75/10,000 worms), cocoon weight (1.49 g), pupal weight (1.16 g) and shell weight (0.31 g), as compared to other breeds such as B4 and B8. While least larval mortality (40.75%) and highest effective rate of rearing (59.25%), cocoon yield by number (5,925 per 10,000 worms) and shell ratio (22.64%) were recorded in B4 breed.

Keywords : Thermotolerance, Bivoltine, *Bombyx mori* L, *Beauveria bassiana*

INDIA occupies the second position in global silk production next only to China. Sericulture in India is being practiced predominantly in tropical regions and to limited extent in temperate region. The existing tropical situation in the country provides scope for the exploitation of multivoltine breeds / hybrids as these breeds show the inherent capacity to perform well under varied and fluctuating environmental conditions. But the quality of multivoltine silk is low compared to the international standards. To meet this standard it is necessary to shift to bivoltine sericulture which assures the production of quantitatively and qualitatively superior cocoons (Sahana *et al.*, 2021).

The silkworm, *Bombyx mori* L. is delicate, sensitive and completely domesticated insect and classic model

organism for lepidoptera. The success of cocoon production depends on disease management. Exploitation of the resistant of silkworm breeds towards different diseases causing pathogens is a better option for managing the crop loss. Diseases in silkworm are fairly common in occurrence inflicting serious losses. Four major categories of disease viz., the microsporidian, viral, bacterial and fungal diseases, which are popularly known as pebrine, grasserie, flacherie and muscardine, respectively, cause damage to silkworms in different seasons. Among these diseases white muscardine caused by *Beauveria bassiana* (Bals.) Vuill. is one of the most devastating silkworm disease common during winter and rainy seasons. In India 10-40 per cent of the total

loss due to disease has been accounted for white muscardine (Janakiraman, 1961; Chandrasekharan and Nataraju, 1998). The climatic condition in the tropics is congenial for the occurrence and easy spread of fungal diseases and spreads easily under high humidity and low temperature conditions (Samson *et al.*, 1990).

It is a well-established fact that, unlike multivoltine silkworms, bivoltines are more vulnerable to different stresses under tropical condition as these bivoltines have originated from temperate region. It is, therefore, imperative to evolve bivoltine silkworm breeds which can give stable yields under both abiotic and biotic stress conditions (Sahana *et al.*, 2021). Keeping this in view, ten thermotolerant bivoltine silkworm breeds *viz.*, B1, B2, B3, B4, B5, B6, B8, APS12 and APS45 under muscardine infection revealed that, B4 breed performed better with respect to cocoon weight, pupal weight, shell weight, shell ratio, filament length and filament weight, followed by B1 and B8 breeds (Keerthana *et al.*, 2019). In order to assess these

thermotolerant silkworm breeds for their physiological responses under muscardine infection, a preliminary assay was done in the present study.

## MATERIAL AND METHODS

The layings of three thermotolerant bivoltine silkworm breeds, *i.e.*, B1, B4 and B8 identified as muscardine resistant from the previous studies (Keerthana, 2018; Sreejith Vakayil, 2019; Jayashree, 2019 and Sahana, 2021) and susceptible bivoltine silkworm breed CSR<sub>4</sub>, were utilized for the present study. The characteristic features of these breeds are given in Table 1.

Larval and cocoon parameters of the thermotolerant bivoltine silkworm breeds used in the experiment.

### Rearing of Silkworm Breeds

Silkworm rearing was conducted during July 2020-April 21 at the Department of Sericulture, UAS, GKVK, Bengaluru. Silkworms were reared in following the standard rearing practices (Dandin

TABLE 1  
Effect of *B. bassiana* infection on growth and survival of thermo tolerant bivoltine silkworm breeds

Breeds	Fifth instar larval Duration (days)		Fifth instar larval weight (g/10 larvae)		Effective rate of rearing (%)		Pupation rate (%)	
	Control	<i>B. bassiana</i> infected	Control	<i>B. bassiana</i> infected	Control	<i>B. bassiana</i> infected	Control	<i>B. bassiana</i> infected
B1	7.29	7.44 <sup>b</sup> (+2.06)	39.17	37.28 <sup>a</sup> (-4.82)	100	58 <sup>a</sup> (-42)	100	68.12 <sup>a</sup> (-31.88)
B4	7.21	7.27 <sup>b</sup> (+0.83)	38.38	33.79 <sup>b</sup> (-11.95)	100	59.25 <sup>b</sup> (-40.75)	100	63.75 <sup>b</sup> (-36.25)
B8	7.33	7.41 <sup>b</sup> (+1.09)	29.66	22.94 <sup>d</sup> (-22.65)	100	44.25 <sup>c</sup> (-55.75)	100	51.87 <sup>c</sup> (-48.13)
CSR4	8.39	8.47 <sup>a</sup> (+0.95)	31.12	27.29 <sup>c</sup> (-12.30)	100	18.38 <sup>d</sup> (-81.62)	100	26.87 <sup>d</sup> (-73.13)
F TEST	**	**	**	**	NA	**	NA	**
SEm±	0.08	0.05	0.09	0.25	-	0.04	-	0.65
CD at 5%	0.26	0.13	0.29	0.79	-	1.11	-	2
CV (%)	2.23	1.17	0.54	1.69	-	1.16	-	2.47

Note: Positive and negative figures in the parenthesis indicate per cent increase (+) or decrease (-) over ; control, respectively; \*\* - Significant @1%; NA – Not analysed

Genotypes	Breed traits
B1	Plain larva spinning oval shaped cocoon
B4	Plain larva spinning oval shaped cocoon
B8	Marked larva spinning peanut cocoon
CSR <sub>4</sub>	Plain larva spinning oval shaped cocoon

and Giridhar, 2014) on V-1 mulberry leaves till spinning. Newly ecdysed fifth instar silkworms (50 silkworms per replication in four replication each) were topically inoculated with *B. bassiana* spore suspension with  $6.86 \times 10^4$  spores per ml at the rate of 0.5 ml per silkworm (Keerthana *et al.*, 2019). Simultaneously, a control batch of all the four breeds was also maintained. Three such rearings was conducted and pooled mean data on fifth larval duration, larval weight, larval mortality, ERR, cocoon yield by number and by weight, cocoon weight, pupal weight, shell weight, pupation rate and shell ratio were recorded. The data so collected was analysed using a completely randomised design (Sundarraaj *et al.*, 1972).

## RESULTS AND DISCUSSION

### Fifth Instar Larval Duration (Days)

Significant differences were observed among the breeds utilized under *B. bassiana* inoculation with respect to larval duration. The fifth instar larval duration was determined from first day of fifth instar till spinning both under normal condition and muscardine inoculation (Table 1).

Among the thermotolerant bivoltine breeds, under normal condition the shorter larval duration was observed in B4 (7.21 days), followed by B1 (7.29 days) and longer larval duration was recorded in B8 (7.33 days) and CSR<sub>4</sub> (8.39) breeds. In *B. bassiana* inoculated batch, CSR<sub>4</sub> breed showed maximum larval duration of 8.47 days, followed by B1 and B8 (7.44 and 7.41 days) and the minimum in B4 breed (7.27 days). In the pathogen inoculated batch, higher increase in larval duration was observed in B1 breed (2.06%) and B8 (1.09%) breeds. While least was seen in B4 (0.85%) breed (Table 2). In a similar study, four thermotolerant silkworm breeds (B1, B4, B6 and B8) and their hybrids were inoculated with different dilutions ( $10^{-2}$ ,  $10^{-4}$ ,  $10^{-6}$ ,  $10^{-8}$ ) of *B. bassiana* where

TABLE 2  
Cocoon yield in thermo tolerant bivoltine silkworm breeds subjected to *B. bassiana*

Breeds	Cocoon yield per 10,000 larvae			
	By number (No.)		By weight (g)	
	Control	<i>B. bassiana</i> infected	Control	<i>B. bassiana</i> infected
B1	10000	5,800	19,349.12	14731.75
B4	10000	5,925	18,620.25	12551.00
B8	10000	4,425	13,292.50	11518.25
CSR4	10000	1,837	16,115.12	9681.80
F TEST	NA	**	**	**
SEm±	-	36.08	33.79	53.36
CD at 5%	-	111.197	104.116	164.45
CV (%)	-	0.160	0.40	0.881

Note: \*\* - Significant @1% ; NA- Not analysed

in B6 (10.58 days) breed, B6 x B1 and B6 x B8 (10.50 days each) hybrids showed prolonged larval duration over all the dilutions compared to control (Jayashree *et al.*, 2020b). In fungus inoculated batch the fifth instar larval duration was extended and the extent of prolongation was higher in B1 breed. (7.44 days). Results of the current study with respect to prolongation of larval duration are in conformity with the findings of Manjunath Gowda *et al.* (2011) under *BmNPV* infection and Keerthana *et al.* (2019) and Sahana *et al.* (2021) under muscardine infection. The prolongation of larval duration is due to reduced metabolic activity in the infected silkworms (Janakiraman *et al.*, 1961). Least prolongation of larval duration is desirable which evolving muscardine resistance breeds and hence B4 breed is suitable.

### Fifth Instar Larval Weight (g)

Fifth instar larval weight was affected significantly among all the thermotolerant bivoltine silkworm breeds due to *B. bassiana* inoculation (Table 2). In the normal batch, among thermotolerant bivoltine silkworm breeds the maximum fifth instar larval weight of 39.17 g/10 larva was observed in B1, followed by B4 (38.38 g/10 larva) and minimum larval weight was recorded in B8 (29.66 g/10 larva) followed by CSR4 (31.12/10 larva g). In muscardine treated batch (Table 1) the larval weight was significantly higher in muscardine resistant thermo tolerant bivoltine breed B1 (37.28 g/10 larva), followed by B4 (33.79 g/10 larva) and lowest larval weight was observed in B8 (22.94 g/10 larva), followed by CSR<sub>4</sub> (27.29 g/10 larva) breeds. Decrease in body weight in *B. bassiana* infected silkworms is due to cessation of feeding, decrease in food consumption, digestion, relative consumption rate and efficiency of conversion of ingested food (Venkataramana Reddy, 1978 and Cai, 1989). In earlier studies B4, B2 and B1 exhibited highest larval weight (21.35 g/10 larvae, 20.78 g/10 larvae and 20.50 g/10 larvae, respectively) under *B. bassiana* inoculation (Keerthana *et al.*, 2020) which supports the present findings. Further, the larval weight was significantly reduced under muscardine inoculation. Among the breeds, the thermotolerant breed B1 lost its body weight from 42.27 g (control)

to 35.39 g (muscardine treated) which was the least reduction in larval body weight (Jayashree *et al.*, 2020b). Results of the current study are in conformity with the earlier findings in thermotolerant bivoltine breeds under muscardine inoculation.

### Larval Mortality (%)

Significant differences were observed for larval mortality among the four thermotolerant bivoltine breeds under muscardine inoculation (Fig.1). In the batch without any fungal treatment, all the four thermotolerant bivoltine breeds had no mortality. Under *B. bassiana* infection, larval mortality among thermotolerant bivoltine breeds was significantly least in breed B4 (40.75%), followed by B1 (42.00%), B8 (55.75%) and maximum larval mortality was observed in CSR<sub>4</sub> (81.63%) (Fig.1).

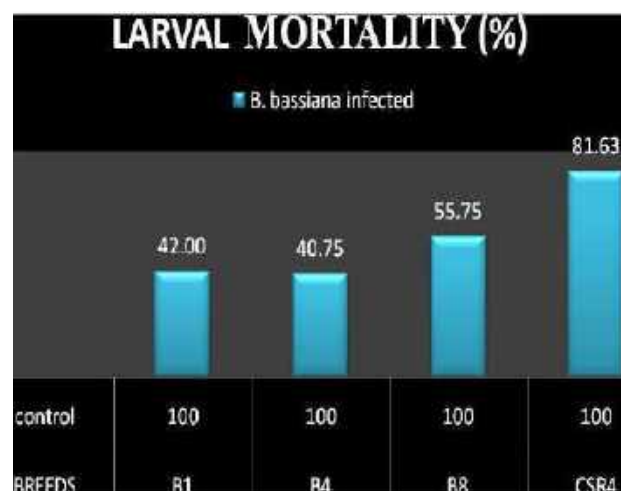


Fig1. Larval mortality (%) of the thermotolerant bivoltine silkworm breeds inoculated by *B. bassiana* ( $6.86 \times 10^4$  spores/ml @ 0.5 ml per/silkworm)

According to Sreejith vakayil (2019), maximum larval mortality was recorded in breed B3 (75%), followed by B8 (72%), B5 and B6 (70% each) and B7 (67%) and minimum larval mortality was recorded by breed B1 (41%) and B2 (57%) under *B. bassiana* inoculation. The thermotolerant bivoltine breeds used in this study also showed variation in the larval mortality due to fungal infection. B4 breed showed minimum larval mortality when infected with fungal pathogen.

### Effective Rate of Rearing (%)

Effective rate of rearing (ERR) among the muscardine resistant thermotolerant bivoltine breeds and muscardine susceptible productive bivoltine breed showed non-significant differences under normal condition and significant differences under muscardine inoculation (Table 1).

In muscardine treated batch, ERR was significantly different among the breeds (Table 1). Among the thermotolerant bivoltine silkworm breed B4 recorded highest ERR of 59.25 per cent, followed by B1 (58.00%) and lowest ERR was observed in B8 (44.25%) and CSR<sub>4</sub> (18.38%) breeds. Previously, when eight races of silkworms *viz.*, Pure Mysore, Hosa Mysore II, C-Nichi, HS<sub>6</sub>, NN<sub>6</sub>D, NB<sub>4</sub>D<sub>2</sub>, KA, J<sub>122</sub> were inoculated with nine conidial concentrations (10<sup>1</sup> - 10<sup>9</sup> spores / ml) of *B. bassiana*, variation in ERR over spore concentration and between the breeds was observed (Venkataramana Reddy, 1978). Infection of the thermotolerant silkworm breeds with *B. bassiana* resulted in highest ERR in B4 (54.67%) breed (Keerthana *et al.*, 2020). So also, the breed B4 performed better with respect to ERR under *B. bassiana* inoculation in the present study. Results of the current study are also in conformity with the earlier findings of Jayashree *et al.* (2020a) and Sahana *et al.* (2021), who reported better performance of B4 & B1 breeds, respectively, under *B. bassiana* infection.

### Pupation Rate (%)

There was no significant difference among the breeds in control batch for pupation rate (Table 1). The thermotolerant bivoltine silkworm breeds inoculated with fungal spores of *B. bassiana* showed significant differences in pupation rate (Table 1). Among the treated batch the maximum pupation rate was recorded in the breed B1 (68.12%) followed by B4 (63.75%) and B8 (51.87%). Minimum pupation rate was observed in breed CSR<sub>4</sub> (26.87%). The literature pertaining to the effect of muscardine infection in silkworms on pupation rate is rather limited. However, Chandrashekar *et al.*, (2006) have observed reduced pupation rate in silkworms infected with *BmNPV* and

the reduction was lesser at lower dose of inoculation of viral inoculation and comparatively higher at the higher dose. Pupation rate was significantly affected by the *BmNPV* infection in different silkworm genotypes. Multivoltine breeds, Pure Mysore, Nistari and C-Nichi and diapausing breed Diazo showed comparatively higher pupation rate than bivoltine breeds CSR<sub>2</sub> and CSR<sub>4</sub> at all doses of *BmNPV* infection (Manjunath Gowda, 2011).

### Cocoon Yield by Number (No. / 10,000 worms)

Thermotolerant bivoltine silkworm breeds when treated with *B. bassiana* showed significant differences for cocoon yield by number per 10,000 worms (Table 2). Under fungal stress, significantly highest number was recorded in breed B4 (5,925/10,000 worms), followed by B1 (5,800/10,000 worms) and B8 (4,425/10,000 worms) and significantly least cocoon yield by number per 10,000 worms was recorded in breed CSR<sub>4</sub> (1,837).

Chandrasekharan and Nataraju (1998) reported that among bivoltine races NB<sub>4</sub>D<sub>2</sub> was the least susceptible to muscardine infection and that NB<sub>18</sub> and NB<sub>7</sub> breeds the most susceptible. Venkataramana Reddy (1978) also recorded higher cocooning per cent in KA, followed by NB<sub>2</sub> and NB<sub>18</sub>, at different dilutions of fungal spores. In the present study B1 and B4 could yield more cocoons per 10,000 worms under fungal infection.

When eight races of silkworm were inoculated with nine conidial concentrations (10<sup>1</sup> - 10<sup>9</sup> spores/ml) of *B. bassiana*, no cocoons were spun at the two highest concentrations, as compared with 96.67 to 100 per cent cocoons formed in the batches with no treatment and 48 to 78 per cent in the case of the batches with the lowest concentration (Raghavaiah and Jayaramaiah, 1990). The breeds B4 (9,066.7 cocoons/10,000 silkworms) and B1 (8,033.33 cocoons/10,000 silkworms) recorded highest cocoon yield by number under *B. bassiana* inoculation (Sahana *et al.*, 2021). Keerthana *et al.* (2020) recorded significantly highest number of cocoons in breed B4 (620/1,000 worms) followed by B1 (500/1,000 worms) under *B. bassiana* inoculation. Results of the



TABLE 3  
Effect of *B. bassiana* infection on cocoon parameters of thermo tolerant bivoltine silkworm breeds

Breeds	cocoon weight (g)		Shell weight (g)		Pupal weight (g)		Shell ratio (%)	
	Control	<i>B. bassiana</i> infected	Control	<i>B. bassiana</i> infected	Control	<i>B. bassiana</i> infected	Control	<i>B. bassiana</i> infected
B1	1.94 <sup>a</sup>	1.49 <sup>a</sup> (-38.65)	0.45 <sup>a</sup>	0.31 <sup>a</sup> (-31.11)	1.48 <sup>a</sup>	1.16 <sup>a</sup> (-21.62)	23.78 <sup>a</sup>	20.97 <sup>a</sup> (-11.81)
B4	1.91 <sup>a</sup>	1.27 <sup>b</sup> (-33.50)	0.47 <sup>a</sup>	0.28 <sup>b</sup> (-40.42)	1.41 <sup>a</sup>	0.97 <sup>b</sup> (-31.20)	24.97 <sup>a</sup>	22.64 <sup>a</sup> (-9.33)
B8	1.36 <sup>c</sup>	1.15 <sup>c</sup> (-15.44)	0.24 <sup>c</sup>	0.20 <sup>c</sup> (-16.66)	1.09 <sup>b</sup>	0.90 <sup>b</sup> (-17.43)	17.35 <sup>b</sup>	17.83 <sup>b</sup> (+2.76)
CSR4	1.69 <sup>b</sup>	1.10 <sup>c</sup> (-34.91)	0.38 <sup>b</sup>	0.22 <sup>c</sup> (-42.10)	1.31 <sup>a</sup>	0.71 <sup>c</sup> (-45.80)	24.24 <sup>a</sup>	22.01 <sup>a</sup> (-9.19)
F TEST	*	**	**	**	**	**	**	**
SEm±	0.05	0.02	0.009	0.004	0.05	0.03	1.23	0.78
CD at 5%	0.16	0.07	0.03	0.014	0.14	0.108	3.77	2.43
CV (%)	6.05	3.68	5.18	3.66	7.06	7.49	10.85	7.56

Note: Positive and negative figures in the parenthesis indicate per cent increase (+) or decrease (-) over control, respectively;  
\*-Significant @ 5%; \*\* - Significant @1%

current study are also in conformity with the earlier findings of Jayashree *et al.* (2020) under *Beauveria bassiana* infection.

#### Cocoon Yield by Weight (g / 10,000 Worms)

Effect of *B. bassiana* infection on cocoon yield by weight showed significant difference and the highest value for cocoon yield by weight was observed in breed B1 (14731.75 g/10,000 worms), followed by B4 (12,551.00 g/10000 worms) and B8 (11,518.00 g/1000 worms) and significantly lowest value was noticed in the breed CSR<sub>4</sub> (9681.80 g/10000 worms) (Table 3).

When four thermo tolerant bivoltine breeds *viz.*, B1, B4, B6, B8 and their hybrids *viz.*, B1× B4, B1× B6, B1× B8, B4× B1, B4× B6, B4× B8, B6× B1, B6× B4, B6× B8, B8× B1, B8× B4 and B8× B6, were inoculated with *B. bassiana*, significantly highest cocoon yield by weight was recorded in B1 (867.00 g/1000 worms) among parents and in B1× B8 (960.47 g/1000 worms) among hybrids (Jayashree,

2019). The thermotolerant bivoltine silkworm breeds B4, B6 and B8 recorded highest cocoon yield by weight when inoculated with *B. bassiana* spores, which might be due to their ability to spin good cocoons even under infected condition (Keerthana *et al.*, 2019). According to Sreejith Vakayil (2019), among the eight thermotolerant bivoltine breeds (B1 to B8), significantly highest cocoon yield by weight was recorded in B4 (1007.50 g / 1000 worms), followed by B1 (971.00 g / 1000 worms) / 1000 worms) under *B. bassiana* infection, which is in alternation with the present study.

#### Cocoon Weight (g)

Significant differences were observed among the thermotolerant bivoltine silkworm breeds for single cocoon weight in *B. bassiana* inoculated and control batches. In *B. bassiana* inoculated batches, maximum cocoon weight of 1.49 g was recorded in B1 followed by B4 (1.27 g) and B8 (1.15 g) breeds and it was minimum in CSR4 (1.10 g) breed. The decrease in cocoon weight over control by 15.44 per cent, 38.65

per cent, 34.91 per cent and 33.50 per cent were observed in B8, B1, CSR4 and B4 respectively (Table 2). Suggesting B8 to be productive under infection. Raghavaiah and Jayaramaiah, (1990) reported that NB7 formed cocoons with maximum weight (1.027 g) compared to NB<sub>18</sub> (0.940 g) when infected with muscardine. Also, Rajitha and Savithri, (2014) reported significant reduction in cocoon weight in PM×CSR<sub>2</sub> when infected with *B. bassiana* spore on first day of fifth instar which was similar to the present study.

### Shell Weight (g)

The breeds showed significantly different cocoon shell weights when they were subjected to *B. bassiana* inoculation. In *B. bassiana* inoculated batch, significantly highest cocoon shell weight was recorded in B1 (0.31 g) followed by B4 (0.28 g) and CSR4 breeds (0.22 g). The breed B8 recorded the lowest cocoon shell weight of 0.20 g. (Table 3). In earlier studies NB7 silkworm spun cocoons with maximum shell weight compared to NB4D2, KA and NB18 bivoltine silkworm breeds when they were infected with different doses of *B. bassiana* spore during fifth instar (Venkataramana Reddy, 1978). Topical application of *B. bassiana* spores to ten thermotolerant silkworm breeds recorded the highest cocoon shell weight in B4 (0.24 g) and B1 (0.19 g) breeds (Keerthana *et al.*, 2020) these findings are corroborating with that observed in the present study.

### Pupal Weight (g)

Pupal weight was significantly affected among the *B. bassiana* inoculated thermotolerant bivoltine silkworm breeds. In *B. bassiana* treated batch, B1 breed exhibited significantly maximum pupal weight of 1.16 g, followed by B4 (0.97 g) and B8 (0.90 g) breeds. Pupal weight was significantly lowest in CSR<sub>4</sub> (0.71 g) breed. The B4 breed recorded highest (31.20%) reduction in pupal weight over control and it was lowest in B8 breed (17.43%) (Table 3). Reduction in pupal weight in cross breed (PM x CSR<sub>2</sub>) silkworm was observed when treated with sub-lethal concentration of *B. bassiana* conidial suspension (Rajitha and Savithri, 2014). The present

findings were also supported by results of Keerthana *et al.* (2020), wherein B1 breed recorded maximum pupal weight of 0.92 g followed by B4, B6, B7 and B8 (0.87 g each) under muscardine infection.

### Shell Ratio (%)

Cocoon shell ratio was significantly affected due to *B. bassiana* inoculation among the thermotolerant silkworm breeds. In *B. bassiana* inoculated batch, B4 breed exhibited significantly highest cocoon shell percentage of 22.64, followed by CSR4 and B1 breeds (22.01 and 20.97%, respectively). Silkworm breed B8 exhibited significantly lowest cocoon shell ratio of 17.83 per cent. Cocoon shell ratio was highly affected in B1 breed as it recorded 11.81 per cent decrease over control while, it increased in B8 (2.76%) breed and least reduction in CSR4 breed (9.19%) (Table 3). In earlier studies, NB<sub>4</sub>D<sub>2</sub> produced highest shell ratio compared to NB<sub>7</sub>, KA and NB<sub>18</sub> (Venkataramana Reddy, 1978) and cross breed, PM x CSR<sub>2</sub> recorded lesser cocoon shell ratio of 12.80 per cent under *B. bassiana* infection compared to control (16.43%) (Seema *et al.*, 2019).

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## Arranging a Set of Accessions with Whole Genome Sequence Amenable for GWAS Studies in Rice

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### ABSTRACT

Water scarcity due to climate change is emerging as a major threat for agriculture in recent years. Growing rice, a water intensive crop in the puddled condition is becoming a major challenge as it consumes around 50 per cent of available fresh water for agriculture. It's critical to improve yield potential of rice under water limited conditions to meet the increasing demands. Though, adapting changes in agronomic practices like growing rice under semi-aerobic conditions could reduce water usage, a concomitant yield loss has occurred. Developing a rice cultivar with minimal water use with increased yield or without reducing the actual yield potential is a great challenge. Addressing this, by evaluating genetic diversity at the DNA/molecular level has become an interested approach with the availability of diversified germplasm resources and genetic information across species generated through high-throughput technologies and innovations. Evaluating the accessions with large phenotypic and genotypic diversity and looking for a desirable qualitative trait through techniques *viz.*, GWAS would lead to a superior cultivar with improved yield potential. Considering Water Use efficiency (WUE) as a qualitative physiological trait and dissecting its subcomponent traits and looking for the variations present in the genes governing the latter would harness the advantages of WUE in crop improvement programs. With this background, a set of 853 diversified accessions with Whole Genome Sequence (WGS) was procured. Further, a subset was assembled based on various criteria like geographical, physiological and molecular diversity that will be amenable for GWAS.

**Keywords :** Water use efficiency, GWAS, Water scarcity, Whole genome sequence

**R**ICE, the most important staple cereal in India is an extremely water intensive crop consuming more than 50 per cent of all the fresh water used in agriculture. With the erratic rainfall leading to decreased water availability growing rice under puddled conditions has emerged as the major constraint to achieve potential productivity. So, it's highly important to focus on water conservation strategy without compromising on the yield, while improving the yield of local varieties (Karthika and Shanker, 2022). A few water saving agronomic practices have been developed, among them, the semi-irrigated aerobic cultivation is known to save up to 40 per cent of irrigation water, with a significant

yield loss of up to 50 per cent (Bouman *et al.*, 2005). The yield reduction could be associated with low spikelet fertility under water limited condition. To overcome this, breeding for rice under water limited conditions is crucial. Genetic enhancement of crops to evolve superior cultivars through improving physiological traits among several drought adaptive traits have been identified and introduction of double haploids technique (Chaitanya and Raju, 2022) are being used for crop improvement (Reynolds and Tuberosa, 2008 and Araus *et al.*, 2008).

Evaluation of genetic diversity is considered as a basic platform for crop improvement. Plant scientists

always evaluated the populations over years and locations to understand genetic diversity present in the species of their interest. Insights on useful trait inheritability, environmental influence, value of different parents for breeding and obtaining desirable offsprings were collectively performed by using replication and sophisticated experiments. But, the introduction of evaluation of genetic diversity at the DNA level has become an interested approach.

Datasets associated with genotyping and phenotyping have been generated to address both basic and applied questions. This interests have been shown in the nature and origin of mutations and functional significance of the genes to obtain qualitative and quantitative traits. Tremendous improvement and innovations in the genomics technology over the last 20 years, through multidimensional approaches collaborating biologists, bioinformaticians and chemists have been driven with the efficient support of the available diversified germplasm resources and genetic information across species through high-throughput genotyping and next generation sequencing (NGS) (Tung *et al.*, 2010 and Elshire *et al.*, 2011).

Whole-genome sequence, emerging as an unifying tool in biology has lead to development of diversified panels and large mapping populations in many crop species to facilitate trait dissection and gene discovery. With the availability of huge genetic information, adapting Genome Wide Association Studies (GWAS) technique would lead to the discovery of QTLs associated with the phenotypic trait of interest.

Among various physiological traits contributing to yield, water use efficiency (WUE) and water use are considered as important traits under water limited conditions. Physiologically WUE is defined as  $\mu\text{mol CO}_2$  fixed per mole of water transpired or is the ratio of biomass to water transpired. Improving crops for WUE most often was not associated with higher yields due to the occurrence of trade-off between photosynthesis and transpiration. However, it is evident that at a given water use, plants with higher WUE would have higher growth rates,

which can be predominantly due to chloroplast carbon assimilatory capacity. Therefore, developing a greater understanding of the various chloroplast and photosynthetic mechanisms governing variability in WUE has high relevance in crop improvement programs.

To harness the advantages of WUE, it is critical to dissect this complex trait into its subcomponents. With this background, a program was conceived to identify the genetic variability in WUE and its sub-component traits using the accurate large-scale phenotyping facility. To accomplish this study, a set of accessions with diversity in geographical origin, genotypic and phenotypic variability was studied with a set of 150 germplasm accessions with whole genome sequence is assembled considering various criteria like population group, days to flowering and allelic variations found in the genes for future phenotyping experiments that will be amenable for GWAS.

## MATERIAL AND METHODS

### Seed Material and Growth Condition

A diversified panel of 853 germplasm accessions with whole genome sequence from 3K Rice genome panel were procured from the IRRI Centre located at ICRISAT, Hyderabad. The selected accessions were grown in the field at ZARS Mandya, during *kharif* 2019 under aerobic condition. Around 30 plants were maintained for each accession and the seeds were dibbled at a depth of 3-4 cm while maintaining 20×10 cm spacing between rows and plants respectively. The recommended agronomic practices were followed. The accessions were grown till the crop reaches its physiological maturity and seeds were collected for the further experiments.

### Selection of a Diversified Subset

The passport data containing various informations on the accessions like, country of origin, sub population group, days to 50 per cent flowering etc., are available at the IRRI website (SNP Seek). These data were used to select a subset of accessions belonging to subpopulation group of Indica. Further, observations on days to 50 per cent flowering recorded at ZARS

Mandya and the data available with passport information were compared to narrow down the accessions with 85 to 95 days to 50 per cent flowering.

### Selection of Genes

Water Use efficiency is contributed by various sub-component traits *viz.*, CO<sub>2</sub> diffusion, carboxylation, photochemical processes, epidermal patterning, stomatal mechanisms, VPD responses, etc. These subcomponent traits are governed by various genes and transcription factors, so a through literature search was done in selecting a set of genes governing different subcomponent traits of WUE and based on the importance and functions.

### Sequencing Data Analysis

Single nucleotide polymorphism data for the set of selected accessions with reference to Nipponbare are available at IRRI website (SNP Seek) and were downloaded for the experimental purpose. Around 2178 SNP markers were detected for 217 accessions and these SNPs were used to perform population structure using the STRUCTURE HARVESTER software with maximum adhoc measure ( $\Delta K$ ) of five. Further Cluster analysis was performed for 217 accessions based on random 2198 SNP markers by Neighbor-joining method using DARwin.

## RESULTS AND DISCUSSION

The main aim of this study was to select a subset from the 853 lines obtained from the IRRI center located at

ICRISAT, Hyderabad. Hence, considering phenotypic and molecular diversity was crucial.

### Selection of a Set of Accessions Based on Subpopulation Group

The 853 accessions obtained from the IRRI center located at ICRISAT, Hyderabad, represented 48 countries Fig. 1, indicating a diversification in their origin and the accessions belong to 12 subpopulation groups (Table 1). Since the goal of our study was to improve crop productivity in Indian conditions, only Indica subpopulation group was selected and the 853 lines were narrowed down to 522 lines.

TABLE 1  
Representing 853 lines from different sub population groups

Subpopulation	No of genotypes
Admixture	27
Aromatic	27
Australian	78
Indica 1 A	41
Indica 1 B	41
Indica 2	176
Indica 3	87
Indicax	178
Japonicax	15
Subtropical	70
Temperate	10
Tropical	103

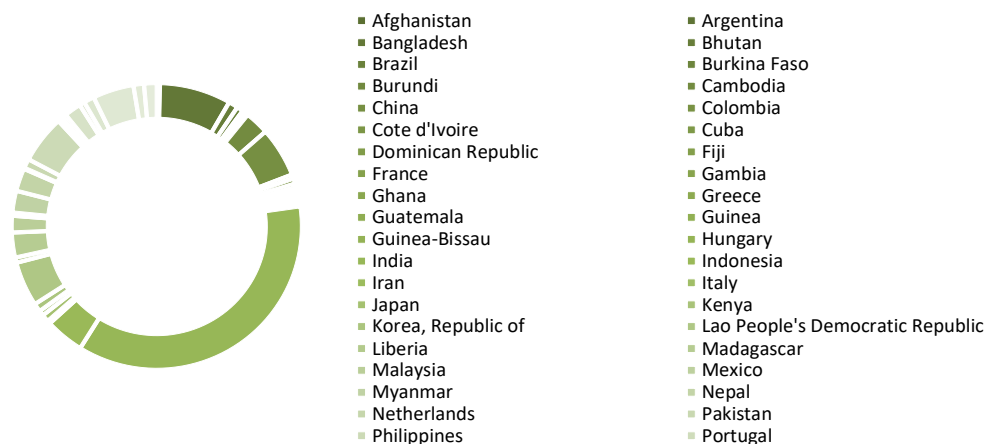


Fig. 1 : Collection of 853 lines from different geographical area



Fig. 2 : Phenotyping of 853 rice accessions at Zonal Agricultural Research Station, V C Farm, Mandya

### Narrowing Down of the Accessions Based on Days to Flowering

Every germplasm will have passport data on various traits including days to flowering. We observed a large variation in days to flowering between 50 and 184 days after sowing from a passport data (<https://snp-seek.irri.org/>). To verify these data, the entire set of 853 accessions was grown in ZARS, VC Farm Mandya during *kharif* 2019 (Fig. 2). Days to Flowering recorded at Mandya did not show any association with passport data while 70 accessions remained unflowered due to photosensitivity (Fig. 3 and Table 2). This indicates the possibility of variations in photosensitivity of germplasm collected

TABLE 2

Comparison of flowering duration between passport data and measured data

Flowering Duration (days)	Passport data	<i>Kharif</i> 2019
51-60	5	3
61-70	47	0
71-80	110	130
81-90	177	125
91-100	142	246
101-110	134	138
111-120	106	78
121-130	68	12
131-140	31	51
141-150	13	-
151-160	11	-
161-170	8	-
Photosensitive	-	70

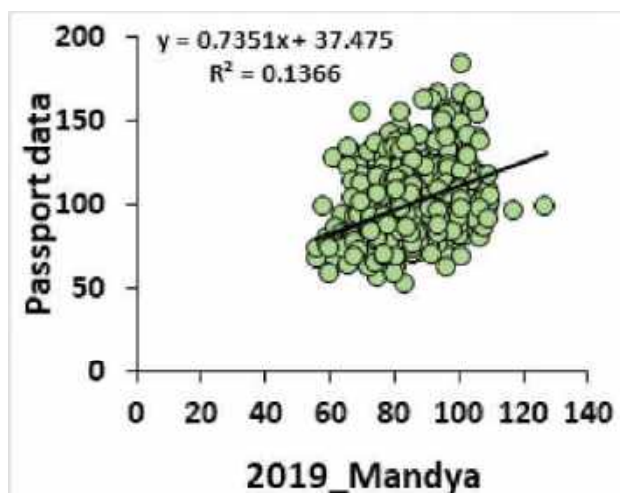


Fig. 3 : Correlation for days to fifty percent flowering taken from passport data and recorded data from VC farm Mandya

from various locations. Flowering time is controlled by many genes, which are expressed or suppressed in close interaction with environmental factors such as day length and temperature (Tsuji *et al.*, 2008). Since the environmental conditions in Phillipines and India are comparatively different we observed a larger variation in days to flowering ( $R^2 = 0.13$ ). Further comparing both the data, around 217 accessions were selected having a range between 85 and 95 days to flowering. Development of early-flowering or photoperiod-insensitive cultivars has been an important objective of rice breeding (Tsuji *et al.*, 2008). Hence the selection was made to meet this objective.

### Dissection of Water use Efficiency in to Subcomponent traits

With the goal of improving crop productivity through WUE, dissecting the subcomponents of

WUE plays a major role. Considering this, around 35 putative genes governing various subcomponents traits of WUE *viz.*, CO<sub>2</sub> diffusion, carboxylation, photochemical processes, epidermal patterning, stomatal mechanisms, VPD responses *etc.* were identified through literature search and genetic diversity analysis has been done to know the variations in sequences of the candidate genes leading to functional differences. Recently (Sheshshayee *et al.*, 2013) provided some experimental evidences, that clearly establish the relevance of WUE as an important physiological trait that can be used for crop improvement. It was further proved that introgression of WUE with other traits such as total water use (roots) and improved acquired tolerance mechanism significantly enhanced growth rate under both well-watered and water limited condition (Raju *et al.*, 2014 and Sheshshayee *et al.*, 2018). Furthermore, after identifying a robust marker associated with WUE and root traits (Raju *et al.*, 2016), a marker assisted back-cross breeding strategy was adopted to introgress WUE and water use characters on to the background of a late variety IR64. The trait introgressed lines on an average resulted in a 23 per cent increased yield under water limited conditions while saving more than 50 per cent of irrigation water (Dharmappa *et al.*, 2019) These clearly emphasize the possibility of improving productivity through an improvement in WUE.

### Structure Analysis

A total of 2591 SNPs present among the 217 accessions were retrieved. After filtering the missing data, a total of 2278 SNPs (Table 3) were selected

TABLE 3  
Number of SNPs on each chromosome used to run cluster analysis

Chromosome	Number of SNPs
CHR 1	251
CHR 2	244
CHR 3	110
CHR 4	403
CHR 5	132
CHR 6	244
CHR 7	158
CHR 8	108
CHR 9	153
CHR 10	113
CHR 11	152
CHR 12	130

for further analysis. Population structure of 217 accessions was analyzed using STRUCTURE with different K values ranging between 1 and 10. STRUCTURE harvester showed the maximum adhoc measure ( $\Delta K$ ) at five (Fig. 4). The optimum K value was fixed at so as to divide the 217 accessions into five sub-populations (Fig. 5).

Neighbor joining analysis using DARwin clustered the accessions also into five clusters, with two distinctly distant clusters (C1 and C2) as illustrated in Fig. 6. A core set of germplasm was constructed using heuristic search available with Power Core and a core collection containing 150 rice accessions was constituted. The selected accessions represented 29 countries of origin (Fig. 6) and five subpopulation

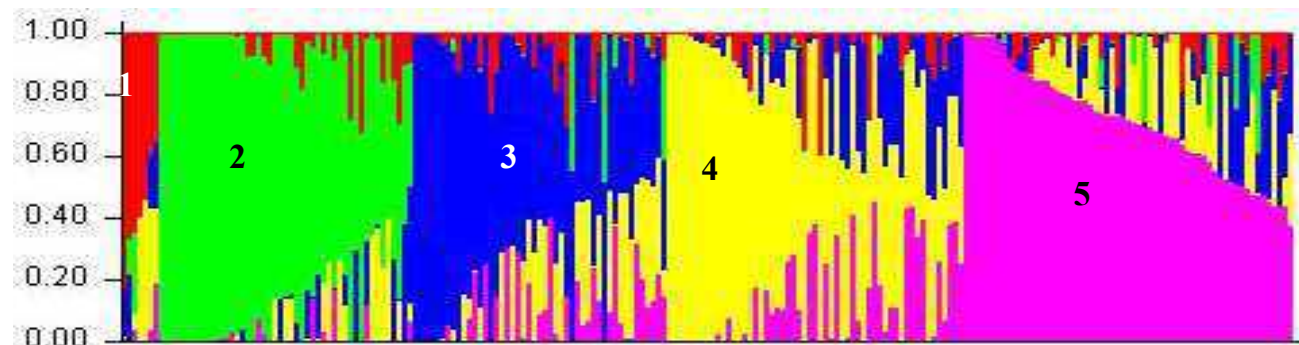


Fig 4. Population structure of 217 accessions



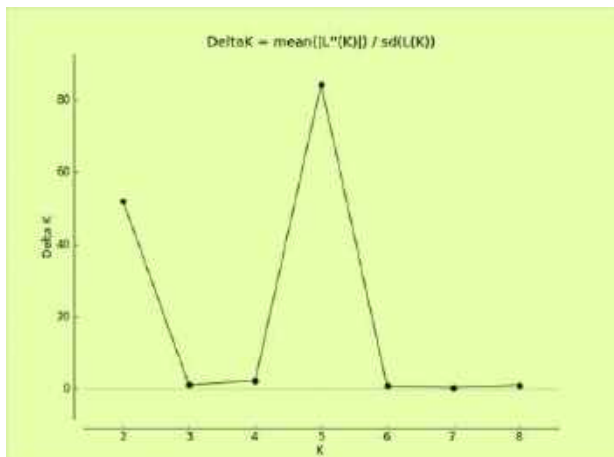


Fig. 5 : Maximum adhoc measure ( $\Delta K$ ) of two observed from STRUCTURE harvester

TABLE 4  
150 lines representing different subpopulation groups

Subpopulation	No of genotypes
Indica1A	11
Indica1B	6
Indica2	55
Indica3	16
Indicax	62

group (Table 4) indicating large geographical diversity maintainance and these accessions will be used for all phenotyping and GWAS activities. Similarly, Nachimuthu *et al.* (2015) assessed the molecular and genetic diversity and relatedness by evaluating the set of 192 diverse rice germplasm lines with 61 genome wide SSR markers leading to identification of 205 alleles revealing two sub groups.

So this indicates that understanding the population structure and assessing the kinship relatedness between superior alleles and traits which is necessary for successful association mapping program.

### Haplotype Analysis

The sequences of the selected genes were downloaded from the IRRI database and aligned to screen for haplotype diversity. Haplotype analysis revealed the existence of large sequence variations in 35 candidate genes (2-15 haplotypes) except Rca and BP-73 which had no polymorphism (Table 5). Diversity analysis and population structure analysis were done for the selected 217 lines using nucleotide sequence polymorphisms prevailing in the 35 genes. The existing SNPs and InDels among the 35 genes were retrieved from the SNPseek database (<https://snpseek.irri.org/>) and used. Haplotype assembly is one

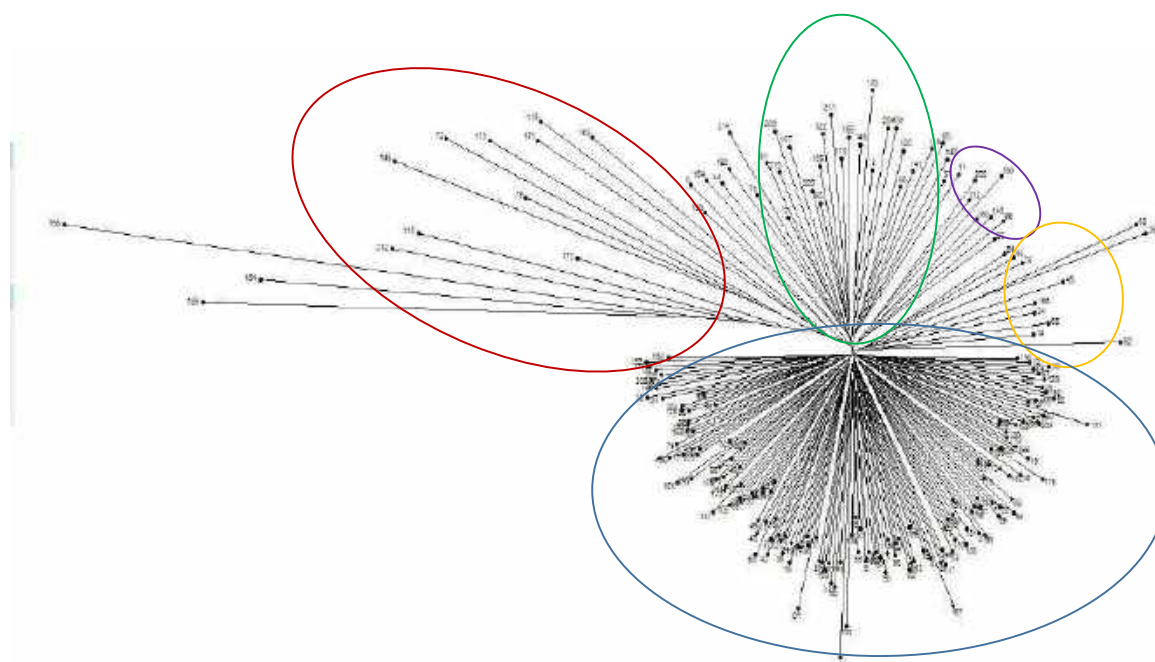


Fig. 6 : Cluster analysis of 217 accessions based on 2278 SNP markers for 35 candidate genes using DARwin

TABLE 5  
Haplotype diversity of selected candidate genes 3K  
panel rice accessions.

Gene	Locus ID	Haplotypes
SDD1	Os03g0143100	12
ERECTA	Os02g0777400	15
SLAC1	Os04g0574700	15
GORK	Os06g0250600	2
OST2	Os03g0689300	4
rbcL	Os12g0277500	3
RbcS	Os12g0274700	15
<b>Rca</b>	Os11g0707000	-
PGK	Os06g0668200	5
GPS	Os04g0615000	15
OsCKX2	Os01g0197700	15
<b>BP-73</b>	Os03g0183100	-
SAP16	Os07g0569700	15
HAP3	Os01g0834400	15
SPS	Os01g0919400	9
SBP	Os01g0916400	15
TMM	Os01g0623000	15
FAMA	Os05g0586300	15
COP1	Os02g0771100	2
DST	Os10g0456800	8
EPFL9	Os01g0914400	12
SAMDC	Os02g0611200	2
P5CS	Os01g0848200	2
NAC	Os01g0191300	3
AP37	Os01g0797600	15
CIPK	Os03g0319400	15
PIP	Os02g0823100	15
SnRK2	Os03g0250000	15
OsCDPK	Os03g0128700	15
SBP	OSNPB_040234600	15
TPI	OSNPB_010147900	2
RPI	OSNPB_030781400	15
FBA	OSNPB_060664200	2
RPE	OSNPB_030169100	15
SPS	OSNPB_020184400	2

of the promising approaches used in breeding program for crop improvement. In recent years, haplo-pheno analysis is extensively used for identification of superior haplotypes. In 3K rice genome panel, haplotypes of 21 genes governing grain yield and quality has been identified (Abbai *et al.*, 2019). Moreover, haplotypes of deep water adaptation in rice and for direct seeded rice (DSR) have been identified (Kuroha *et al.*, 2018 and Chen *et al.*, 2019). Hence in this study more number of haplotypes is a indirect indicator of vast genetic and phenotypic variability in the selected population.

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## Studies on Effect of Different Levels of Fertigation on Growth and Yield of Ginger Cultivars in Eastern Dry Zone of Karnataka

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### ABSTRACT

Field experiment was carried out at the Department of Horticulture, UAS, Bangalore, in well-drained red sandy loam soils of the experimental block with a pH of 6.2. The soil was low in organic carbon and available nitrogen, whereas available phosphorus and potassium were in medium range. The experiment consisted of two ginger cultivars (Rio-de-Janeiro and Himachal) and 11 fertigation levels which were replicated thrice in the RCBD. Among the eleven fertigation treatments tried on two ginger cultivars, significant variation was observed for growth and yield parameters. Maximum plant height at 30, 60, 90, 120, 150, 180 and 210 DAS was recorded with 200 per cent RDF (200:100:100 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg /ha) through fertigation + FYM 30t/ha, Neem cake 2t/ha in both the varieties. Maximum leaf area (1387.52 cm<sup>2</sup> and 2708.02 cm<sup>2</sup>) and leaf area index (1.85 and 3.61) at harvest were also recorded in the same treatment for Rio-de- Janeiro and Himachal, respectively. Highest number of primary fingers, secondary fingers per clump, maximum fresh weight and length of mother rhizome, dry matter production at harvest were recorded with the application of 200 per cent RDF + neem cake @ 2 t ha<sup>-1</sup> which was on par with the 150 per cent RDF along with 30 tons of FYM and 2t of neem cake in both the varieties. Highest growth and yield parameters were recorded in the Himachal variety as compared to Rio-de-Janeiro with respect to the fertigation levels.

Keywords : Ginger, Fertigation, RDF, Neem cake, Bio fertilizers

GINGER (*Zingiber officinale*) belonging to family Zingiberaceae is the major and widely used spice throughout the world. It occupies a prime position among the spice crops in India. India is a major producer and grower of ginger. It is grown in an area of 1.72 lakh ha accounting to an annual production of 18.43 lakh tonnes with an average productivity of 10.71 t/ha. In India it is grown to a larger extent in Madhya Pradesh (25,402 ha), Karnataka (21,683 ha) and Assam (19,351 ha) with a production of 410950 t, 278000 t and 183160 t, respectively (Anonyomous., 2019). Ginger is extensively prized for its aroma flavour, pungency and medicinal properties since ancient times.

Among the major spices grown in the country, ginger occupied an important place, as it is the major GDP earner. Dry ginger, green ginger, oleoresin and essential oils of ginger are the other important value added products and export of these products is increasing year after year. The refreshing aroma and the pungent taste make ginger an essential in the best ingredient of food and several processed products.

Ginger is a long duration crop and needs a balanced and sustained supply of nutrients for higher fresh rhizome yield with a better quality, which can be supplied by both inorganic sources and organic sources. Imbalanced nutrient supply is one of the

major constraints for reduced yield coupled with poor quality. Ginger is a nutrient exhaustive crop and application of organic manures, fertilizers and bio fertilizers are absolutely essential. Ginger rhizomes are mainly N and K exhausting, intermediary in P and Mg removal and the least in Ca removal (Nagarajan and Pillai, 1979). Verma *et al.* (2019) reported an accumulation of macronutrients in the decreasing order *viz.*, N, K, Ca, Mg, S and P similarly micronutrients in the order of Fe, Mn, Zn, B, Cu. However, nutrients uptake varies considerably with soil type, varieties or hybrids, climatic conditions, nutrient levels in the soils.

Fertigation is a method of fertilizer application in which fertilizers are supplied along with water by drip system. In this system fertilizer solution is distributed evenly in water and directly supplied to the active root zone through drippers which results in higher nutrient use efficiency. As water and fertilizers are supplied evenly to the ginger crop there is possibility of getting 25-50 per cent higher yield (TNAU Agritech portal, 2016). Fertilizer use efficiency through fertigation varied between 80-90 per cent, which helps to save on an average of 25 per cent of applied nutrients.

Till today farmers are practicing conventional method of nutrients applications in ginger cultivation getting average yield coupled with poor quality of rhizomes. This warrants the use of alternative nutrients application method. Nutrients application through fertigation offers a viable option as this enhances the Nutrient use efficiency in turn increased yield. This present study focus on the standardization of fertigation level for increasing the nutrient use efficiency increased yield of ginger cultivars.

#### MATERIAL AND METHODS

Field experiments were carried out to evaluate the performance of ginger cultivars and to study the effect of different fertigation levels on growth and yield of ginger cultivars by increasing the nutrient use efficiency in Southern dry zone of Karnataka during the *kharif* season of 2021 at the

Department of Horticulture, GKVK, University of Agricultural Sciences, Bangalore.

The experiment was carried out on the red sandy loam soil in the Horticulture experimental block, Gandhi Krishi Vignana Kendra, University of Agricultural Sciences, Bangalore. It is situated in the Southern dry zone of Karnataka with the mean annual rainfall of 800-900mm which is fairly distributed from May to November. The soils of the experimental block are well drained red sandy loam, with a neutral pH of 6.4. The soil is low in organic carbon (0.35%), low in available nitrogen (201.14 kg ha<sup>-1</sup>), medium in available phosphorus (34.0 kg ha<sup>-1</sup>) and medium in available potassium (152 kg ha<sup>-1</sup>).

The field experiment was laid out in a randomized complete block design with three replications. Eleven fertigation levels were tried on two ginger cultivars namely Rio-de-Janeiro and Himachal. Ginger cultivars were grown in the net plot size of 10.0 m × 1.0 m with a spacing of 45 cm between the rows and 30 cm between the plants. The treatment details are described below.

Treatment	Details
T <sub>1</sub>	RDF (100:50:50 N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg /ha normal fertilizers) + FYM 30 t/ha, Neem cake 2t/ha
T <sub>2</sub>	200 % RDF (200:100:100 N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha
T <sub>3</sub>	150 % RDF (150:75:75 N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg/ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha
T <sub>4</sub>	100 % RDF (100:50:50 N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg/ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha
T <sub>5</sub>	75 % RDF (75:37.5:37.5 N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg/ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha
T <sub>6</sub>	50 % RDF (50:25:25 N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg/ha) NF soil application + 50% Fertigation (50:25:25 NPK kg/ha) WSP + FYM 30 t/ha, Neem cake 2t/ha
T <sub>7</sub>	100 % RDF (100:50:50 N: P <sub>2</sub> O <sub>5</sub> : K <sub>2</sub> O kg/ha) NF + Azotobacter + PSB + AMF + KMB + FYM 30t/ha, Neem cake 2t/ha

- T<sub>8</sub> 75 % RDF (75:37.5:37.5 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha WSF) + Azotobacter + PSB + AMF + KMB + FYM 30t/ha, Neem cake 2t/ha
- T<sub>9</sub> 50 % RDF (50:25:25 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha WSF) + Azotobacter + PSB + AMF + KMB + FYM 30 t/ha, Neem cake 2t/ha
- T<sub>10</sub> 50 % RDF (50:25:25 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha WSF) + Ginger Special 25 % Foliar spray at 60, 90, 120 DAP + Azotobacter + PSB + AMF + KMB + FYM 30 t/ha, Neem cake 2t/ha
- T<sub>11</sub> 100% RDF (100:50:50 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha WSF) + Azotobacter + PSB + AMF + KMB + FYM + Neem cake

The land was prepared and brought to a fine tilth by ploughing followed by harrowing. Ridges and furrows were formed after leveling the plots. The seed material was procured from ZARS, Chamaraj nagara. Healthy and disease free, uniform sized fingers of cultivars with well-developed buds were selected for planting. The fingers were planted in the furrows by adopting a proper spacing between rows and plants. After the preparation of land and plots, farm yard manure was applied at 30 tonnes per hectare as a basal dose. The fertilizers in the form of urea (46% N), single super phosphate (16% P<sub>2</sub>O<sub>5</sub>) and muriate of potash (60% K<sub>2</sub>O) were applied. Nitrogenous fertilizer was applied in two splits, phosphatic and potassic fertilizers were applied as a basal application. The fertilizers were applied in the rows and mixed in the soil. Each plot, the required quantity of fertilizers was calculated as per the treatments. Weed management and plant protection measures were taken whenever is required to get the optimum growth and yield.

The crop was harvested as per maturity standards as indicated by the drying up of the leaves, pseudostem and drooping of the plants. One day before digging of the rhizomes, light irrigation was provided. Rhizomes were removed by digging and were separated.

Observations on vegetative growth parameters were recorded from five randomly selected and marked plants from each treatment at 30, 60, 90, 120, 150,

180 and 210 days after planting (DAP). The leaf area (Y) was calculated by an equation  $y = 0.6995 \ln - 0.768$ . This is an accurate method for determining leaf area in ginger and expressed as leaf area per plant in cm<sup>2</sup>. The dry weight (g) was recorded by drying the fresh plants parts at 70 °C till consistent weights were obtained. The average value was expressed as dry matter per plants in grams. Fresh rhizomes from each plot were weighed and recorded as kg per plot. The gross plot yield was computed and expressed as fresh yield in tonnes per hectare. The number of primary fingers arising from the mother rhizomes (plant material) in each of the labeled plants was counted and mean was worked out. Length of primary fingers was measured by using a non-stretchable string and expressed in centimeter. The number of secondary fingers arising from the primary fingers was counted in all the labeled plants and average was worked out. The length of secondary fingers was measured by using a non-stretchable string from the selected five fingers and expressed in centimeter.

The experimental data was subjected to standard analysis of FRCBD. The level of significance @ 5 per cent employed in F test and critical differences (CD) were calculated where ever F test was significant.

## RESULTS AND DISCUSSION

### Plant Height (cm)

The data pertaining to effect of different fertigation levels on plant height recorded at different stages of crop growth (30, 60, 90, 120, 150, 180 and 210 days) for different cultivars during 2021 are presented in the Table 1.

Plant height differed significantly among the cultivars at all the stages of crop growth and also among the fertigation levels.

Application of 200 per cent RDF (200:100:100 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha) through fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>2</sub>) to Himachal cultivar recorded highest plant height of 19.40, 19.92, 35.90, 39.12, 39.90 and 41.50 cm at 60, 90, 120, 150, 180 and 210 days after planting, respectively as compared to

TABLE 1  
Effect of fertigation levels on plant height of different cultivars of ginger

Treatments/ Fertigation levels	Plant height (cm) of the ginger varieties																	
	Rio – de – Janeiro (Days after planting)									Himachal (Days after planting)								
	30 DAP	60 DAP	90 DAP	120 DAP	150 DAP	180 DAP	210 DAP	30 DAP	60 DAP	90 DAP	120 DAP	150 DAP	180 DAP	210 DAP				
T <sub>1</sub>	6.48	12.23	16.00	26.13	28.10	29.93	30.03	6.86	14.33	16.95	28.30	31.04	34.00	38.04				
T <sub>2</sub>	6.84	15.16	18.40	33.66	37.03	39.46	40.24	6.96	19.40	19.92	35.90	39.12	39.90	41.50				
T <sub>3</sub>	6.40	14.33	17.66	32.43	36.86	38.90	39.94	6.83	18.40	18.90	34.86	38.90	39.12	40.84				
T <sub>4</sub>	6.48	12.34	17.24	30.26	35.33	37.12	38.25	6.74	14.34	18.36	33.12	37.56	38.73	39.66				
T <sub>5</sub>	6.72	12.29	16.22	31.12	33.34	36.48	38.10	7.10	15.12	17.84	32.22	36.90	37.90	39.12				
T <sub>6</sub>	6.46	13.10	17.21	29.83	34.10	37.50	39.05	6.89	16.03	18.03	31.50	37.04	38.84	39.04				
T <sub>7</sub>	6.54	13.50	17.90	30.34	33.12	36.84	38.45	6.98	16.26	18.56	31.64	37.12	38.56	39.26				
T <sub>8</sub>	6.90	13.26	16.94	31.25	34.20	36.56	39.24	7.04	17.03	18.12	31.08	36.50	37.64	38.93				
T <sub>9</sub>	6.60	12.90	17.10	30.16	33.46	37.22	38.15	7.20	16.10	18.04	32.43	37.15	39.03	39.88				
T <sub>10</sub>	6.46	13.35	17.30	31.45	34.15	37.58	39.26	6.88	16.90	18.12	33.03	37.24	38.87	39.00				
T <sub>11</sub>	6.74	13.45	17.94	30.24	33.90	37.25	39.50	6.95	17.10	18.90	33.12	38.04	39.08	39.34				
F test	*	*	*	*	*	*	*	*	*	*	*	*	*	*				
SEm+	0.072	0.465	0.271	0.283	0.432	0.323	0.346	0.015	0.171	0.016	0.241	0.104	0.301	0.309				
CD @ 5%	0.212	1.374	0.801	0.837	1.276	0.953	1.021	0.046	0.507	0.048	0.712	0.308	0.890	0.912				

Rio-de-Janeiro cultivar (15.16, 18.40, 33.66, 37.03, 39.46 and 40.24 cm at 60, 90, 120, 150, 180 and 210 days after planting, respectively), which is on par with the application of 150 per cent RDF (150:75:75 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha) through fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>3</sub>) with respect to the plant height of both the cultivars. Among the two cultivars tested, Himachal cultivar performed very well for the fertigation levels when compared to Rio-de-Janeiro cultivar. Among the fertigation levels tried on the cultivars, application of 200 per cent RDF (200 : 100 : 100 kg N : P<sub>2</sub>O<sub>5</sub> : K<sub>2</sub>O/ha) through fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>2</sub>) is performed well among the different fertigation levels tested in both the cultivars. The lowest plant height at 60, 90, 120, 150, 180 and 210 days after planting was recorded with the application of RDF (100:50:50 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg /ha normal fertilizers) + FYM 30 t/ha, Neem cake 2t/ha (T<sub>1</sub>) to the Himachal (12.23, 16.00, 26.13, 28.10, 29.93 and 30.03 cm, respectively) and Rio-de-Janeiro cultivar (14.33, 16.95, 28.30, 31.04,

34.00 and 38.04 cm, respectively). The results are in confirmation with the findings of Mathew and Sreekala (2019), who analyzed the nutrients effect on growth as well as yield in transplanted ginger (Soumya *et al.*, 2009).

### Leaf Area Per Plant

It is evident from Table 2 that leaf area per plant differed significantly at 180 days after planting in both the cultivars with different fertigation levels.

Himachal cultivar recorded maximum leaf area (2708.02 cm<sup>2</sup>) as compared to the Rio-de-Janeiro cultivar (1387.52 cm<sup>2</sup>) with 200 per cent RDF (200:100:100 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O/ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>2</sub>) among the different fertigation levels applied to cultivars and minimum leaf area was recorded in Rio-de-Janeiro (731.80 cm<sup>2</sup>) as compared to Himachal cultivar (898.28 cm<sup>2</sup>) with the application of RDF (100:50:50 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg /ha normal fertilizers) + FYM 30 t/ha, Neem cake 2t/ha (T<sub>1</sub>) among the different fertigation levels.

TABLE 2  
Effect of fertigation levels on leaf area and leaf area index (LAI) of different cultivars of ginger at 180 days after planting

Treatments	Leaf Area Index (LAI)			
	Rio – de - Janeiro		Himachal	
	Leaf area (cm <sup>2</sup> )	Leaf area Index	Leaf area (cm <sup>2</sup> )	Leaf area Index
T <sub>1</sub>	731.80	0.97	898.28	1.19
T <sub>2</sub>	1387.52	1.85	2708.02	3.61
T <sub>3</sub>	1277.00	1.70	2286.75	3.05
T <sub>4</sub>	1266.48	1.69	1845.04	2.46
T <sub>5</sub>	1215.81	1.62	1678.86	2.34
T <sub>6</sub>	1015.36	1.35	1642.04	2.19
T <sub>7</sub>	1164.28	1.53	1136.75	1.52
T <sub>8</sub>	1212.46	1.62	1331.72	1.78
T <sub>9</sub>	1185.93	1.58	1407.70	1.88
T <sub>10</sub>	1256.34	1.64	1487.99	1.99
T <sub>11</sub>	1290.25	1.75	1442.36	1.92
F test	*	*	*	*
SEM+	74.09	0.099	204.49	0.273
CD @ 5%	218.27	0.293	603.25	0.807



### Leaf Area Index

The data pertaining to the leaf area index is presented in the Table 2. The leaf area index per plant for different cultivars differed significantly when observed at 180 DAP with the different fertigation levels.

During *kharif* 2021, Himachal cultivar recorded highest leaf area index (3.61) as compared to Rio-de-Janeiro cultivar (1.85) with 200 per cent RDF (200:100:100 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>2</sub>) which was on par with 150 per cent RDF (150:75:75 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>3</sub>) as compared to different fertigation levels. Lowest leaf area index was recorded with RDF (100:50:50 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg /ha normal fertilizers) + FYM 30 t/ha, Neem cake 2t/ha (T<sub>1</sub>) in both the ginger cultivars. Mathew and Sreekala (2019) analyzed the nutrients effect on growth as well as yield in

transplanted ginger. The results of the study indicated that ginger transplants intercropped in coconut garden, with mulching @ 30 t ha<sup>-1</sup> (half at transplanting and remained at 2 MAT) along with 150:100:100 kg NPK ha<sup>-1</sup> apart from basal application of 30 t ha<sup>-1</sup> of farm yard manure resulted in better growth and higher yield.

### Number of Primary Fingers

The production of primary fingers per plant differed significantly among the cultivars and fertigation levels (Table 3). Higher number of primary fingers per plant were produced by Himachal cultivar (4.96) compared to Rio-de-Janeiro cultivar (3.56) with 200 per cent RDF (200:100:100 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>2</sub>) which is on par with 150 per cent RDF (150:75:75 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>3</sub>) as compared to other fertigation levels. Lower number of primary fingers were found in Rio-de-Janeiro cultivar (2.20) with the application of RDF

TABLE 3  
Effect of fertigation levels on number of primary and secondary fingers per clump of different cultivars of ginger

Treatments	Number of primary and secondary fingers per clump			
	Rio – de - Janeiro		Himachal	
	Primary fingers	Secondary fingers	Primary fingers	Secondary fingers
T <sub>1</sub>	2.20	1.76	2.40	2.00
T <sub>2</sub>	3.56	2.93	4.96	3.80
T <sub>3</sub>	3.50	2.90	4.73	3.46
T <sub>4</sub>	3.16	2.93	3.80	3.26
T <sub>5</sub>	2.86	2.36	3.66	2.73
T <sub>6</sub>	3.20	2.60	3.53	2.93
T <sub>7</sub>	3.13	2.60	4.53	2.33
T <sub>8</sub>	3.26	2.00	3.40	3.33
T <sub>9</sub>	2.94	2.40	3.66	2.00
T <sub>10</sub>	3.10	2.58	3.93	3.20
T <sub>11</sub>	3.36	2.53	4.20	2.23
F test	*	*	*	*
SEM+	0.153	0.132	0.289	0.203
CD @ 5%	0.453	0.390	0.853	0.600

only among the different fertigation levels as compared to Himachal cultivar (2.40). Among the cultivars, Himachal cultivar recorded higher number of primary tillers as compared to Rio-de-Janeiro cultivar (Viswanatha *et al.*, 2000).

### Number of Secondary Fingers

The observations on the production of secondary fingers per plant differed significantly among the cultivars and fertigation levels (Table 3).

The number of secondary fingers varied from 2.00 to 3.80 in Himachal and 1.76 to 2.93 in Rio-de-Janeiro. Himachal produced higher number of secondary fingers with the application of fertigation levels, maximum number (3.80) was recorded in 200 per cent RDF (200:100:100 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>2</sub>) and lowest was recorded in RDF (100:50:50 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg /ha normal fertilizers) + FYM 30 t/ha, Neem cake 2t/ha (T<sub>1</sub>) (2.00) which was on par with 50 per cent RDF

(50:25:25 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha WSF) + *Azotobacter* + PSB +AMF + KMB + FYM 30 t/ha, Neem cake 2t/ha (T<sub>9</sub>) (2.00) as compared to Rio-de-Janeiro.

Similarly, cultivar Rio-de-Janeiro recorded maximum number of secondary fingers (2.93) in T<sub>2</sub> and lowest number (1.93) of secondary fingers was produced with RDF (100:50:50 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg /ha normal fertilizers) + FYM 30 t/ha, Neem cake 2t/ha (T<sub>1</sub>).

### Wight of Mother Rhizomes

It was observed that the weight of mother rhizomes per plant differed significantly among the cultivars with different fertigation levels (Table 4).

Himachal cultivar recorded highest weight of mother rhizome per plant (51.33 g) as compared to Rio-de-Janeiro (39.66 g) in the treatment T<sub>2</sub> which was on par with T<sub>3</sub> among the different fertigation levels. Lowest weight of mother rhizomes was found in the Rio-de-Janeiro (22.33 g) and Himachal (26.00 g) with the application of RDF (100:50:50 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg /

TABLE 4

Effect of fertigation levels on fresh weight of mother rhizome, primary and secondary fingers per clump of different cultivars of ginger

Treatments	Fresh weight of mother rhizome, primary and secondary fingers per clump (g)					
	Rio – de - Janeiro			Himachal		
	MR	PF	SF	MR	PF	SF
T <sub>1</sub>	22.33	57.00	21.33	26.00	61.00	24.33
T <sub>2</sub>	39.66	83.00	31.33	51.33	124.66	44.33
T <sub>3</sub>	32.00	71.33	25.33	47.33	119.33	39.66
T <sub>4</sub>	26.00	57.00	21.33	33.33	114.00	29.00
T <sub>5</sub>	29.33	106.66	26.00	35.00	92.00	31.66
T <sub>6</sub>	31.00	105.33	37.33	38.00	101.66	30.00
T <sub>7</sub>	31.66	84.33	27.33	38.66	115.33	34.00
T <sub>8</sub>	29.00	67.00	21.66	41.66	106.33	33.00
T <sub>9</sub>	29.66	66.00	23.66	36.00	113.33	41.33
T <sub>10</sub>	31.25	73.00	24.34	42.33	123.33	37.66
T <sub>11</sub>	32.54	68.00	24.34	43.00	91.00	34.00
F test	*	*	*	*	*	*
SEM+	1.587	5.261	0.542	0.859	1.793	0.682
CD @ 5%	4.682	15.519	1.600	2.533	5.291	2.013

ha normal fertilizers) + FYM 30 t/ha, Neem cake 2t/ha ( $T_1$ ).

### Weight of Primary and Secondary Fingers

The weight of primary fingers per plant varied significantly in both the cultivars with application of different fertigation levels (Table 4). Among the different fertigation levels applied, Himachal cultivar responded very well as compared to Rio-de-Janeiro. Application of 200 per cent RDF (200:100:100 kg N:  $P_2O_5$ :  $K_2O$  /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha ( $T_2$ ) resulted in higher weight of primary fingers in Himachal (124.66 g) and Rio-de-Janeiro (83.00 g). Lowest primary finger weight was recorded in Rio-de-Janeiro (57.00 g) and Himachal (61.00 g) with the application of RDF (100:50:50 N:  $P_2O_5$ :  $K_2O$  kg /ha normal fertilizers) + FYM 30 t/ha, Neem cake 2t/ha ( $T_1$ ).

Similar trend was observed in weight of secondary fingers. Among the cultivars, Himachal recorded higher weight of secondary fingers as compared to

Rio-de-Janeiro. The maximum weight was recorded with the application of 200 per cent RDF (200:100:100 kg N:  $P_2O_5$ :  $K_2O$  /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha ( $T_2$ ) in Himachal (44.33 g) and Rio-de-Janeiro (31.33 g). This treatment was on par with the 150 per cent RDF (150:75:75 N:  $P_2O_5$ :  $K_2O$  kg/ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha ( $T_3$ ) in both the ginger cultivars. The lowest weight of secondary fingers was recorded with 100 per cent RDF ( $T_1$ ) in Rio-de-Janeiro (21.33 g) and Himachal (24.33 g). The similar results were reported by Sharath Pal *et al.* (2014) and Yadgirwar *et al.* (2017).

### Length of Mother Rhizomes

Significant variation in the length of mother rhizome was observed in Himachal and Rio-de-Janeiro cultivars of ginger with different fertigation levels (Table 5). The length of mother rhizome ranged from 2.54 to 4.51cm in Himachal cultivar and 2.42 to 3.65 cm in Rio-de-Janeiro cultivar with different fertigation levels and maximum length of mother

TABLE 5  
Effect of fertigation levels on length of mother rhizome, primary and secondary fingers per clump of different cultivars of ginger

Treatments	Length of mother rhizome, primary and secondary fingers per clump (cm)					
	Rio – de - Janeiro			Himachal		
	MR	PF	SF	MR	PF	SF
$T_1$	2.42	3.23	3.96	2.54	3.34	4.10
$T_2$	3.65	3.67	3.93	4.41	3.75	4.30
$T_3$	3.36	3.60	3.85	4.28	3.46	4.15
$T_4$	2.86	3.35	3.78	3.22	3.75	4.21
$T_5$	2.86	3.16	3.54	3.54	3.57	3.96
$T_6$	2.97	3.02	3.77	3.25	3.43	4.22
$T_7$	3.13	3.31	4.13	3.32	3.19	4.91
$T_8$	2.94	3.43	3.86	3.09	3.05	3.87
$T_9$	3.24	3.15	3.89	2.98	3.46	3.91
$T_{10}$	3.05	3.58	4.12	2.98	3.28	3.68
$T_{11}$	3.18	3.11	4.76	3.80	3.80	4.06
F test	*	*	*	*	*	*
SEM+	0.138	0.049	0.137	0.211	0.058	0.138
CD @ 5%	0.410	0.147	0.407	0.623	0.173	0.418

rhizome was observed with the application of 200 per cent RDF (200:100:100 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>2</sub>) followed by 150 per cent RDF (150:75:75 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>3</sub>) and least length of mother rhizome was observed in 100 per cent application to both the cultivars.

### Length of Primary and Secondary Fingers

Length of primary and secondary fingers showed the significant variation among the cultivars and fertigation levels (Table 5). Maximum length of primary fingers was observed in Himachal (3.75 cm) and Rio-de-Janeiro (3.67cm) with the application of 200 per cent RDF (200:100:100 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>2</sub>) followed by 150 per cent RDF (150:75:75 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>3</sub>). The minimum length was observed with T<sub>1</sub> with 100 per cent RDF in both the cultivars. Similar trend was observed with the length of

secondary fingers in Himachal and Rio-de-Janeiro. Application of 200 per cent RDF (200:100:100 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>2</sub>) resulted in maximum length of secondary fingers and 100 per cent RDF recorded minimum length in both the cultivars. The similar results were quoted by Sharath Pal *et al.* (2014).

### Dry Matter Production per Hectare (t/ha)

The dry matter production per hectare varied significantly among the cultivars and different fertigation levels (Table 6).

Himachal cultivar recorded higher dry matter production (68.25 g/plant and 9.10 t/ ha) and Rio-de-Janeiro recorded the dry matter production of 34.31 g/ plant and 4.57 t/ha with the application of 200 per cent RDF (200:100:100 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>2</sub>) followed by 150 per cent RDF (150:75:75 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg/ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha (T<sub>3</sub>). The least dry matter production was

TABLE 6  
Effect of fertigation levels on total dry matter production of different cultivars of ginger

Treatments	Total dry matter production			
	Rio – de - Janeiro		Himachal	
	g/ plant	t/ha	g/ plant	t/ha
T <sub>1</sub>	22.86	3.04	26.06	3.47
T <sub>2</sub>	34.31	4.57	68.25	9.10
T <sub>3</sub>	28.17	3.75	47.99	6.36
T <sub>4</sub>	26.32	3.50	37.71	5.02
T <sub>5</sub>	23.03	3.06	31.42	4.18
T <sub>6</sub>	28.15	3.75	46.08	6.40
T <sub>7</sub>	27.19	3.62	44.00	5.86
T <sub>8</sub>	19.69	2.62	34.28	4.57
T <sub>9</sub>	26.20	3.49	32.85	4.42
T <sub>10</sub>	28.20	3.86	40.31	5.36
T <sub>11</sub>	29.12	4.10	53.31	7.10
F test	*	*	*	*
SEM+	1.065	0.172	4.767	0.636
CD @ 5%	3.143	0.510	14.063	1.877

recorded with the RDF (100:50:50 N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O kg / ha normal fertilizers) + FYM 30 t/ha, Neem cake 2t/ha (T<sub>1</sub>) in both the cultivars. Himachal cultivar of ginger responded very well for the application of fertigation levels as compared to Rio-de-Janeiro. Variations could be attributed to the difference in genetic makeup and response to inputs by these genotypes. The results are in conformity with the findings of Sudha *et al.* (2020).

The Himachal ginger cultivar performed well for the application of different fertigation levels as compared to the Rio-de-Janeiro cultivar with respect to the plant height, leaf area, leaf area index, number of primary fingers, secondary fingers, fresh weight of mother rhizomes, primary fingers, secondary fingers and length of mother rhizomes, primary fingers, secondary fingers and total dry matter production with different fertigation levels. Application of 200 per cent RDF (200:100:100 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O /ha) Fertigation + FYM 30t/ha, Neem cake 2t/ha resulted in higher plant growth and yield attributes as compared to different fertigation levels in both the ginger cultivars.

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## Evaluation of Genetic Diversity in Germplasm Accessions of Finger Millet [*Eleusine coracana* (L.) Gaertn.]

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### ABSTRACT

Finger millet [*Eleusine coracana* (L.) Gaertn.] is an annual herbaceous small millet crop with nutraceutical value grown in diverse environments. It is a crop of antiquity with great historical, cultural and nutritional importance, particularly in Asia and Africa. Utilization of diverse germplasm is important in breeding programme to improve the yield of the crop. In this regard, 224 finger millet germplasm accessions representing world collections were characterized for various quantitative traits adopting multivariate analysis. Wide variation was observed for most of the traits indicating their importance for direct selection. Among the quantitative traits evaluated, number of productive tillers had a significant positive association with grain yield per plant. Principal component analysis indicated that the first two principal components accounted 56.7 per cent of the total variability. Principal component analysis revealed that days to 50 per cent flowering, plant height, number of productive tillers, main ear length, number of fingers per ear and grain yield per plant contributed most to genetic diversity. Two hundred and twenty-four finger millet germplasm accessions evaluated for seven quantitative traits were grouped into 10 clusters. Thus, the grouping of accessions shall be of practical value to finger millet researchers to select and tap the genetic potential of elite accessions from different clusters as donor parents in crossing programmes.

**Keywords :** Finger millet, Germplasm, Principal component analysis, Cluster analysis, Genetic diversity

**F**INGER millet (*Eleusine coracana* (L.) Gaertn.) is an annual herbaceous small millet crop widely grown and consumed in Africa and Asia. It was domesticated around 5000 years ago in Eastern Africa and introduced in India about 3000 years ago (Hilu *et al.*, 1979). It is an important staple food after rice, wheat, pearl millet and sorghum in India. Finger millet is grown throughout India and is cultivated over 0.97 million hectares with 1.68 mt production and 1662 kg/ha productivity during 2019-2020 (Dept. of Economics and Statistics, DAC & FW, Government of India, New Delhi). The major finger millet-growing states are Tamil Nadu, Karnataka, Orissa, Andhra Pradesh, Uttar Pradesh, Maharashtra, Bihar, Madhya Pradesh and Gujarat.

It serves as a food-security crop because of its high nutritional value and excellent storage qualities (Chandra and Sharma 2016; Ramashia *et al.*, 2019). Finger millet grain is gluten-free, rich in calcium, fiber, iron and essential amino acids, with excellent malting qualities and has low glycemic index (Bruntha Devi *et al.*, 2011). More remarkably, finger millet grain contains higher calcium than other cereals (Kumar *et al.*, 2016) and it is also endowed with abundant phytochemicals, with distinguished health-beneficial properties, making the crop reservoir of health-giving nutrients (Chandra and Sharma, 2016).

In any crop improvement programme genetic variability and diversity play very important roles. The higher diversity between parents shows the higher heterosis and more chance of getting transgressive segregation. To develop improved crop variety over existing cultivated variety breeder has to identify diverse parents having high genetic variability for combining desirable characters. Assessment of large number of germplasm accessions for genetic diversity is of immense help in the selection of diverse genotypes for hybridization programme (Reddy *et al.*, 2015). Multivariate hierarchical cluster analysis helps in the initial grouping of accessions. Principal Component Analysis is used to confirm the diversity pattern brought about by cluster analysis. Hence the present study was undertaken to characterize the germplasm accessions for yield and attributing traits by means of descriptive statistic and to understand the association of various characters, PCA and cluster analysis which would enable breeders to classify the available germplasm into distinct groups on the basis of genetic diversity.

#### MATERIAL AND METHODS

*Experimental Material* : The experimental material consisted 224 finger millet germplasm accessions representing collections across the world (Table 1) which are maintained at the National Active Germplasm Site, Project Coordinating Unit, AICRP

on small millets, University of Agricultural Sciences, GKVK, Bengaluru.

*Evaluation of Experimental material* : The present experiment was conducted during *kharif* 2020 at the experimental farm of All India Coordinated Research Project on small millets, University of Agricultural Sciences, GKVK, Bengaluru located at 13° 05" N latitude and 77° 34" E longitudes. The center is at an altitude of 924 meters above mean sea level. The annual rainfall ranges from 528 mm to 1374.4 mm with the mean of 915.8 mm. 224 germplasm accessions consisted of indigenous collections, land races, exotic collections and white seeded accessions that were sown in a single row of 3m length and row to row spacing of 22.5cm. The experimental material was divided into 14 blocks; each block consisted of 16 accessions and 3 checks (KMR 340, GPU 66 and KMR 630) following the augmented design. The crop was supplied with recommended dose of fertilizers as per the package of practices. Observations were recorded from five randomly selected plants in each accession for seven quantitative characters *viz.*, Days to 50 per cent flowering, Days to maturity, Plant height (cm), Number of productive tillers, main ear length (cm), number of fingers per ear and grain yield per plant(g).

*Statistical analysis* : Summary statistics like, mean, range, variances and standard deviation were estimated using Microsoft Excel. The data recorded on the quantitative traits was subjected to analysis of variance (Federer and Raghavarao, 1975). Phenotypic correlation coefficients were calculated as suggested by Johnson *et al.*, (1955). PCA was computed for 7 quantitative traits to find out the relative importance of different traits in capturing the variation in the entire germplasm set using OPSTAT statistical package as suggested by Johnson and Wichern (1988). The seven quantitative characters were subjected to diversity analysis to determine K means cluster using R software version 4.0.2.

#### RESULTS AND DISCUSSION

Analysis of variance (Table 2) showed significant differences for all quantitative characters suggesting

TABLE 1

Details of 224 finger millet germplasm accessions evaluated in the experiment

Source	Number of accessions evaluated
Uganda	10
Kenya	5
Malawi	15
Ethiopia	4
Zambia	25
Zimbabwe	4
Tanzania	2
India	159
Total	224

TABLE 2  
Analysis of variance (ANOVA) of yield in finger millet germplasm accessions

Source of Variation	DF	Sum of Squares	Mean Squares	Significance
Blocks (eliminating treatments)	13	22.192	1.707	0.0699
Treatment (ignoring blocks)	226	10,863.60	48.069 *	0
Checks	2	128.554	64.277	0
Accessions	223	10,735.03	48.139	0
Checks vs.accessions	1	0.022	0.022	0.87379
Error	26	22.643	0.871	
Total	265	10,908.44		

TABLE 3  
Summery statistics of various quantitative traits evaluated in 224 finger millet germplasm accessions

Characters	Mean	Range	Variance	Standard division
Days to 50% flowering	72.112	58.00 - 88.00	35.66	5.972
Days to maturity	113.241	99.00 - 128.00	36.444	6.037
Plant height (cm)	96.882	65.0 - 133.00	124.871	11.175
No. of productive tillers	4.761	2.00 - 10.60	1.822	1.35
No. of fingers /ear	6.356	3.00 - 8.00	0.626	0.791
Main ear length (cm)	5.442	3.10 - 14.50	1.628	1.276
Grain yield per plant (g)	19.18	6.00 - 58.33	48.139	6.938

significant variability for all the traits. The mean, range, variance and standard deviation for all the seven quantitative traits are presented in Table 3. Days to 50 per cent flowering ranged from 58 days (GE 133) to 88 days (GE 4969) days with mean value of 71.11. Accessions GE 249, GE 289 and GE 68 were early to flower; hence these accessions could be utilized for developing early to medium duration varieties in finger millet. The accessions *viz.*, GE 581 which is a land race called Nalla Gidda Ragi and GE 80 source from Africa were the most dwarf with height of 65 to 69 cm, while the accession GE 118 was the tallest with a height of 133 cm. These accessions can be utilized as a donor for breeding dwarf varieties. Wide range of variation was observed for number of productive tillers, main ear length and number of fingers per ear head. The accession GE 135 recorded

a higher number of productive tillers among the accessions. The accession GE 4705 had higher number of fingers (8 no's), whereas GE 12 had longer ear head (14.50cm). The accession GE 135 expressed superior grain yield levels (58.33 g/plant) and produced numerous productive tillers (10 tillers). Most of the high yielding accessions were from Indian origin followed by Africa and Zambia. The landraces collected from India are low yielders. Most promising trait donors for various quantitative traits identified based on average are presented in Table 4.

#### Relationship Between Traits

The correlation coefficients between traits are presented in Table 5. The evaluation of data provides a valuable opportunity for assessing relationships among traits to test the similarity between different



TABLE 4  
Identity of the promising finger millet trait donors with desirable quantitative traits

Characters	Promising accession
Days to 50% flowering (61-63 days), early duration	GE 249, GE 289, GE 68, GE 118, GE 155, GE 15, GE 31, GE 62, GE 100, GE 114, GE 117, GE 142, GE 157
More number of tillers (7-10)	GE 135, GE 274, GE 98, GE 59, GE 150, GE 153, GPU -W-1B, GE 15, GE 4801
Plant height (cm) (65-75 cm), dwarf accessions	GE 581, GE 80, GE 10, GE 6636, GE 4682, GE 170, GE 4706, GE 5155, GE 4895
Main ear length (8-14cm)	GE 12, GE 153, GE 69, GE 4, GE 86, GE 1324, GE 46, GE 169, GE 4805, GE 4808, GE 4902
More number of fingers per ear (7-8)	GE 4701, GE 5, GE 6, GE 26, GE 44, GE 133, GE 153, GE 198, GE 29
High yield (33-58g)	GE 135, GE 15, GE 1318, GE 249, VL356, GE 150, GE 4981, GE 67, GE 3371

TABLE 5  
Phenotypic correlation co-efficient between 7 quantitative traits in 224 finger millet germplasm

Characters	Days to 50% flowering	Days to maturity	Plant height (cm)	No. of productive tillers	No. of fingers /ear	Main ear length (cm)	Grain yield per plant (g)
Days to 50% flowering	1	0.979	0.383	-0.312	-0.297	0.127	-0.17
Days to maturity		1	0.388	-0.3	-0.289	0.126	-0.179
Plant height (cm)			1	-0.246	-0.128	0.426	-0.101
No. of productive tillers				1	0.134	-0.078	0.382
No. of fingers /ear					1	0.128	0.013
Main ear length (cm)						1	-0.115
Grain yield per plant (g)							1

groups. This practice simplifies work and saves resources. Association studies among different characters are important for finger millet breeders in the effective selection of desirable genotypes. The knowledge on the association between grain yield and other attributing quantitative traits helps in improving the efficiency of selection. Number of productive tillers (0.382) had a significant positive association with grain yield per plant. This association suggests that number of productive tillers will be effective selection indices for grain yield. Similarly, Dagnachew *et al.*, 2012, Kadam *et al.*, 2009 and Nandini *et al.*, 2018, reported that number of productive tillers had significant positive association with grain yield per plant. Significant negative correlation was observed for days to 50 per cent

flowering and days to maturity with grain yield per plant. This indicates that increase in one trait would lead to decrease in other trait. The negative association of days to 50 per cent flowering with grain yield is beneficial association because early maturing types are most preferred by farmers. Early duration types will escape from drought situations. Thippeswamy and Sajjanar, 2017, reported days to 50 per cent flowering have negative association with grain yield in foxtail millet and Nandini *et al.*, 2018 in finger millet.

#### Principal Component Analysis (PCA)

PCA was applied as a reductionist approach to the multivariate data, to measure the importance and contribution of each component to the total variance.

TABLE 6  
Vector loadings and percentage of variation explained by the first five principal components in 224 finger millet germplasm

	PC1	PC1	PC1	PC1	PC1
Eigen values	2.677	1.295	1.133	0.791	0.613
Variance explained (%)	0.382	0.185	0.162	0.113	0.088
Cumulative variance explained (%)	0.382	0.568	0.729	0.842	0.93
<b>Eigen vectors</b>					
Days to 50% flowering	0.547	-0.22	0.146	0.35	-0.039
Days to maturity	0.547	-0.214	0.148	0.359	-0.055
Plant height (cm)	0.381	0.355	0.301	-0.344	0.218
No. of productive tillers	-0.335	-0.108	0.546	0.191	-0.668
No. of fingers /ear	-0.234	0.508	-0.031	0.754	0.301
Main ear length (cm)	0.193	0.671	0.298	-0.149	-0.269
Grain yield per plant (g)	-0.228	-0.244	0.691	-0.054	0.582

The *per cent* variation explained by the first four principal components (PCs) and vector loadings for each trait is presented in Table 6. The first four PCs accounted to 84.2 *per cent* of the total variation and subsequent components contributed 5 per cent or less variation. Vaishali *et al.*, (2021) reported that first five principal components explained 99.97 *per cent* of the entire variability in finger millet. Similarly, Patil *et al.*, 2019, reported genetic diversity in finger millet using principal component analysis and found first three principal components showed 98.31 *per cent* of total variation. PC1 accounted to 38.2 per cent of the total variation. PC1 was attributed to days to 50 per cent flowering, days to maturity and plant height for largest positive loadings indicating its significant importance in these components. Number of productive tillers and number of fingers per ear and grain yield per plant had negative loadings. PC2 accounted for 18.5 per cent of the total variation and contributed to positive loadings for plant height, number of fingers per ear and main ear length whereas, all other traits contributed negatively. PC3 accounted for 16.2 per cent of the total variation. All the traits contributed to positive loadings except number of fingers per ear head. Similarly, PC4 accounted for 11.3 per cent of

the total variation, the major traits that contributed highly to the variation include number of fingers per ear head, days to 50 per cent flowering and days to maturity, whereas, number of productive tillers contributed least to the variation. Similar findings with regard to grain yield per plant, plant height, days to 50 per cent flowering and productive tillers per plant were reported by Salini *et al.*, 2010 in proso millet and Nandini *et al.*, 2018; Aradhana *et al.*, 2019 in finger millet. Principal component analysis revealed that days to 50 per cent flowering, plant height, number of productive tillers, main ear length, number of fingers per ear and grain yield per plant contributed most to genetic diversity.

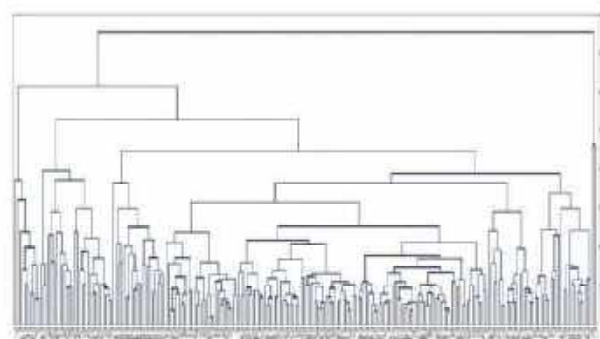


Fig. 1 : Hierarchical clustering pattern of 224 finger millet germplasm accessions

### Cluster Analysis

K means cluster analysis based on agronomic traits grouped 224 finger millet germplasm into 10 clusters. Hierarchical clustering pattern of 224 finger millet germplasm accessions presented in Fig. 1. Cluster II, with highest number of germplasm accessions (49) was identified as the largest cluster, whereas clusters VII, VIII and X was with 10 germplasm accessions. The high grain yielding accessions (GE 135, GE 15 GE 249 and GE 150) and accessions with more number of productive tillers were registered in cluster I and are Indian origin. Cluster V comprised land races, whereas white seeded accessions registered in clusters VII and X. The dwarf accessions GE 4682, GE 6636 and GE 5155 were found in cluster II. This indicated that accessions in these clusters have wide diversity for various characters. Similar results have been reported by Nandini *et al.* (2020) in which the barnyard millet germplasm accessions used were grouped into 23 clusters and in foxtail millet, 23 clusters (Nandini *et al.* 2018) have been made using 1312 foxtail millet germplasm accessions. Manimekalai *et al.*, 2018, grouped 61 genotypes of barnyard millet germplasm in 13 clusters based on the bouches clustering technique. Likewise, Sood *et al.*, 2015 grouped 95 germplasm accessions of barnyard millet into two groups. Group A comprised of 43 accessions and Group B comprised of 51 accessions using two-way cluster analysis.

Cluster means of all seven quantitative traits are presented in Table 7. The accessions in the cluster I showed more mean grain yield followed by cluster III and IX. Accessions which are present in these diverse clusters can be utilized for hybridization to get transgressive segregants for that trait, which could be used for developing superior high grain yielding varieties. The dwarf accessions could be utilized in developing dwarf varieties in finger millet. Likewise, accessions in diverse clusters could be utilized in hybridization programme to develop high grain yielding, dwarf and high tillering ability types in finger millet.

Relationship among the accessions and assessment of genetic diversity among various accessions is fundamental importance to exploit the finger millet genetic resources. Multivariate statistical analysis provides a means for estimating morphological diversity between germplasm accessions. The potential breeding value of germplasm can be evaluated by these tools. The PCA and cluster analysis provided a simplified classification finger millet germplasm accessions for use in breeding. Principal component analysis revealed that days to 50 per cent flowering, plant height, number of productive tillers, main ear length, number of fingers per ear and grain yield per plant contributed most to genetic diversity. Number of productive tillers had significant positive association

TABLE 7  
The average of 7 quantitative traits for each cluster in 224 finger millet germplasm

Cluster Number	Days to 50% flowering	Days to maturity	Plant height (cm)	No. of productive tillers	No. of fingers /ear	Main ear length (cm)	Grain yield per plant (g)
I	63.82	104.82	85.49	6.98	6.60	4.96	35.58
II	74.55	116.00	101.64	4.51	6.12	5.56	15.18
III	71.68	112.90	106.96	4.67	6.32	6.00	27.02
VI	65.80	106.70	77.80	4.71	6.48	4.57	17.43
V	65.00	106.10	94.62	4.87	6.70	5.28	17.00
VI	75.94	116.91	89.35	4.66	6.29	5.18	15.71
VII	81.50	122.30	114.08	3.43	6.09	5.65	16.20
VIII	72.20	113.20	122.03	4.52	6.28	7.12	15.07
IX	72.19	113.25	93.50	5.24	6.59	5.42	23.00
X	82.80	124.10	104.77	3.84	5.86	5.64	22.07

with grain yield per plant. This association suggests that number of productive tillers will be effective selection indices for grain yield. Hybridizing the accessions belonging to different clusters would maximize opportunities for transgressive segregation because of higher probability that unrelated genotypes will contribute unique desirable alleles at multiple loci (Beer *et al.*, 1993). Thus, the grouping of accessions by multivariate methods in present study will be of practical value to the finger millet breeders in allowing them to choose elite accessions from different clusters as parental line for hybridization programmes.

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## Molecular Detection and Virulence Profiling of Associated Bacterial Pathogens Causing Bacterial Blight in Rice

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### ABSTRACT

Bacterial leaf blight of rice caused by *Xanthomonas oryzae* pv. *oryzae* (*Xoo*) is one of the most destructive diseases. The disease severity of this differs from geographical regions, strain (race) and the host genotype. Hence, isolates were collected from different locations of Karnataka to know the virulence of *Xoo* and recorded disease severity. Cultured isolates were cultured in the laboratory and were confirmed by 16S rRNA sequencing as well isolated *Pantoea ananitis* associated with bacterial blight and producing similar kind of symptoms as that of *Xoo*. The ClustalW analysis of the 16S rRNA gene sequences revealed two groups in the case of both *Xoo* and *P. ananitis*. The second group containing only two isolates *Xoo* 4 and *Xoo* 9 are more similar to the MH444307 isolate from Maharashtra and is delineated from the rest of the *Xoo* isolates. The 35 isolates of *P. ananitis* also formed into two groups and had more similarity with Japan isolate AB304809 followed by South Korean isolates HE672167 and HE716948 respectively. Around 42 resistance genes of rice (*Xa* genes), have been identified to confer resistance against various strains of *Xoo*. An understanding of pathotype diversity within the target pathogen population is required to identify the *Xa* genes to be deployed for developing resistant rice cultivars. The isolates of *Xoo* and *P. ananitis* collected from different parts of Karnataka formed 2 major pathotypes of both *Xoo* and *P. ananitis* and were distinguished based on their reaction towards *Xa* genes in the monogenic lines. The resistance gene *Xa21* conferred resistance to more than 70 per cent of the *Xoo* and *P. ananitis* population. Further the amounts of exopolysaccharide (EPS) produced were quantified and varied depending on *Xoo* isolates. Isolate *Xoo* 3 produced a distinctly high amount of EPS (92.3 mg) followed by *Xoo* 6, *Xoo* 8 and *Xoo* 4. On the contrary, *Xoo* 2 produced distinctly less amount of EPS that has been co-related with the virulence nature of the pathogen on the rice cultivar, TN-1.

Keywords : *Xanthomonas oryzae* pv. *oryzae*, *Pantoea ananitis*, Pathotype, EPS

RICE, one of the most important food crop, is constantly challenged by bacterial pathogens, such as those causing bacterial leaf blight (BB), leaf streak, and bacterial panicle blight. Leaf blight is a serious problem in rice agroecosystems of many countries, causing significant economic losses worldwide (Chukwu *et al.*, 2019). The impact of leaf blight on rice farming systems appears to be increasing, and causes yield loss up to 70 per cent in various countries. This disease is most prevalent and

destructive in tropical Asia (Mew *et al.*, 1993). In India, the first report on BLB was made by Bhapkar *et al.* (1960) and it is one of the most devastating diseases during monsoon season and a major production constraint in rice cultivation particularly in irrigated and rainfed lowland ecosystems of rice growing states of India. For decades, the gram-negative bacterium *Xoo* has been widely regarded as the only causal agent of leaf blight in rice (Chien *et al.*, 2019).

However, several studies in recent past have indicated that many species from the genus *Pantoea* are capable of causing the symptoms of leaf blight disease. *Pantoea* spp. is commonly associated with plants as epiphytes or pathogens (Deletoile *et al.*, 2009). Recent evidence has provided additional support, where the *Pantoea* species have re-emerged as a threat to global rice production as they have been shown to cause various rice diseases in several rice growing areas of the world (Azizi *et al.*, 2020). The *Pantoea* spp. was described as a causal agent of leaf blight in India (Mondal *et al.*, 2011) and Korea (Lee *et al.*, 2010). Recent reports by Toh *et al.* (2019) and Azizi *et al.* (2020) indicated that *P. ananatis*, *P. dispersa* and *P. stewartii* were all recorded to be the causal agents of leaf blight outbreak in Malaysia.

Bacterial blight is characterized by a high degree of race cultivar specificity. *Xoo* races is differentially infecting rice cultivars and distributed in geographic regions over time periods. Hence, *Xoo* is a rapidly evolving pathogen (Salzberg *et al.*, 2008). The selection of cultivated rice varieties may facilitate the race shift or result in the emergence of new races. Indeed, a prior study showed that the shifting of the major race over time in the Philippines might be caused by a dramatic change in the host genotypes (Quibod *et al.*, 2016). The population dynamics of *Xoo* monitored in resistant and susceptible rice cultivars showed that bacterial populations in compatible and incompatible interactions increased almost equally in the initial stage of infection. There after, however, the virulent population multiplied more rapidly and extensively than the avirulent ones (Noda and Kaku, 1999).

Chemical control is not very successful in India and elsewhere for the management of the disease (Laha *et al.*, 2009). Therefore, deploying of resistance genes to increase host plant robustness is economically and environmentally advantageous and the most promising strategy to manage BLB. So far, more than 45 BLB resistance genes have been identified from diverse sources. Some of them have been introgressed to suitable agronomically important rice cultivars to develop resistance against BB in several countries

(Yugander *et al.*, 2018). The interlinked evolution of *Xoo* and rice harbours the potential for selecting emerging virulence factors (Mishra *et al.*, 2013). Therefore, regular monitoring of BB pathogen is essential to identify the existing and evolving pathotypes of *Xoo* in a particular region as a prerequisite for developing of durable BB resistant rice cultivars.

Analysis of BLB pathogen population structure using both classical pathotyping on rice differentials, International Rice Bacterial Blight (IRBB near isogenic lines) and genetic diversity using molecular markers has been reported by several workers both in India and elsewhere (Yugander *et al.*, 2017). The present study was undertaken to identify the pathotypes and virulence profile of *Xoo* and *P. ananatis* isolates collected from selected areas of Karnataka. The findings of this study will help for breeding BB resistant rice varieties suitable for the region.

## MATERIAL AND METHODS

### Collection of BB Diseased Leaf Samples

A field survey was undertaken in major rice growing regions of Karnataka India, during November 2020 and June 2021. A total of 45 samples were collected from different locations in Karnataka. Plants with typical bacterial leaf blight symptoms *viz.*, yellow water-soaked lesions at the margin of the leaf blade, the lesions run parallel along the leaf, presence of bacterial discharge on young lesion early in the morning resembling the milky dewdrop, drying up of leaf blade with white lesions as wavy margin were collected in blotting paper folds, labelled, and then in a paper envelope. The samples were brought to the laboratory and stored at 4 °C for isolation of the pathogen.

### Isolation of the Pathogen

Isolation of pathogen was carried out from infected rice leaves collected during the survey. The leaves showing typical symptoms of leaf blighting, exuded bacterial ooze from the cut section were used for isolation. The diseased portion with healthy tissues was cut into 1.5 to 2 cm pieces. These diseased pieces

were surface sterilized for 30 seconds in 0.1 per cent sodium hypochlorite solution, followed by three subsequent washing with sterilized distilled water in aseptic conditions to remove the traces of sodium hypochlorite (Bakade and Kumar, 2020). The bacterial suspension was prepared with the cut pieces of infected leaf samples. Following serial dilution, the suspension was streaked on Peptone Sucrose Agar (PSA) medium with the help of a sterilized wire loop. The inoculated plates were incubated at room temperature ( $27 \pm 2$  °C) for 48 hrs. Single colonies of cultures isolated from disease samples were picked up from PSA plates and the pure culture, thus obtained was preserved in slants for further investigations in the refrigerator at 4 °C for routine work and in 50 per cent glycerol at -80 °C for long term storage.

### Pathogenicity Test

To confirm the pathogenicity of the bacteria isolated from the bacterial blight samples, the isolates were multiplied in a nutrient broth medium. The pathogenicity test was carried out on rice plants (cv. TN-1) in the glasshouse using leaf clip inoculation technique (Kaufmann, 1973). The pots were filled with sterilized soil and TN-1 seeds were sown in the prepared pots. Two seedlings per pot were raised for 50 days, covered with plastic to avoid air borne infection. The pots were labelled, watered gently and all the agronomical practices were adopted for growing the rice plants.

### Molecular Confirmation of the Pathogen Using 16S rRNA and Phylogenetic Analysis

Genomic DNA of the bacterium was extracted as described by Zhang *et al.* (1998) with some modification. 0.3 g of washed bacterial cells pellet was suspended in 200 µl of Cetyltrimethylammonium bromide (CTAB) buffer (50 mM Tris, pH 8.0; 0.7 mM NaCl; 10 mM EDTA; 2 Cetyltrimethylammonium hexadecyltrimethylammonium bromide) followed by 100 µl of 10 per cent sodium dodecyl sulfate and incubated at 65 °C for 10 min. DNA was purified with chloroform and precipitated with iso-propanol at -20 °C overnight. Purified DNA was washed with 70 per cent ethanol, then the pellet was air

dried and resuspended in 30 µl of DNase free sterile distilled water. DNA concentration was measured using NanoDrop (De Novex) at 260 / 280 nm. DNA quality was checked on 0.8 per cent agarose gel in Tris-Acetate-EDTA (TAE) buffer (45 mM Tris-acetate, 1 mM EDTA, pH 8.0). After electrophoresis, the genomic DNA was further used for PCR amplification with universal 16S rRNA primers (F-5'-GAGTTTGATCCTGGCTCA-3'; R-5'-AGAAAGGAGGTGATCCAG-3'). PCR was performed in a thermal cycler (Eppendorf, Vapo protect), with 100 ng of genomic DNA, 1 µl of each primer, 10 µl of 2 x PCR master mix and sterile distilled water to make a final volume of 20 µl. The thermal cycler was programmed with an initial denaturation at 94 °C for four min followed by 35 cycles at 94 °C for 1 min, 58 °C for 1 min and 72 °C for 1 min with the final extension at 72 °C for 10 min. The amplified products were purified and directly sequenced from both ends using a commercial facility (Eurofins Scientific India Pvt. Ltd.). Obtained DNA sequences were analysed and compared with the available NCBI database using BLAST analysis.

The phylogenetic analysis using 16S rRNA gene sequences from *Xoo* strains and *P. ananitis* was done using CLC genomic workbench (version 4.7.2) software programme using neighbor-joining method. The percent identity was determined using ClustalW multiple alignments of BioEdit software. The type strains of *P. ananitis* NR 026045, *Sphingomonas endophytica* MN932317, the common epiphytes in rice were kept as outgroups in determining the phylogenetic relationship among the *Xoo* strains. The GenBank accession numbers of 16S rRNA gene from *Xoo* strains are OP048090, OP048091, OP048092, OP048093, OP048094, OP048095, OP048096, OP048097, OP048098 and OP048099. The type strains of *Exiguobacterium* MH753639, *Xanthomonas oryzae* pv. *oryzae* AB68014, *Sphingomonas endophyticum* MN932317 the common epiphytes in rice were kept as outgroups in determining the phylogenetic relationship among the *P. ananitis* strains. The GenBank accessions of 16S rRNA genes from *P. ananitis* strains are



OK576910, OK576911, OK576912, OK576913, OK576914, OK576915, OK576916, OK576917, OK576918, OK576919, OK576920, OK576921, OK576922, OK576923, OK576924, OK576925, OK576926, OK576927, OK576928, OK576929, OK576930, OK576931, OK576932, OK576933, OK576934, OK576935, OK576936, OK576937, OK576938, OK576939, OK576940, OK576941, OK576942, OK576943 and OK576944.

### Pathotype Analysis

The seeds of bacterial blight differential rice lines (IRBB-1, 3, 4, 5, 7, 8, 10, 11, 13, 14, 21), the susceptible check line (IR24) and the resistant check line (RP Bio-226) were provided by the Zonal Agricultural Research Station (ZARS), Mandya. The plants were grown in pots in a greenhouse. Sowing of the differentials was done in pots and 50 days old plants were preferably used for inoculating the pathogen (Yugander *et al.*, 2022). The trays were irrigated every day. Adequate plant protection measures were taken to ensure the healthy and vigorous growth of the plants. Plants were clip inoculated with bacterial suspensions of  $10^9$  cfu/ml. Four leaves per plant were inoculated for each isolate-cultivar combination for 50 days old plants. Disease observations were taken 14 days after inoculation by measuring lesion length. Lesion lengths  $\leq 5$  cm was considered as resistant; 5-10 cm was considered as moderately resistant and  $>10$  cm was considered susceptible. Pathotype grouping was done based on the reaction pattern onto the differentials. Cluster analysis was carried out using the unweighted neighbor joining method and the robustness of the tree/cluster was assessed with 1000 bootstraps using the DARW in software.

### Measurement of Exopolysaccharide (EPS)

The measurement of exopolysaccharide (EPS) was conducted as described by Jeong *et al.* (2008). A single colony of each *Xoo* isolate was inoculated in 40 ml of nutrient broth medium and incubated for 72 h at 28 °C with agitation. The optical density of the bacterial cultures was adjusted to 1.0 at 600 nm with NB. The culture supernatants were transferred into new 50-ml tubes and supplemented with 1.0 per cent potassium chloride (w/v; final concentration). Two volumes of absolute ethanol were added to each solution and the tubes were placed at -20 °C overnight. The precipitated crude EPS was collected by centrifugation for 30 min at 83,000×g. The EPS pellets were dried at 55 °C for 12 h and the dry weight of each was measured.

## RESULTS AND DISCUSSION

### Survey and Collection of Infected Leaf Samples

The bacterial blight infected leaf samples of rice collected from different regions of Karnataka exhibited the characteristic symptom of yellow water-soaked lesions at the margin of the leaf blade and the lesions run parallel along the leaf with the bacterial discharge appears on young lesion early in the morning resembling the milky dew drop, as the disease progress the leaf dries up with white lesions and the leaf blade as wavy margin (Plate 1). Similarly, symptoms were reported by (Bakade and Kumar, 2020) collected infected rice leaf samples from different rice growing regions of Karnataka, Andhrapradesh and Tamil Nadu.



Plate 1: Symptoms of bacterial blight on rice leaves as observed under field conditions

### Isolation and Pathogenicity Assay

The BLB infected rice samples collected during survey were homogenized in sterile distilled water for isolation of pathogen/s on PSA medium. Individual bacterial colonies on PSA medium were yellow, mucoid and convex as reported by Chauhan (1973) and Ou (1985). Pathogenicity of the 45 isolates was proved by adopting the leaf clip inoculation technique (Kaufmann, 1973). The bacterial suspension of all the isolates were inoculated on TN-1 (45 days old plants) in glass house. Symptoms of typical BLB were observed from the 3<sup>rd</sup> day of inoculation. The pathogen was re-isolated from the BB infected plants and thus confirmed its identity and pathogenicity (Plate 2). Based on the pathogenicity tests, the bacterium was confirmed as *Xanthomonas oryzae* pv. *oryzae*. This pattern of morphological characters of *Xoo* observed in the present study was in accordance with the description given by Chauhan (1973) and Ou (1985). Symptoms on the grown-up plant were manifested as rolling of leaves, greyish lesions with wavy margins, wilting of the plant (Laha *et al.*, 2009). The 45 isolates were subjected to 16Sr RNA sequencing for molecular confirmation. Of the 45 strains, 35 were identified as

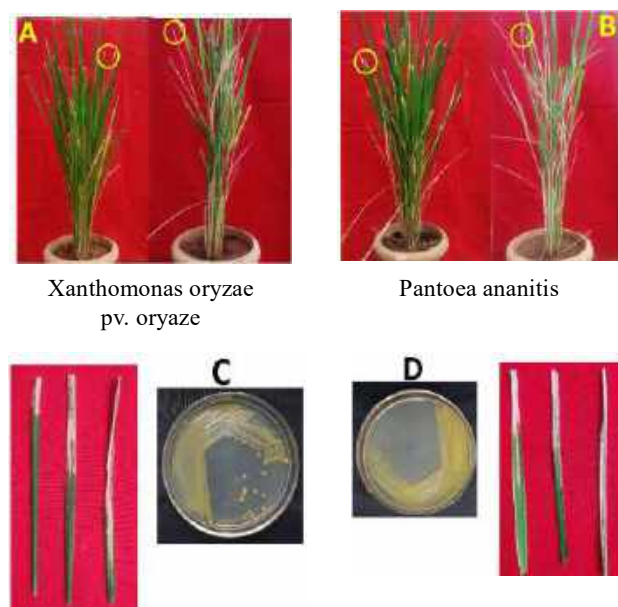


Plate 2: A, B: Symptoms of bacterial blight on rice leaves as observed under glasshouse conditions upon inoculation with *Xoo* and *P. ananitis*, respectively; C, D: Colony morphology of *Xoo* and *P. ananitis*, respectively

*P. ananitis* and 10 as *Xoo*. Reported symptoms observed from *P. ananitis* infection are almost consistent with *Xoo* caused leaf blight in our study. Further, the plants infected with *P. ananitis* exhibited the tale like lesions along the leaf margin as described by Doni *et al.* (2019).

### Molecular Confirmation Using 16SrRNA and Phylogenetic Analysis

The genomic DNA of 45 bacterial isolates were amplified using 16s rRNA universal primers and observed the amplicon of expected 1500bp size in all the isolates (Bakade and Kumar, 2020). The 16s rRNA gene of all the *Xoo* isolates were sequenced and compared with the previously published 16s rRNA gene sequences of *Xoo* using the NCBI-BLAST program. The sequence identities among these 10 isolates are 99-100 per cent. Phylogeny based on 16S rRNA gene revealed that all 10 strains of *Xoo* from Karnataka formed two clusters sharing 100 per cent identity among them, suggesting that geographical distance does not necessarily contribute to the variation in *Xoo* strains with respect to the gene loci, 16S rRNA (Fig. 1) (Kalyan, 2013). The ClustalW analysis of the 16S rRNA gene sequences revealed two groups. The first group includes almost all the isolates viz., *Xoo* 1, *Xoo* 2, *Xoo* 3, *Xoo* 5, *Xoo* 6, *Xoo* 7, *Xoo* 8 and *Xoo* 10. The second group contains only two isolates *Xoo*4 and *Xoo*9 which are from the Koppal district of Karnataka and are more similar to the MH444307 isolate from Maharashtra. The first cluster was divided into two sub clusters where the isolates *Xoo* 1, *Xoo* 2, *Xoo* 6 and *Xoo* 10 from Gangavathi showed high similarity with ON908986 *Xoo* from Shivamogga. The isolate *Xoo* 5 showed high similarity with JQ269244 *Xoo* from Tamilnadu, where, JQ269244 *Xoo* was found to be sub- ancestor for *Xoo* 3, *Xoo* 7 and *Xoo* 8. The 35 isolates of *P. ananitis* also formed into two groups and are more similar with Japan isolate AB304809 followed by South Korean isolates HE716948, HE672167 and all the *P. ananitis* isolate have clustered with in the Karnataka isolates (Fig. 2).

The use of 16S rRNA gene sequences to study bacterial phylogeny and taxonomy has been by far

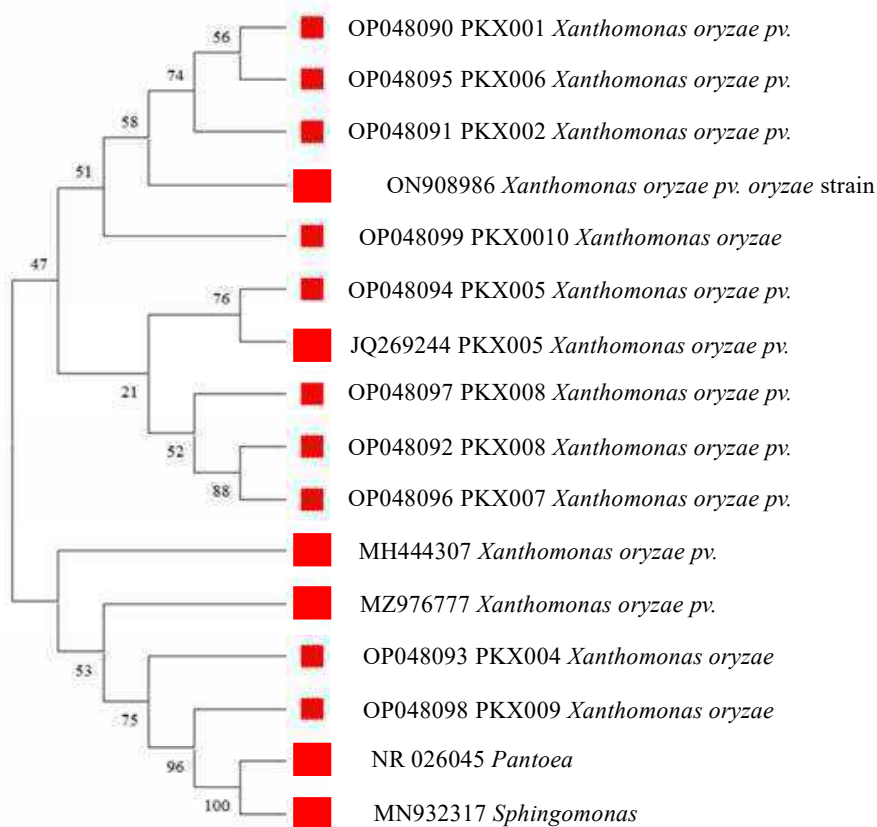


Fig 1: Phylogenetic tree construction using ClustalW analysis of 16S rRNA sequence of Indian Xoo isolates

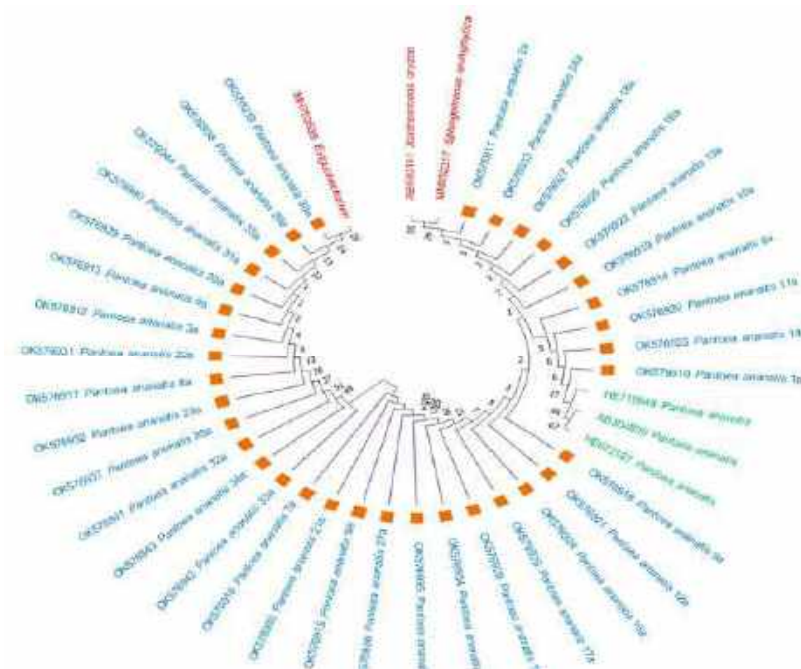


Fig 2: Phylogenetic tree construction using ClustalW analysis of 16S rRNA sequence of Indian *P. ananitis* isolates

the most common housekeeping genetic marker (Ashmawy *et al.*, 2015). 16S rRNA gene was used to identify the tested isolates and study the genetic variability among 35 isolates of *P. ananitis* and 10 isolates of *Xoo*. Results obtained from all tested isolates gave a band in the right expected molecular length. DNA sequences of tested isolates revealed that the sequences belong to *P. ananitis* and *Xoo*. The findings confirm with data from Krawczyk *et al.* (2010).

### Pathotype Analysis

The phylogenetic neighbour-joining tree constructed using phenotypic data of differential reaction on differential lines divided 35 isolates of *P. ananitis* into two major clusters and a sub-cluster under each (Fig. 3). The clusters were designated as pathotypes and were classified based upon the similarity coefficient given based on differential reaction produced by the differential lines upon inoculation with the pathogen. Pathotype I, the largest cluster, contained 19 *P. ananitis* isolates while pathotype II

contained 16 *P. ananitis* isolates. Pathotype I and II were further divided into two pathotypes IA, IIA, IB and IIB (Table 1). The frequency of the isolates under pathotypes IA, IB, IIA, and IIB is 42, 11, 25 and 20 per cent respectively. The results indicate that majority of the isolates fall under pathotype IA since most of the collections were obtained from Gangavathi under the Tungabhadra command area where BB was drastic during the respective year 2020. The results indicate a strong correlation between pathotype diversity since the isolates were collected only from two command areas of Karnataka and within the cluster the isolates exhibited pathogenic diversity based on the differential reaction produced by the monogenic differential lines were in pace with results as obtained by (Mishra *et al.*, 2013). Considering the individual R-genes, *Xa21* appeared as the most broadly effective, conferring resistance against 85 per cent of the isolates, followed in decreasing order by *Xa11* (54%), *Xa13* (45%), *Xa14* (45%), *Xa8* (28%), *Xa5* (28%), *Xa3* (22%), *Xa4* (20%) (20%), *Xa7* (14%), *Xa10* (14%), *Xa1* (5%) (Fig. 4). The R-genes *Xa1*, *Xa3*, *Xa4*, *Xa7* and *Xa10* were the least effective against any pathotypes.

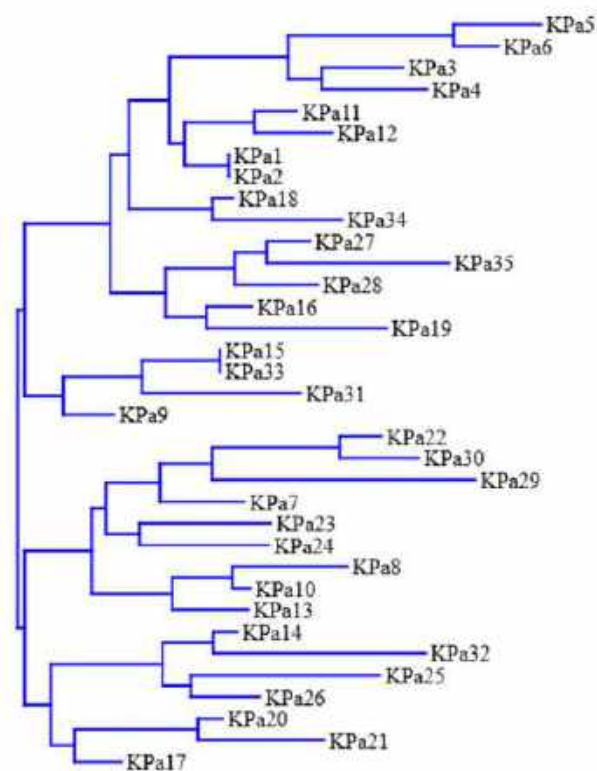


Fig. 3: Dendrogram showing the pathogenic relatedness of 35 *P. ananitis* isolates collected from Karnataka

TABLE 1

Details of the number of pathotypes and isolates belonging to each pathotype

Patho- types	No. of isolates	<i>Pantoea</i> isolates
IA	15	KPa #- 1-6, 11-12, 16, 18-19, 27-28, 34-35
IB	4	KPa #- 9, 15, 31, 33
II A	9	KPa #- 7-8, 10, 13, 22-24, 29-30
II B	7	KPa #- 14, 17, 20-21, 25-26, 32

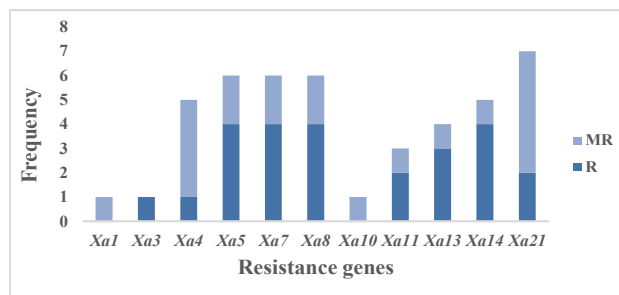


Fig. 4: Effectiveness of Xa genes against Indian *P. ananitis* isolates. The x-axis indicates resistance gene and the y-axis indicates the frequency of isolates against which this gene confers resistance

Similarly, pathotype analysis was performed for the 10 *Xoo* isolates, pathotype I is the largest cluster, containing 8 *Xoo* isolates and pathotype II contained 2 *Xoo* isolates (Fig. 5). The results did not indicate a strong correlation between pathotype diversity since the isolates were collected only from one command area of Karnataka *i.e.*, Tungabhadra command area. Within the cluster, the isolates exhibited the pathogenic diversity based on the differential reaction produced by the monogenic differential lines. Considering the individual R-genes, *Xa21* appeared as the most broadly effective, conferring resistance against 70 per cent of the isolates, followed in decreasing order by *Xa5* (60 %), *Xa7* (60 %), *Xa8* (60 %), *Xa14* (50 %), *Xa4* (50 %) *Xa13* (40 %) (Fig. 6). The R-genes *Xa1*, *Xa3*, *Xa10* and *Xa11* were least effective against any of the pathotypes. With all the pathotypes of *Xoo* and *P. ananitis*, *Xa21* exhibited more than 70 percent of resistance. These *Xa21*-mediated basal pathways included mainly those related to the basic material and energy metabolisms and many related to phytohormones such as cytokinin, suggesting that *Xa21* triggered redistribution of energy, phytohormones and resources among essential cellular activities before invasion thus providing broader resistance (Peng *et al.*, 2015).

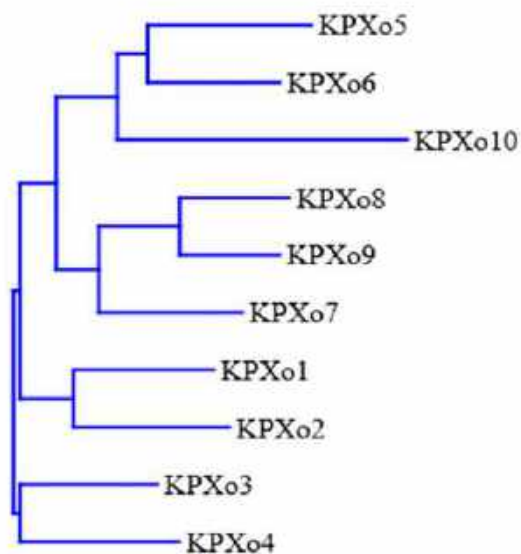


Fig. 5: Dendrogram showing the pathogenic relatedness of *Xoo* isolates collected from Karnataka

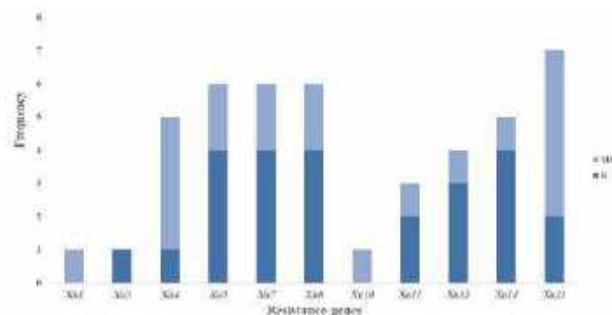


Fig. 6 : Effectiveness of *Xa* genes against Indian *Xoo* isolates. The x-axis indicates resistance gene and the y-axis indicates the frequency of isolates against which this gene confers resistance

### Variation in the Production of Exopolysaccharide (EPS) by *Xoo* Isolates

The production of exopolysaccharide (EPS) is a characteristic feature of *Xanthomonads*. A biochemical assay was carried out to assess the EPS accumulation in different isolates. Isolate *Xoo* 3 produced a distinctly high amount of EPS in this study (92.3 mg) followed by *Xoo* 6, *Xoo* 8 and *Xoo* 4. On the contrary isolates *Xoo* 2 produced distinctly less amount of EPS. The isolates *Xoo* 1, *Xoo* 5 and *Xoo* 7 gave 35-55 mg dry weight of EPS (Fig. 7). To assess the severity of disease infection by different isolates, the susceptible cultivar TN1 was challenged with *Xoo*. All tested isolates caused leaf blight on leaf surface on the 6<sup>th</sup> day after inoculation. Symptoms of BB appeared on leaves as pale green to grey-green water-soaked streaks near the leaf tip and margin. These lesions coalesced and became yellowish-white with wavy edges. On the leaf sheath of susceptible cultivars, the affected leaves will turn yellow, roll up and wilt rapidly. Under greenhouse inoculation, the

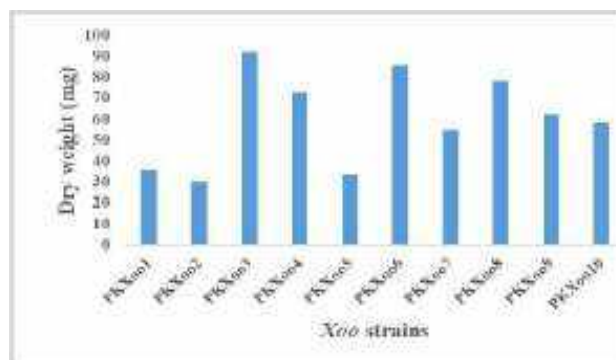
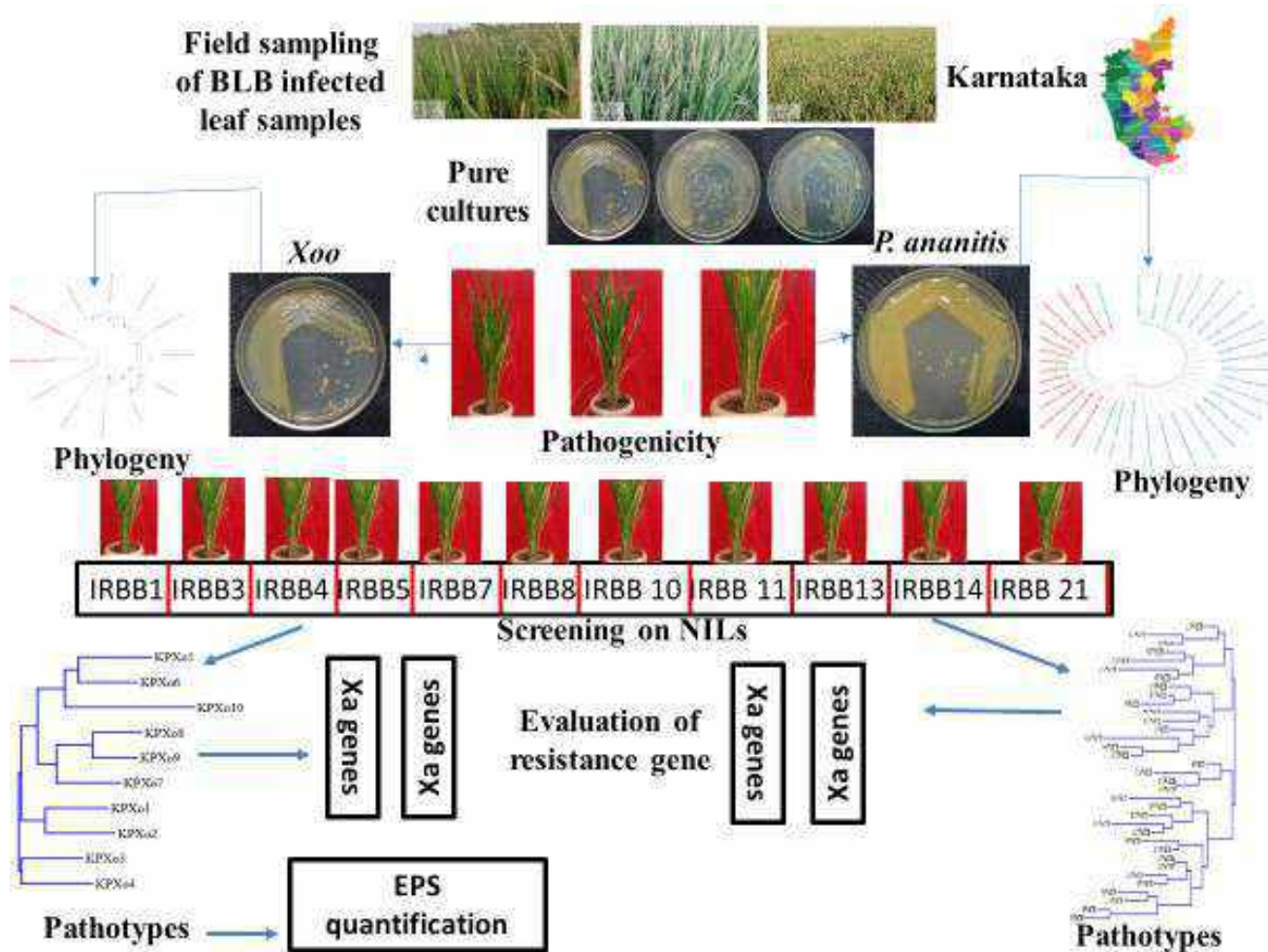


Fig. 7: EPS dry weights of ten Indian *Xoo* isolates

systemic infection produced tannish grey to white lesions along the vein. Differences in virulence among the *Xoo* isolates were quantified according to the lesion length of the necrotic area. *Xoo* 3, *Xoo* 6, *Xoo* 8, *Xoo* 4 and isolates were shown to cause symptoms on day 5 after inoculation while *Xoo* 7, *Xoo* 9 and *Xoo* 10 isolates were shown symptoms on 6 dpi followed by *Xoo* 1, *Xoo* 2, *Xoo* 5 on 7 dpi. It also showed the maximum disease development and increased virulence to susceptible rice cultivar TN-1 at 21 d post inoculation (Fig. 8). The average lesion length was 10-15 cm at 21 d post inoculation. Isolate *Xoo* 2 showed the lowest lesion length and *Xoo* 3 was the highest lesion length at day 21 post inoculation. The isolate *Xoo* 3 revealed a high yield of EPS which



Fig. 8: Leaf phenotype of TN-1 showing increased susceptibility to pathogen isolates, lesion length count in cm up to 21 dpi, leaves were photographed on 21 days after inoculation



Schematic representation of sampling of infected BLB leaf samples, screening on nearly isogenic lines, grouping of isolates in to pathotypes and EPS quantification

is further evident by the higher virulence of *Xoo* that shows the aggressiveness in the disease severity index. On contrary with *Xoo 2* revealed a low yield of EPS with less aggressiveness in the disease index (Bakade and Kumar, 2020). The screening of isolates for virulence-related genes of *Xoo* needs further evaluation toward revealing the complex and overlapped bacterial pathogenesis mechanisms, especially in the early stage of infection.

In conclusion, while the most commonly reported causal agents for leaf blight disease in rice are still the various *Xoo* strains, the involvement of *Pantoea* spp. in also causing the disease cannot be ignored. The various strains of *Xoo* have been extensively studied and that will help to incorporate genes conferring resistance to *Xoo* associated leaf blight. The recent findings of the association of *Pantoea* spp. as a pathogen in causing leaf blight create new challenges in combating this disease (Doni *et al.*, 2021). Although the full impacts of this new pathogenic species are yet to be determined, a thorough investigation of the role of *Pantoea* spp. such as the range of symptoms, mode of infection, infestation, pathogenicity, genetics and evolutionary shift from *Xoo*, will be crucial for ensuring a successful control and management of *Pantoea* spp. associated leaf blight.

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## Biology of Field Bean Pod Borer, *Adisura atkinsoni* (Moore) (Lepidoptera : Noctuidae) on Dolichos Bean under Laboratory Condition

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### ABSTRACT

The studies on the biology of field bean pod borer, *Adisura atkinsoni* (Moore) on field bean pods was carried out in the Department of Entomology, College of Sericulture, Chintamani under laboratory condition during 2021-22. The mean pre-oviposition, oviposition and post-oviposition period on field bean pod lasted for  $2.2 \pm 0.78$ ,  $6.4 \pm 3.02$  and  $2.2 \pm 1.22$  days, respectively. The females laid on an average of  $343.9 \pm 232.4$  eggs in her life time. The incubation period and hatching percentage were observed to be  $3.6 \pm 0.51$  days and  $94 \pm 6.99$ , respectively. The mean length and breadth of eggs was reported about  $0.609 \pm 0.03$  mm and  $0.405 \pm 0.01$  mm, respectively. The mean total larval duration observed to be  $14.4 \pm 1.42$  days. The fully grown larva measured  $1.83 \pm 0.03$  mm &  $26.34 \pm 1.51$  mm with respect to head width and body length. The pupal period lasts for  $15.3 \pm 1.25$  days. The male pupa measures about  $12.22 \pm 0.12$  mm and  $4.09 \pm 0.064$  mm with respect to length and breadth. Whereas, the mean length and breadth of female pupae observed to be  $13.24 \pm 0.34$  mm and  $4.35 \pm 0.08$  mm, respectively. The male moths measured  $11.8 \pm 0.2$  and  $2.59 \pm 0.11$  mm with respect to body length and width. The average body length and breadth of female moth was  $12.8 \pm 0.14$  and  $2.78 \pm 0.13$  mm, respectively. Whereas, the wing expansion with respect to male and female moth reported was  $26.7 \pm 0.388$  mm and  $27.74 \pm 0.50$  mm. The sex ratio of male and female was 1:1.4.

Keywords : *Adisura atkinsoni*, Field bean, Morphometrics, Sex ratio, Life cycle

THE field bean (*Dolichos lablab* L.) is an important pulse-cum-vegetable crop in India popularly known as hyacinth bean, dolichos bean, country bean, butter bean and poor-man's bean. Field bean is cultivated for its tender and matured pods, seeds and also as fodder. The foliage of the crop provides hay, silage and green manure. The crop is cultivated in dry tropical parts of Asia, Africa, East and West Indies, South Central America and China. In India, it is being cultivated in Karnataka, Tamil Nadu, Andhra Pradesh, Kerala and Assam. In Karnataka, *Dolichos* bean is cultivated in 0.77 lakh hectares with an annual production of 0.17 lakh tonnes with a productivity rate of 183 kg/ha (Anonymous, 2019). Though the crop is cultivated in almost all regions of

Karnataka, it is largely grown as a mixed crop with finger millet and sorghum mainly in many parts of Karnataka. However, it is also grown as a pure crop under rain fed as well as irrigated conditions.

On local cultivars, more than ten species of pod borers embracing Lepidoptera, Coleoptera and Diptera are recorded. Among the several pests infesting this crop, pod borer complex comprising of *Helicoverpa armigera* (Hubner), *Adisura atkinsoni* (Moore), *Maruca vitrata* (Geyer) and *Exelastis atomosa* (Walshingham) found to be major cause for the severe yield loss (Rashmi *et al.*, 2019).

After 1990's, the plant breeders were successful in evolving photo-insensitive varieties, like the *Lablab*

plants which bloom throughout the year and this caused the change in the seasonal occurrence, relative abundance and composition of species of pod borers on *Lablab* cultivars. Interestingly, species of pod borers like webworm, *Maruca* and polyphagous pod borer, *Helicoverpa* increased in abundance and their occurrence extended to the entire year. Hence, wherever photo insensitive cultivars and varieties are being sown there the seasonal occurrence and relative abundance of *Adisura* is declining (Chakravarthy and Rajendraprasad, 2016).

The pod borer, *A. atkinsoni* is a dominant and specific insect pest of field bean occurring from August to March under field conditions which coincides with flowering and pod formation stage of the crop. It was found feeding only on the flowers and pods of field bean.

*Adisura* is a specific, locally adapted, economically important pod borer on *Lablab* beans. The life cycle of the pod borer appears to have co-evolved with the life cycle of the plant. Understanding the biology of the pest in the crop will help to identify its most damaging stages as well as the particular time to carry management practices against the pest, which will be beneficial for strategizing the management options of that particular pest. Hence, present investigation has been carried out to study the biology of field bean pod borer on field bean pods under laboratory conditions.

## MATERIAL AND METHODS

The present study was carried out at College of Sericulture, Chintamani during 2021-22. The detailed biology was studied in Insect rearing laboratory, Department of Entomology, College of Sericulture, Chintamani.

### Collection and Rearing of Field Bean Pod Borer *Adisura Atkinsini*

The initial inoculum culture of insect was collected from field bean (HA4 variety) crop. Collected larvae were reared on field bean pods in the laboratory. The temperature of  $25 \pm 2$  °C and relative humidity of  $69 \pm 5\%$  was maintained during the study. The pupa

obtained during rearing were sex separated and kept separately in plastic containers for adult emergence.

### Mating and Oviposition

The adults emerged within 12 to 15 days after pupation. The newly emerged adults (males and females) were collected and about 10 pairs were allowed for pairing in the ratio of 1:1. Each pair provided with individual mating cages with a cotton swab dipped in 1:1 solution (honey: water) for adult feeding. A fresh inflorescence of field bean was arranged for egg laying (Plate 1). The inflorescence was maintained in water filled container to maintain the turgidity. Inflorescence was changed daily and examined for the eggs/ egg masses. The collected eggs/ egg masses were kept separately in plastic containers. The black head stage eggs were provided with fresh young field bean pods and were changed regularly at an interval of one- two days throughout the study.

Observations on incubation period, larval instars and duration, pupal duration, adult emergence, adult longevity, fecundity and hatching percentage were recorded. Larval instars were determined based on exuviae.

### Morphometric Study

Starting from the egg stage, *A. atkinsoni* undergoes four moultings *i.e.*, five larval instars followed by pupation and adult stage. Changes in length and breadth during different developmental stages of larva were noticed. Diameter of eggs, instar wise larval measurements, pupal and adult measurements were recorded by using computerized micrometer (Leica).

## RESULTS AND DISCUSSION

### Pre-oviposition, Oviposition and Post-oviposition Period

The observation on pre-oviposition, oviposition and post-oviposition period revealed that, the pre oviposition period varied from 1 to 3 days with an average of  $2.2 \pm 0.78$  days. The oviposition period ranged between 1 to 10 days with an average of  $6.4 \pm 3.02$  days. The post-oviposition period ranged between 1 to 4 days with an average of  $2.2 \pm 1.22$  days (Table 1). These observations are almost close

TABLE 1

Pre-oviposition, oviposition, post-oviposition period and fecundity of *A. atkinsoni* on field bean pod.

Observation	Minimum	Maximum	Mean±SD
Pre-oviposition period (days)	1	3	2.2 ± 0.78
Oviposition period (days)	1	10	6.4 ± 3.02
Post-oviposition period (days)	1	4	2.2 ± 1.22
Fecundity	7	648	343.9 ± 232.4



Plate 1: Mating and oviposition cage

to the findings of Govindan (1974), who reported that the pre-oviposition, oviposition and post-oviposition was 2 to 3 days, 7 to 10 days and 1 to 2 days, respectively.

### Fecundity

The observations on fecundity of *A. atkinsoni* revealed that, the female moth laid an average of  $343 \pm 232.4$  eggs, ranging from 7 to 648 eggs (Table 1). The observations are in accordance with Govindan (1974), who reported female laid eggs ranging from 145 to 364 with an average of 272 eggs on field bean pod. Sunil and Mohan observed that *Phthorimaea absoluta* laid 125.5 eggs per day on tomato in captivity. The present findings are on corroboration with these reports.

### Eggs

The freshly laid eggs were creamy whitish in color and spherical or ovoid in shape with flat base. The color of eggs changed gradually from yellowish to brownish towards hatching with the mean hatching percentage was  $94 \pm 6.99$ , which was ranging from

80 to 100 per cent. The incubation period occupied between 3 to 4 days with an average of  $3.6 \pm 0.51$  days under laboratory conditions (table 2). The present findings are in conformity with observations of Govindan (1974) and Ramachandra rao (1918), who recorded average incubation period of *A. atkinsoni* on field bean pod was 3.5 and 3 days, respectively.

The morphometrics of egg stage was recorded. The mean length of eggs were  $0.60 \pm 0.032$ mm and ranging from 0.56 to 0.68mm. Whereas, the mean breadth of eggs  $0.40 \pm 0.10$ mm which ranges from 0.39 to 0.42 mm as shown in table 3. According to Govindan (1974), egg measures 0.46 mm in height and 0.49 mm in diameter, these observations are varying with present observations may be due to nutrient contents.

### Larva

The development period of larva ranged from 12 to 18 days with an average of  $14.4 \pm 1.42$  days during this period, larvae moulted 4 times with 5 instars (Table 2). According to Ramachandra rao (1918) and Krishnamurti and Appanna (1948), the larval period of *A. atkinsoni* occupied from 13 to 15 days and 17 to 18 days, respectively with five instars. The larval duration of *A. atkinsoni* in the present study was coinciding with the results of Nunes *et al.* (2017), who reported a *Helicoverpa armigera* mean larval duration of 14.54 days.

The mean duration of 1<sup>st</sup> larval instar was  $3.3 \pm 0.48$ , which ranges from 3 to 4 days. The mean 2<sup>nd</sup> larval instar duration was  $2.4 \pm 0.51$  days, which was ranging from 2 to 3 days. After 2<sup>nd</sup> instar the three remaining instars (III, IV and V) were look similar except size. The mean 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> instar larval duration was  $2.2 \pm 0.42$  days,  $2.3 \pm 0.48$  days and

TABLE 2  
Duration (days) of different developmental stages of *A. atkinsoni* on field bean pod

Growth stage	No. of individuals observed	Period (in days)		
		Minimum	Maximum	Mean±SD
Egg (incubation period)	10	3	4	3.6 ± 0.51
<i>Larval duration</i>				
1 <sup>st</sup> instar	10	3	4	3.3 ± 0.48
2 <sup>nd</sup> instar	10	2	3	2.4 ± 0.51
3 <sup>rd</sup> instar	10	2	3	2.2 ± 0.42
4 <sup>th</sup> instar	10	2	3	2.3 ± 0.48
5 <sup>th</sup> instar	10	3	5	4.2 ± 0.63
Total larval duration		12	18	14.4 ± 1.42
<i>Pupal duration</i>				
Pre-pupa	10	2	3	2.3 ± 0.48
Pupa	10	12	14	12.8 ± 0.78
Total pupal duration		14	17	15.3 ± 1.25

4.2 ± 0.63 days, which were ranging from 2 to 3 days, 2 to 3 days and 3 to 5 days, respectively (table 2). The present findings are matching with the recordings of Govindan (1974), who reported the mean 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> larval instar durations was 2.6 days, 2.8 days, 2.75 days, 3.8 days and 4.7 days and ranged from 2 to 3 days, 2.5 to 3 days, 2 to 3 days, 3 to 4 days and 4 to 5 days, respectively.

The larval morphometrics of *A. atkinsoni* recorded from first instar to fifth instar, which was reared on field bean pod and the larval size increased with each moulting (Plate 2).

The morphometrics of different larval stages was given in table 3. The mean head width of 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> larval instar was 0.299 ± 0.02 mm, 0.69 ± 0.014 mm, 1.137 ± 0.02 mm, 1.46 ± 0.03 mm and 1.83 ± 0.03 mm, respectively. Which, ranges from 0.27 to 0.35 mm, 0.68 to 0.71 mm, 1.1 to .19 mm, 1.41 to 1.5 mm and 1.75 to 1.86 mm with respect to 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> larval instars. Govindan (1974) showed the mean head width was 0.5 mm, 0.75 mm, 1.0 mm, 1.25 mm and 1.9 mm with respect to 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> instars, while except for first instar the observations were in accordance with present study observations.



Plate 2: Change in larval size and colour from first instar to fifth instar

The mean body length of 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> larval instars was 2.42 ± 0.23 mm, 10.02 ± 0.52 mm, 12.37 ± 0.42 mm, 18.65 ± 0.58 mm and 26.34 ± 1.51 mm, respectively. The body length of 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> larval instars was ranging from 2.1 to 2.7 mm, 9.3 to 11 mm, 11.9 to 13 mm, 18 to 19.5 mm and 24 to 28 mm, respectively (table 3). The host nutrition and temperature could be the reason for variation in larval body size. The body length of different larval stages of *A. atkinsoni* on field bean showed by Govindan (1974) was ranging from 2.0 to 2.5mm, 4.5 to 5.5mm, 7 to 10mm, 14 to 16mm and 21.5 to 23.0mm with respect to 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup>, 4<sup>th</sup> and 5<sup>th</sup> larval instars and the present findings are in accordance with these results.

## Pupa

The fully grown caterpillar stop feeding wandered for some time and entered into the soil. It prepares an oval earthen cell within which the caterpillar become inactive and body shrunk. The skin ruptured along the mid-dorsal line and pupation occurred. The caterpillar was also found to pupate inside the pod even in absence of the soil. The pre-pupal duration ranged from 2 to 3 days with an average of  $2.3 \pm 0.48$  days (table 2). Mahakunda and Tiwari (2020) also reported pre-pupal period from 2 to 3 days in *M. vitrata* on pigeon pea and the present results are in agreement with the above reports.

The freshly formed pupa is light-green in color and soft and later become brown and hard (Plate 2). The mean pupal period of *A. atkinsoni* was  $12.8 \pm 0.78$  days which ranged from 12 to 14 days (table 2). The present results are in contradiction with the reports of Govindan (1974), who recorded pupal period of *A. atkinsini* varied from 10 to 18 days on field bean

under laboratory condition and variation of pupal period may be depending on light hours.

The mean length of male pupa was  $12.22 \pm 0.12$  mm and ranged from 11.95 to 12.29 mm in which, the width ranged from 4.01 to 4.19 mm with an average of  $4.09 \pm 0.06$  mm. The female pupa length ranged from 12.6 to 13.6 mm with an average of  $13.24 \pm 0.34$  mm whereas; the mean width of female pupa was  $4.35 \pm 0.08$  mm, which ranged from 4.1 to 4.4 mm (Table 3).



Plate 3: Colour changes observed in pupa of *A. atkinsoni*

TABLE 3  
Morphometrics of different stages of *A. atkinsoni* on field bean pod

Growth stage	Parameter(mm)	No. of individuals observed	Minimum(mm)	Maximum(mm)	Mean± SD
Egg	Length	10	0.56	0.68	$0.609 \pm 0.03$
	Breadth		0.39	0.42	$0.405 \pm 0.01$
<i>Larval stages</i>					
1 <sup>st</sup> instar	Head width	10	0.27	0.35	$0.299 \pm 0.02$
	Body length		2.1	2.7	$2.42 \pm 0.23$
2 <sup>nd</sup> instar	Head width	10	0.68	0.71	$0.69 \pm 0.01$
	Body length		9.3	11	$10.02 \pm 0.52$
3 <sup>rd</sup> instar	Head width	10	1.1	1.19	$1.137 \pm 0.02$
	Body length		11.9	13	$12.37 \pm 0.42$
4 <sup>th</sup> instar	Head width	10	1.41	1.5	$1.46 \pm 0.03$
	Body length		18	19.5	$18.65 \pm 0.58$
5 <sup>th</sup> instar	Head width	10	1.75	1.86	$1.83 \pm 0.03$
	Body length		24	28	$26.34 \pm 1.51$
<i>Pupal stages</i>					
Male	Length	10	11.95	12.29	$12.22 \pm 0.12$
	Width		4.01	4.19	$4.09 \pm 0.06$
Female	Length	10	12.6	13.6	$13.24 \pm 0.3$
	Width		4.1	4.4	$4.35 \pm 0.08$

TABLE 4  
Morphometrics of adult moths of *A. atkinsoni*

Adult	Parameter	Minimum (mm)	Maximum (mm)	Mean± SD
Male	Body length	11.5	12.1	11.8 ± 0.2
	Body width	2.4	2.8	2.59 ± 0.11
	Wing expansion	26.1	27.1	26.7 ± 0.38
Female	Body length	12.6	13	12.8 ± 0.14
	Body width	2.6	3	2.78 ± 0.13
	Wing expansion	27.1	28.5	27.74 ± 0.50

### Adult

The body length of male and female was ranged from 11.5 to 12.1 mm and 12.6 to 13 mm with an average of  $11.8 \pm 0.2$  mm and  $12.8 \pm 0.14$  mm, respectively. The mean body width of male and female moths was  $2.59 \pm 0.11$  mm and  $2.78 \pm 0.13$  mm, which ranged from 2.4 to 2.8 mm and 2.6 to 3 mm, respectively. The wing expanse of male and female moth ranged from 26.1 to 27.1 mm and 27.1 to 28.5 mm with an average of  $26.7 \pm 0.38$  mm and  $27.74 \pm 0.50$  mm, respectively (Table 4).

### Sex Ratio

The male and female moths did not show any distinct sexual dimorphism (Plate 4). Whereas in dry preserved male specimen, the tip of abdominal genital valvae are seen extruded, but in case of female, the abdomen is somewhat stout and the tip is pointed. The sex ratio of laboratory reared culture was found to be 1:1.4 (male: female). Out of 100 adults examined, 41 observed to be males and 59 were females. Govindan

(1974) observed sex ratio of lab reared culture was found to be 5:4 (male: female) out of 10 specimens.

The present study indicated the biology of *A. atkinsoni* on field bean under laboratory conditions. Since, *A. atkinsoni* is the major pest of field bean crop having enough potential to cause severe damage to the crop and yield. The larval size increased in each instar. The mean larval duration of 5<sup>th</sup> instar is more compared to other instars. The pupal and adult female body is increased compared to male body. Differences in crop phenology and agroclimatic conditions may influence the biology and life cycle of *A. atkinsoni*.

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Plate 4: Male and Female moths of *A. atkinsoni*

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## Growth of Maize Ecosystem in India and Karnataka Vis-a-Vis Associated Risk in Production : An Economic Insight

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### ABSTRACT

Maize is one of the most versatile emerging crops having wider adaptability under varied agro-climatic conditions. Globally, maize is known as queen of cereals because of its highest genetic yield potential among the cereals. In addition to staple food for human being and quality feed for animals, maize serves as a basic raw material as an ingredient to thousands of industrial products. Growing area under maize and increasing MSP for maize will reflect importance of maize and its multiple benefits. In this study, the growth in area, production and productivity of maize in India and Karnataka and risks associated with maize production in Karnataka were analyzed. The primary data pertaining to study was collected from major maize growing districts of Karnataka and secondary data on area, production and productivity of India and Karnataka was collected from Indiastat.com and the Directorate of Economics and Statistics, Government of Karnataka. Significant growth rates in maize area (1.12), production (3.51) and productivity (2.36) were recorded in India as a whole for the period 1970-2019. The share of maize in total food grain production has increased from 3.40 per cent in 1950-51 to 9.98 per cent in 2020-21. The cumulative annual growth rate of maize area (4.80), production (6.08) and productivity (1.52) has showed positive significant scores in Karnataka. Water scarcity, non-availability of inputs at right time and unstable yield were major risks associated with maize production before the infestation of fall armyworm, but pest and disease, unwanted moisture in the field and low quality of fodder were major risks associated with maize production after the introduction of fall armyworm in the study area.

*Keywords* : Area, Production & productivity of maize, Growth rate, Production risk, Fall armyworm

MAIZE (*Zea mays* L) being one of the versatile emerging crop with wider adaptability under different agro-climatic environmental conditions. Because of its high level of genetic yield potential among all cereals, maize is known as 'Queen of Cereal's globally (Manjanagouda and Kalyanamurthy, 2018). Maize is a vital crop for millions of people in the form of food, fodder, feed and industrial raw material. Globally, around 1147.7 million metric tonnes of maize is produced from 193.7 million hectare with an average yield of 5.75 tonnes per hectare in 170 countries (Meena and Nirupma, 2021) with a diverse range of soil, climate, biodiversity and

management approaches, accounting for 36 per cent of world grain production.

Maize is widely used for a many purposes around the world, including feed 61 per cent, food 17 per cent and industry 22 per cent. China leads the globe in maize area under cultivation, followed by the United States, which together account for 39 per cent of global maize area. Since 2005, India has ranked fourth in terms of area and seventh in terms of production, accounting for about 4 per cent of global maize area and 2 per cent of overall production. In India, the maize acreage grew to 9.2 million hectares in



2018-19 (Meena and Nirupma, 2021). During 1950-51 India produced 1.73 million tonnes maize, which has increased to 27.8 million tonnes by 2018-19, recording close to 16 times increase in production. During this period, average productivity surged by 5.42 times, from 547 kg/ha to 2965 kg/ha, meanwhile the area under maize cultivation nearly tripled to 9.2 million hectares. The United States produces 34 per cent of the world's maize, followed by China 22 per cent. Since 1961, India has ranked top ten among the maize producers in the world, with an annual production of roughly 28 million tonnes. In India, maize productivity is slightly higher than 3 t/ha, which is slightly higher than half of the global average (5.6 t/ha).

After rice and wheat, maize is India's third most significant food crop, having the largest output potential among cereals. Since mid-1980s there is a distinct shift in maize cultivation, when larger area under maize shifted to peninsular India. Currently peninsular India represents over 40 per cent of maize area and 50 per cent total maize production. Karnataka (1.3 mha), Madhya Pradesh (1.3 mha), Maharashtra (1.0 mha), Telangana and Andhra Pradesh (0.9 mha), Rajasthan (0.8 mha) are the principal maize growing states of the country.

Currently 47 per cent of maize produced in India is consumed in feed industry, while 13 per cent as animal feed. Starch industry consumes around 14 per cent of maize and other industries use maize as a primary raw material, including starch, oil, protein, alcoholic drinks, food sweeteners, pharmaceutical, cosmetic, film, textile, gum, package and paper industries *etc.* Over the decade use of maize as direct food has reduced considerably, now pegs at around 13 per cent. However, there is an increasing trend to use maize as processed food, which contributes to around 7 per cent of annual maize consumption in the country. Use of specialty corns, *viz.*, sweet corn, baby corn and popcorn is a recent dimension where maize cultivation is getting integrated with rural entrepreneurship and agro-business. With all of these benefits, maize is the best crop for accomplishing the government's goal of doubling farmer's income.

In Karnataka, maize is grown over an area of approximately 1.3 mha producing 26.4 lakh tonnes. Particularly in Central part of Karnataka *viz.*, Shimoga (0.585 lakh ha), Davangere (1.74 lakh ha) and Chitradurga (0.67 lakh ha) districts. In the past ten years maize has registered tremendous increase in area compared to any other crops and has replaced other rainfed area crops in the state like potato, tobacco, cotton, groundnut, ragi and sorghum.

Due to drastic expansion of area under maize and its increased cultivation, risk components have also increased. Since the majority of maize is rainfall dependent and the production is unstable due to lack of irrigation facilities is one among the major limiting factors and along with other production risks already present in the production of maize, the fall armyworm's introduction in 2018 started having a significant influence on maize output.

#### MATERIAL AND METHODS

Karnataka is the major maize producing state in the country and has registered a positive significant growth rates in last two decades overtaking other states and becoming number one in terms of area and production. Based on area, production, productivity and fall armyworm incidence on maize, four major maize growing districts *viz.*, Davanagere, Haveri, Hassan and Chikkaballapur were selected for the study. The ultimate sample of farmers numbering 50 from each district was chosen randomly from the cluster of villages to form overall sample size of 200 maize farmers.

A structured schedule was prepared and pretested before it was administered to the respondent farmers. The schedule covered general information on maize farmers, their asset position and details of maize crop production in terms of input usage, costs, income, production risk associated and damage caused by fall armyworm *etc.* For assessing the production risk associated, yield loss. Secondary data on area, production and productivity of India as a whole were collected from Indiastat.com for the period 1970-2019 and for Karnataka State Secondary data on area, production and productivity and rainfall as a whole

were collected from Directorate of Economics and Statistics, Govt. of Karnataka, Bengaluru for the period 2000-2020.

### Analytical Tools Used

#### Exponential Growth Model

Growth rates for area, production and productivity of maize in India and Karnataka were computed for a period of 50 years from 1970 to 2019 and 2000 to 2020 for India and Karnataka, respectively. Several functional forms were used to estimate the growth rates of the selected economic variables. Finally, exponential growth model was selected for the analysis and the model is of the following form.

$$Y = ab^t e \dots (1)$$

Where

Y = Dependent variable for which the growth rate is estimated (area, production, productivity of maize).

a = Intercept

b = Regression coefficient

t = Time variable (1970 to 2019) for area, production, productivity for India) (2000 to 2020 for Karnataka)

e = Error term

The compound growth rate was obtained from the logarithmic form of the equation (1) as below.

$$\ln Y = \ln a + t \ln b$$

The per cent compound growth rate (g) was derived using the relationship

$$g = (\text{Anti ln of } b - 1) \times 100$$

#### Instability Analysis

The coefficient of variation was used as a measure to study the variability in area, production, productivity and input use in rainfed maize in Karnataka. The coefficient of variation (CV) or index of instability was computed using the following formula

$$CV = \frac{\text{Standard Deviation}}{\text{Mean}} \times 100 \dots (2)$$

Linear trend was fitted to the original time series data, for a period of 50 years from 1970 to 2019 for India and 2000 to 2020 for Karnataka. The trend coefficients were tested for their significance. Whenever the trend of series was found to be significant, the variation around the trend rather than the variation around mean was used as an index of instability. The formula suggested used to compute the degree of variation around the trend, mean, coefficient of variation was multiplied by the square root of the difference between the unity and coefficient of multiple determination ( $R^2$ ) in the cases where  $R^2$  was significant to obtain the Instability Index.

$$\text{Instability Index} = \frac{\text{Standard Deviation}}{\text{Mean}} \times 100 \times \sqrt{(1 - R^2)} \dots (3)$$

$R^2$  = Coefficient of Determination

A high degree of instability index signifies violent variations.

#### Garret's Ranking Technique

An attempt was made to recognize the problems faced by the growers in the cultivation of Maize. The identified problems of growers in the cultivation of maize were ranked by making use of Garrett's Ranking Technique. The technique was used to rank the preference mentioned by the respondents on different factors and aspects of the cultivation process. It is used to find the most significant factor which had influenced the respondent in their practices. Founded on the Garret's Ranking technique, the study had the respondents rank different problems and outcome based on their impact thereby converting into score value and rank with the help of the following formula:

$$\text{Percent position} = \frac{100 (R_{ij} - 0.5)}{R_{ij}}$$

Where,

$R_{ij}$  = Rank given for the  $i^{\text{th}}$  variable by  $j^{\text{th}}$  respondents

$N_j$  = Number of variable ranked by  $j^{\text{th}}$  respondents

With the help of Garrett's table, the per cent position estimated is converted into scores by referring to the table. Then for each factor, the scores of each individual were added and then total value of scores and mean values of score was calculated. The factors having highest mean value was considered to be the most important factor.

## RESULTS AND DISCUSSION

### Growth Rates in Area, Production, Productivity of Maize

Compound growth rates were computed to comprehend the trends in area planted, production and productivity in maize cultivation. The study period was from 1970 to 2019 and (sub divided into Period-I (1970-1979), Period-II (1980-1989),

Period-III (1990-1999), Period-IV (2000-2009) and Period-V (2010-2019)) and 2000 to 2020 for India and Karnataka, respectively and the exponential growth function was employed to find out the growth rates during the above mentioned period.

The results of estimated growth rates are presented in Tables 1 and 3. Negative growth was observed in area (-0.13), production (-0.63) and productivity (-0.50) of maize during Period-I (1970-1979) and found statistically non-significant, because the major traditional maize growing areas like Bihar, Madhya Pradesh, Uttar Pradesh and Rajasthan most farmers use to grow local maize varieties during rainy season, low use of input levels far below than recommended level and seed replacement is very low (Joshi, *et al.*, 2005). During Period-II (1980-1989) again negative growth was observed in area but production and productivity has shown a positive growth rate but found statistically non-significant and medium instability in production was observed during the

TABLE 1

Decade wise temporal variation of area, production and productivity of maize in India

Particulars	Area ( '000 ha)	Production ( '000 t)	Productivity (Kg/ha)
Period-I (1970-1979)			
Mean	5843	6172.9	1055.6
CAGR	-0.13	-0.63	-0.5
p value	0.65	0.66	0.7
CV %	2.34	12.08	11.09
Instability Index	2.45	12.65	11.64
Period-II (1980-1989)			
Mean	5839.3	7451.2	1274.5
CAGR	-0.19	1.87	2.06
p value	0.5	0.3	0.21
CV %	2.28	15.5	14.5
Instability Index	2.34	15.32	13.85
Period -III (1990-1999)			
Mean	6104.3	9928.6	1623.6
CAGR	0.95 ***	3.28 ***	2.32 ***
p value	0.0003	0.001	0.01
CV %	3.14	11.26	8.78
Instability Index	1.38	6.2	5.92

Table 1 Continued.....

Particulars	Area ( '000 ha)	Production ( '000 t)	Productivity (Kg/ha)
Period-IV (2000-2009)			
Mean	7463.7	15072.3	2007.4
CAGR	2.93 ***	5.29 ***	2.28 **
p value	5E-06	0.001	0.05
CV %	8.91	18.32	11
Instability Index	2.38	9.99	9.01
Period-V (2010-2019)			
Mean	9067.8	24787.6	2728.4
CAGR	1.06 ***	3.47 ***	2.39 ***
p value	0.01	0.0002	0.001
CV %	4.11	11.4	8.4
Instability Index	2.75	4.82	4.4

same period. Since, majority of maize area in India is rainfed the production is dependent on good rainfall whenever there is a of lack of rainfall during cropping period leads to instability in production the variance of production of maize was mainly due to factor other than area and productivity (Kiran, *et al.*, 2018) Whereas during Period-III (1990-1999) there was a positive compound annual growth rate of area (0.95), production (3.28) and productivity (2.32), which was statistically significant at one per cent level. This is mainly due to expansion of area and use of hybrid seeds resulting in higher yields in non-traditional maize growing area like Karnataka and Andhra Pradesh. Farmers in these areas grow maize as a commercial crop and there was close linkage between maize production and the poultry sector (Joshi, *et al.*, 2005). The average productivity during the period stood at 1623.6 kg/ha which is 350 kg higher compared to previous period and the coefficient of variation was 8.68 per cent which was lower compared to 14.50 per cent in the previous period.

In the Period-IV (2000-2009) significant growth rates were observed in area, production and productivity of maize however, along with increase in growth rates the instability index was also increased compared to previous period. Further, during Period-V (2010-2019) the positive and significant trend in area, production and productivity of maize

was observed. Also during the same period the instability index indicated improved values in production (4.82) and productivity (4.40) compared to previous period. The average productivity has increased considerably from 2007.40 kg/ha in Period-IV (2000-2009) to 2728.40 kg/ha in Period V (2010-2019) the area expansion is mainly because depletion of groundwater, the farmers were shifting from unprofitable cultivation of rice to maize because the maize crop can be grown using three to four irrigation and wider adoption of high yielding varieties lead increase in productivity as well as production (Yadav *et al.*, 2016).

Growth of 1.12 per cent in area, 3.51 per cent in production and 2.36 per cent in production of maize in India was observed for overall period from 1970 to 2019 and was statistically significant at one per cent level (Table 2). Mean productivity of maize during the period in India stood at 1737.9 kg/ha, whereas medium instability (15.70) was observed for the period 1970 to 2019 in production of maize in India.

Significant positive growth was observed in area production and productivity of maize in Karnataka for the period 2000-2020 (Table 3). The instability in production of maize in Karnataka was 22.26 followed by 17.61 in productivity and 11.73 in area the mean productivity for the period was 2941 kg/ha. A study by Joshi *et al.* (2005) observed that the positive

TABLE 2  
Temporal variation of area, production and productivity of maize in India

Particulars	Area (lakh ha)	Production (lakh t)	Productivity (Kg/ha)
Period – (1970-2019)			
Mean	68.64	126.83	1737.9
CAGR%	1.12***	3.51***	2.36***
p value	0.00000006	0.00000002	0.00000001
CV%	19.16	55.92	35.83
Instability index	8.44	15.70	9.97

TABLE 3  
Decadal temporal variation of area, production and productivity of maize in Karnataka

Particulars	Area (lakh ha)	Production (lakh t)	Productivity (Kg/ha)
Period – (2000-2020)			
Mean	11.38	33.37	2941.00
CAGR%	4.80***	6.08***	1.52**
p value	0.00000001	0.00000002	0.03
CV%	28.15	39.56	19.63
Instability	11.73	22.26	17.61

growth of maize in Karnataka is attributed to adoption of modern varieties, strong seed sector, timely rainfall or proper irrigation and strong demand for maize from the rapidly growing poultry sector. Singha and Naphde (2012) reported that lack of irrigation was one of the key reasons responsible for many farmers switched from rice to maize cultivation.

During 1950-51, the per cent share of maize area to total food grain area in India was at 3.25 per cent and 3.40 per cent in production, whereas during 1990-91 the percent share of maize in total food grain has increased to 4.20 per cent in area and 5.08 per cent in production. Further in 2020-21, the per cent share of maize area to total food grain area in India was increased to 7.62 per cent which was more than double and production 9.98 per cent which was three times higher compared to the year 1950-51 (Table 4).

The rise in the per cent share of area and production of maize to total food grain in India is driven by the area expansion as well as yield improvement by adoption of high yielding varieties, hybrids with relatively high use of inputs and strong demand driven by poultry sector for feed followed by multiplicity uses of maize and implementation of government of India sponsored 'Integrated scheme of oilseeds, pulses, oil palm and maize' (ISOPOM) (Ranjit Kumar *et al.*, 2014). As per Indian institute of maize research 47 per cent of maize is utilized for poultry feed and the remaining produce is utilized for a variety of purposes, comprising 13 per cent for food and livestock feed, 12 per cent for industrial usage, 14 per cent for the starch industry, 7 per cent for processed foods and 6 per cent for export and other uses.

TABLE 4  
Decadal trend in percent share of maize to total food grain production in India

Year	Particulars	Total food grain India	%Share of maize
1950-51	Area	973.21	3.25
	Production	508.30	3.40
1990-91	Area	1404.28	4.20
	Production	1763.90	5.08
2000-01	Area	1210.48	5.46
	Production	1968.10	6.11
2010-11	Area	1266.71	6.75
	Production	2444.90	8.89
2020-21	Area	1297.95	7.62
	Production	3107.40	9.98

### Sources of Production Risk Associated with Maize Production

The sources and extent of production risk faced by sample farmers was assessed by Garret's ranking technique which is presented in Table 5. Results revealed that before introduction of Fall armyworm the major problem in production of maize among sample farmers was drought/scarcity of water/extreme heat with an average score of 136.67 followed by unstable yield (120.25), availability of inputs (119.17), labour scarcity (116.87) and pest and diseases (116.02)

TABLE 5  
Sources of production risk associated with maize production

Sources of Production risk	Before introduction of Fall Armyworm (n=200)		After Introduction of Fall Armyworm (n=200)	
	Average score	Rank	Average score	Rank
Drought/ water scarcity /extreme heat	136.67	1	126.62	2
Unstable yield	120.25	2	118.91	3
Availability of inputs ( Seeds, Fertilizers, Pesticides etc) at right time and quality	117.58	3	116.45	5
Pests and diseases	117.34	4	143.47	1
Labour scarcity	116.87	5	117.28	4
weed infestation	115.66	6	113.85	8
Credit availability at low interest rate	112.96	7	110.89	9
Information access on maize production methods	109.36	8	110.21	10
Low quality of Fodder/Straw	107.91	9	114.88	7
Floods/unwanted moisture in the field	107.79	10	115.14	6

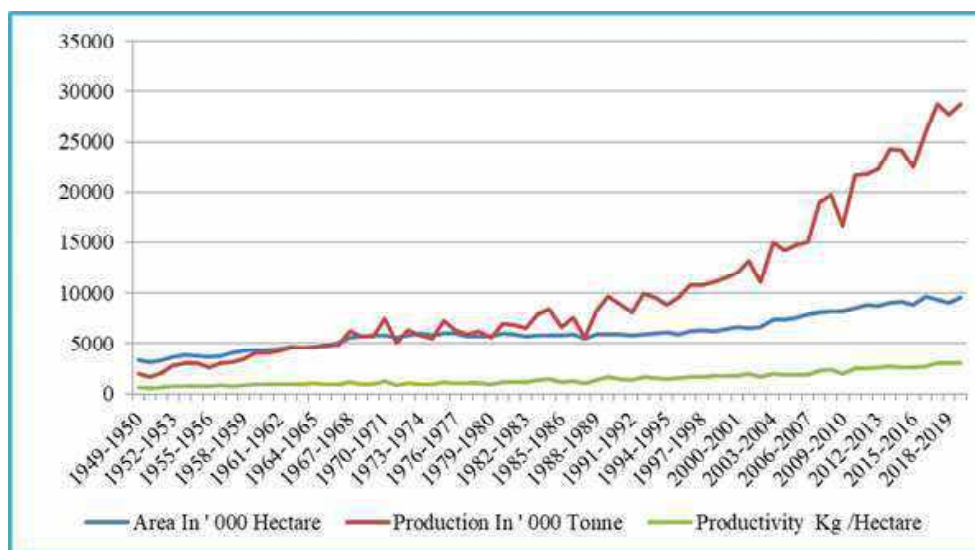


Fig. 1 : Trends in area, production and productivity of maize in India

were the major production risk faced by maize farmers in the study area. Weed infestation and credit availability was also felt as production risk by farmers with average score of 115.66 and 112.96, respectively. The least production risk in maize was associated with floods/unwanted moisture in the field with an average score of 104.64. The situation was changed after introduction of fall army worm in production of maize

with major production problem being faced by farmers was pest and diseases with an average score of 145.98 among 200 sample farmers. Second major production risk as opined by farmers was drought/scarcity of water/extreme heat with average score of 124.35 followed by unstable yield (119.81), labour scarcity (117.28), availability of inputs (116.45). The least production risk in maize faced by sample farmers was

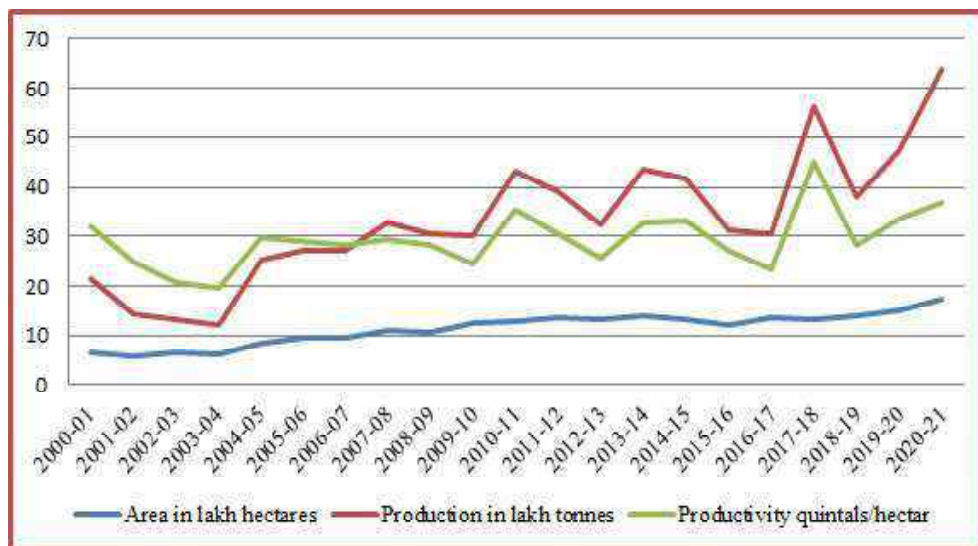


Fig. 2. Trends in area, production and productivity of maize in Karnataka  
Decadal trend in percent share of maize to total food grain production in India

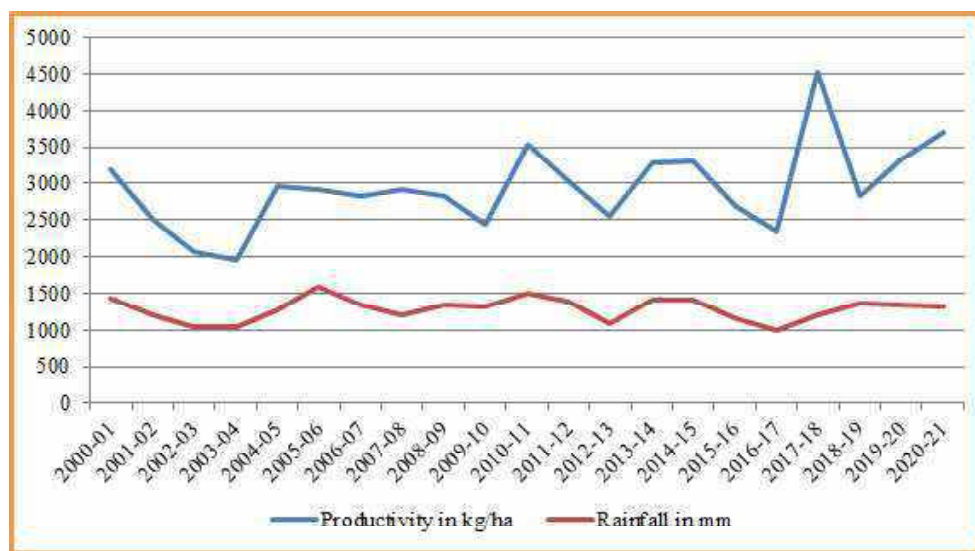


Fig. 3. Trends in rainfall and productivity of maize in Karnataka

information access to maize production methods with an average score of 110.21.

Drought was the major production risk faced by farmers before introduction of fall army worm in maize indicating importance of rainfall as it is apparent from the Fig. 1 that the productivity of maize is in synchronizing trend with the rainfall pattern of Karnataka shows the high dependency of maize production to rainfall hence, farmers opinion

regarding same is correct with regard to drought and unstable yield. Availability of inputs at right time was also a major problem faced by farmers as this leads to low input use and low yield as opined by the farmers. Labour scarcity was given fourth rank in the production risks faced by maize farmers, due to availability of jobs in other sectors with higher wage rates labour problem is faced by all farmers in rural areas irrespective of crops. In general, it was considered that, maize is less risky crop as pest and

disease incidents are minimal but the introduction of fall armyworm in 2018 has changed the scenario with major problem being pest and diseases in the production of maize among sample farmers. Fall armyworm is causing devastating yield losses ranging from 22 to 67 per cent in Africa since its introduction in 2016 (Balla *et al.*, 2019) and involves high management cost. Results are in line with the study conducted by Kathy *et al.* (2021) wherein they reported that they spent US\$600 million in 2009 for controlling FAW. The average management cost of fall army worm was estimated to be \$40/ha.

During past three decades maize production in India and Karnataka has increased significantly adding new regions and seasons with growing demand from the poultry, animal feed, starch and ethanol industries facilitated by government policies and adoption of single cross hybrids and high yielding varieties and switching from other crops to maize due to its high yield potential and low water requirement and having multiple uses has made the crop special from other cereals. Due to drastic expansion of area under maize and its increased cultivation, risk components have also increased. Since the majority of Indian maize is rainfall dependent and the production is unstable due to lack of irrigation facilities this is one among the major limiting factors and the incidence of fall armyworm started making serious impact on maize output further, reducing the national average yield of maize to less than 3.1 tonnes per hectare which is low as compared to other Asian countries This. calls for change in strategic approach and attention for planning and devising adaptation and mitigation strategies for future pest management programmes like framing stringent policies for control of invasive pest and diseases into country for sustainable production.

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## Development of a Scale to Analyse the Environmental Sustainability through Urban Gardens

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### ABSTRACT

Research was conducted to understand how gardeners perceive environmental sustainability for which there is a need to measure using a psycho metric scale. Though several scales available were intended to measure the other forms of sustainability *i.e.* economic, agriculture, social *etc.* whereas environmental sustainability per se was not available. So, the study was taken to develop a scale to measure environmental sustainability through urban nutri-gardens in Bangalore district. Likert summated scaling technique was employed. Based on operational definition of construct - environmental sustainability through urban nutri-gardens responses for 70 items belongs to 6 domains were obtained from 56 judges. Based on relevancy test, 59 items were retained. Responses for these 59 items were obtained from 32 urban gardeners of non-sample area. Criterion group performed to evaluate individual items and critical ratio was calculated using t-test. Forty eight statements with highest 't' values equal to or greater than 2.45 were selected and subjected to reliability in non-sample area consisting of 32 urban gardeners. Correlation coefficient was 0.9210 and 'r' value after adjusting for scale was 0.9592 which was significant at 1:00 per cent level indicating the high reliability of the instrument. Validity, which was found to be 0.974 for scale. Hence, the validity coefficient was also found to be appropriate and suitable for the tool developed. All the components considered were appropriate in measuring the environmental sustainability of the urban gardener's through urban gardens. Final scale composed of 48 statements. The developed scale was administered to 32 gardeners in the non-sample study area as a pilot study and it was found that 40.63 per cent of the gardeners had medium level of environmental sustainability. This specifically developed scale can be used to measure the gardener's environmental sustainability in study area as well as beyond the study area.

**Keywords :** Statements, Urban gardens, Environmental sustainability, Reliability, Validity

ACCORDING to the United Nations prediction that by 2050 about 64 per cent of the developing countries and 86 per cent of the world will be urbanized. In India more than 30 per cent of the population lives in urban areas which is expected to grow further. Urban population growth in Karnataka, especially in Bengaluru is increasing at a faster pace. Bengaluru metropolitan region ranked 24<sup>th</sup> and 4<sup>th</sup> in the world and India, respectively. By 2030, it will be 12<sup>th</sup> most populous urban region in the world. Urbanization is an inevitable consequence of socio-

economic development, but in many countries, it is proceeding at a faster rate that is outpacing the growth of services and employment. Increased urbanization along with exponential growth in population has led to contraction of cultivable farm areas and migration of rural population to urban areas in search of jobs. This increase in population has outstretched the problems of food and nutrition security. It is influencing all phases of food production and consumption. Urban poverty and food scarcity are increasing along with the unemployment rate, beside

air and water pollution most common in urban areas. The land available for agriculture is also getting reduced due to rapid conversion of land into housing, industrial development and highways will lead to the most important environmental challenge faced by human beings. (Suresh and Shivamurthy, 2017).

Urban agriculture contributes greatly to the food security of major cities across the globe. With increase in global population and reduced area under agriculture over years, urban farming is seen as a big solution to traditional agriculture. Urban as well as peri-urban agriculture can help in achieving nutritional security; though conventional agriculture needs to be continued, but urban agriculture can supplement traditional farming. One of the studies noted that one square meter of urban farming is capable of producing 36 heads of lettuce for every 60 days, 10 cabbages for every 90 days and 100 onions for every 120 days. Moreover, urban farming is capable of bolstering more social and political inclusion, sustainability in environment, economic progress and unified water and land policies (Cabannes, 2012 and Nugent, 2000). On the whole, urban farming is a novel initiative which has been encouraged across the urban areas so as to re-create clean, green and sustainable urban areas in the near future. According to FAO report, urban garden lands are 15 times more productive than rural holdings. Urban farming paves way to nutritional security of the population and ensures environmental sustainability.

Sustainable development is a development that meets the needs of the present without compromising the needs of future generations to meet their own needs (Brundtland, 1987). The principles of sustainability are the foundations for three pillars: the economy, society, and the environment. Environmental sustainability is the responsibility to conserve natural resources and protect global ecosystems to support health and wellbeing, now and in the future (Grossarth and Hecht, 2007). It means committing to environmentally sustainable practices to build thriving communities and secure future growth potential. For continuous change in dynamics of urban-rural

interface will increase pressure on environment for its resources, it will lead to exploitation of resources and increase pollution levels in globe, degradation of natural resources and change in land usage. As environmental sustainability is an ecological factor which can be measured in social science by development of statements or items which will access the way it brings the sustainability through urban nutri-gardens by the gardeners. Many of the earlier studies deliberated on scales constructed on overall sustainability, which inclusively economic sustainability and agricultural sustainability etc. these sustainability scales will measure all components of sustainability, such as economic, social and environmental, but there is no scale particularly related to environmental sustainability. Measuring environmental sustainability will helps in understanding the way in which planning the production systems and urbanisation of cities.

There is no scale to analyze the environmental sustainability of urban gardens, hence the present research study was taken up to develop and standardize a scale to analyze the environmental sustainability of urban gardens towards gardening.

#### METHODOLOGY

Study was conducted in Bengaluru as a greater number of people are interested in practicing urban farming as they are more concerned about their health. The intention of the study was to have a birds eye on the urban nutri-gardens effect on environmental sustainability. In the current study, a scale was instrumented to measure the environmental sustainability of urban gardens. Scale consisted six domains *viz.*, targeting renewable resources, conservation of ecosystem, pollution mitigation, health and welfare, intergenerational decisions and intrinsic rights. 110 items were framed after reviewing the related literature. Further, by following the 14 criteria enunciated by Edwards (1969), 70 items were drawn in line with label, abstract, construct and concept of the study. These items were subjected to relevancy test among ninety seven environmental, social, horticultural, food and nutritional scientists

working in State Agricultural Universities, Indian Council of Agricultural Research Institutes and Developmental Departments, to critically evaluate the relevancy of each items. As a result, 56 responses were received duly filled. The responses were obtained on five-point continuum from most relevant to not relevant further computed relevancy percentage, relevancy weightage and mean relevancy scores as given below.

*Relevancy Percentage (RP)* : Relevancy percentage was worked out by summing up the scores of Most Relevant (MR), Relevant (R), Somewhat Relevant (SWR), Less Relevant (LR) and Not Relevant (NR) categories, which were converted into percentage.

$$R.P. = \frac{MR \times 5 + R \times 4 + SWR \times 3 + LR \times 2 + NR \times 1}{\text{Maximum possible score}} \times 100$$

*Relevancy Weightage (RW)* : It was obtained by using the following formula

$$R.W. = \frac{MR \times 5 + R \times 4 + SWR \times 3 + LR \times 2 + NR \times 1}{\text{Maximum possible score}}$$

*Mean Relevancy Score (MRS)* : It was worked out using the following formula

$$M.R.S. = \frac{MR \times 5 + R \times 4 + SWR \times 3 + LR \times 2 + NR \times 1}{\text{Number of judges/experts responded}}$$

Statements having relevancy percent more than 75 per cent and above, relevance weightage more than 0.75 and above and mean relevancy score more than 3.75 were considered for the final selection of statements. Out of 70 statements, 11 statements did not qualify and hence were deleted (Kumar and Popat, 2016; James and Lakshminarayan, 2017; Biradar, *et. al*, 2021; Jiragal and Ganesamoorthi, 2022). These 59 relevant statements were then subjected for item analysis by interviewing 32 gardeners from Chintamani town of non-study area through personal interview technique and responses were obtained on five point continuum. Than score of the respondents for obtained summing up the score of all 59

statements. 25 per cent of respondents with highest total score and 25 per cent of with lowest scores were selected. These 2 groups provided the criterion groups in terms of evaluating the individual statements as suggested by Edwards (1969). The critical ratio was calculated by t-test to differentiate the high group from the low group. The 't' value was calculated by using the formula suggested by Edwards.

$$t = \frac{XH - XL}{\sqrt{\frac{SH_2}{nH} + \frac{SL_2}{nL}}}$$

Where,

$X_H$  = the mean score on given statement of the high group

$X_L$  = the mean score on given statement of the low group

$SH_2$  = the variance of the distribution of responses of high group to the statement

$SL_2$  = the variance of the distribution of responses of low group to the statement

$nH$  = Number of subjects in the high group

$nL$  = Number of subjects in the low group

$t$  = the extent to which a given statement differentiates between the high and low groups.

The selected items were then subjected for reliability testing using split half method. Scale was split into 2 halves on the basis of odd and even numbered items. These two forms were simultaneously administered to 32 gardeners of the study area. Chintamani town was selected and data were collected by personal interview technique. Collected responses were analysed using Karl Pearson's product moment correlation coefficient. To adjust the split half reliability in to the full test reliability Spearman-Brown prophecy formula was applied. Content validity of scale was established as selection of statements were made by seeking expert opinion. Construct validity was obtained by finding the correlation coefficient of sub domain score with the total score of the test.

Half test reliability formula

$$r_{1/2} = \frac{N(\Sigma XY) - (\Sigma X)(\Sigma Y)}{\sqrt{(N\Sigma X^2 - (\Sigma X)^2)(N\Sigma Y^2 - (\Sigma Y)^2)}}$$

Where,

$\Sigma X$  = Sum of the scores of the odd number items

$\Sigma Y$  = Sum of the scores of the even number items

$\Sigma X^2$  = Sum of the squares of the odd number items

$\Sigma Y^2$  = Sum of the squares of the even number items

Whole test reliability formula

$$r_{1/1} = \frac{2r_{1/2}}{1 + r_{1/2}}$$

Where,

$r_{1/2}$  = Half test reliability

The developed scale with 48 statements were administered to 90 respondents of Bengaluru district. The responses are obtained on 5 continuum *viz.*, strongly agree, agree, undecided, disagree and strongly disagree with scores of 5, 4, 3, 2 and 1, respectively. The environmental sustainability scores of each respondent was calculated by adding up the scores obtained on all the items. The environmental sustainability scores on the scale range from minimum of 48 to maximum of 240 based on their scores urban gardeners are divided into 5 categories likewise very high, high, medium, low and very low using the mean and standard deviation as a measure of check.

## RESULTS AND DISCUSSION

### Relevancy Analysis

Statements having the Relevancy percentage more than 75 per cent, Relevancy weightage more than 0.75 and the Mean relevancy scores more than 3.75 were considered for the final selection of statements. Out of 70 statements, 11 statements didn't qualify and hence these statements were deleted. So, total items considered for the next step were 59. Details of relevancy test was given in the Table 1.

### Item Analysis

Results of the item analysis are presented in the Table 2. After computing 't' values for all the items, statements with highest 't' value equal to or greater than 2.45 were selected. This enabled to select the items to be retained in the scale based on the highest discriminating values, besides eliminating those with poor discriminating ability and questionable validity. These 11 statements were disqualified.

### Reliability of the Scale

Retained 48 statements are given in Table 3. These statements when subjected to split-half reliability the value of correlation coefficient was 0.9210. The r value after adjusting using the Spearman-Brown prophency formula was 0.9592 which was significant at 1:00 percent level indicating the high reliability of the instrument. Therefore, the test is reliable to measure the environmental sustainability of the gardening (Ahmed, *et.al*, 2019)

### Validity

It refers to how well a scale analyses what it is purported to measure. The data was subjected to statistical validity, which was found to be 0.974 for scale which is greater than the standard requirement of 0.700. Hence, the validity coefficient was also found to be appropriate and suitable for the tool developed. Thus, the developed scale to analyze the environmental sustainability through urban nutri-gardens was feasible and appropriate. Table 4. Depicted summary of items retained across the different domains in different stages of the scale construction. Items identified initially were 70 and after judge's responses 11 were deleted. After item analysis based on t-test 48 items were retained finally.

### Scale Administration

It was found that 40.62 per cent of respondents belonged to medium, followed by 25.00 per cent very low, 18.75 per cent very high, 9.38 per cent high and 6.25 per cent very low environmental sustainability category. Ahmed, *et.al.*, (2019) reported that the adoption of organic farming, agro-ecosystems will retain its sustainability through the creation of a safe and diverse agro ecosystem that could meet the food requirements of the society in a sustainable way,

TABLE 1  
Results of relevancy test of pooled items

Statements	Relevancy Percentage	Relevancy weightage	Mean relevancy score
<i>A. Targeting renewable resources</i>			
Urban gardening enhances the efficiency of natural resources	85.17	0.85	4.26
Growing Urban gardening will reduce the dependence on the use of power consuming devices for moderating the room temperature	83.10	0.83	4.16
Urban gardens increases the utilisation of natural resources	84.83	0.85	4.24
Urban gardens consume waste water for growing plants which reduces the dependence on fresh water	84.14	0.84	4.21
Urban gardens reduce the no of visits to markets for purchases, so decrease fuel requirements and reduces carbon foot prints	84.14	0.84	4.21
Urban gardening promotes water harvesting in the cities	85.17	0.85	4.26
Bio fortification is possible through urban gardening	74.48	0.74	3.72
Government policies which enable to afford renewable resources	74.48	0.74	3.72
Cost is more in urban gardening while shifting toward non-renewable energy sources	78.97	0.79	3.95
Usage of non-renewable energy sources in urban areas is inherently different from that of rural areas	80.69	0.81	4.03
Proper utilisation of non-renewable energy resources in urban gardening is cost and time effective	82.76	0.83	4.14
Urban gardening efficiently uses the natural resources which are otherwise considered as near waste	78.62	0.79	3.93
Crops grown on the rooftops are efficiently absorb and use the sunlight for their metabolism	85.52	0.86	4.28
Soil medium which normally acts as growth media can be replaced by any other alternative media in urban gardens thus saving top soil	84.14	0.84	4.21
<i>B. Conservation of ecosystem</i>			
Urban gardens contributes towards increase bio life of the earth	82.41	0.82	4.12
Urban gardens contributes towards reducing global warming	86.90	0.87	4.34
Urban gardening facilitates in creation of the favourable micro climate of surrounding locality	88.62	0.89	4.43
Urban gardens minimise accumulation of heavy metals in the soil	80.69	0.81	4.03
Urban gardens reduce the usage of non-renewable energy sources by decreasing use of agrochemicals	84.83	0.85	4.24
Urban gardening contributes in mitigating the greenhouses effect and global warming through its ability to sequester carbon in the soil	81.72	0.82	4.09
Urban gardening encourage birds and other natural predators to live happily on gardens which assists in natural pest control	81.38	0.81	4.07

Table 1 Continued

Statements	Relevancy Percentage	Relevancy weightage	Mean relevancy score
Urban gardening helps in producing food in the locality which intern minimises transportation related greenhouse gas emission (carbon foot prints)	86.21	0.86	4.31
Urban gardens provide fresh and healthy food using less energy	84.14	0.84	4.21
Urban gardens absorb the greenhouse gases to maximum extent.	79.66	0.80	3.98
Urban farming and gardening act as a saviour of local flora and fauna thereby maintain local biodiversity	81.38	0.81	4.07
Urban gardening is a tool to effectively convert food waste into a nutritive compost	87.24	0.87	4.36
Urban gardening protects and recharges earth's resources like soil and ground water	81.72	0.82	4.09
<i>C. Pollution mitigation</i>			
Urban gardens reduces the pollution to greater extent when compared to conventional farming	74.14	0.74	3.71
Use of pesticides and chemical spray on plants in urban garden contaminates the soil, water and surrounding environment	84.14	0.84	4.21
Urban garden reduces he possibility of use of plastic bags to carry food items like vegetables and fruits	87.24	0.87	4.36
Due to improvement in micro-climate, it reduces the dependence on air-conditioners thereby contributes for low CfC emission	85.17	0.85	4.26
Green foliage cover on these gardens acts as a natural sink for common contaminants	82.07	0.82	4.10
Plant photosynthesis minimises Co <sub>2</sub> emitted in urban area	83.45	0.83	4.17
Urban gardening reduces the soil compaction and loosens soil structure	72.41	0.72	3.62
Urban gardening increases the microbial activity in the soil	81.72	0.82	4.09
Urban gardening helps in recycling waste water for irrigating urban gardens	84.14	0.84	4.21
Some tree species will reduce the noise pollution by reducing frequency of sound waves	77.59	0.78	3.88
Urban gardening destroys the soil structure in the garden area	63.79	0.64	3.19
Urban gardening reduces the wind speed to some extent as natural barrier	74.83	0.75	3.74
Urban gardening ultimately leads to chemical free environment	74.83	0.75	3.74
<i>D. Health and Welfare</i>			
Biodiversity in urban areas increases lifespan by producing healthy ecosystem such as clean air, water and soil	85.52	0.86	4.28
Growing by themselves can also substantially helps to minimise food wastage	84.14	0.84	4.21
Green cover in and around residential area helps in preventing runoff	82.76	0.83	4.14
Urban gardening generates employment opportunities	81.38	0.81	4.07

Table 1 Continued

Statements	Relevancy Percentage	Relevancy weightage	Mean relevancy score
Urban gardening increases the nutritional diversity among households	85.17	0.85	4.26
Day to day gardening activities helps in burning excess calories	84.48	0.84	4.22
Urban gardening decreases excess waste of manpower at house hold level	82.41	0.82	4.12
Urban gardening reduces the mental stress & provide some sort of relaxation	87.59	0.88	4.38
Threatened and endangered bird species may find suitable habitat on these gardens	82.07	0.82	4.10
Contribute forcommunity gardens by using wastage of urban gardens	83.10	0.83	4.16
<i>E. Intergenerational decisions</i>			
Urban gardens produce which is grown local has high perceived value and less likely to be sorted as trash	82.41	0.82	4.12
Urban gardening helps the children learn about the gardening practices along with their parents	85.86	0.86	4.29
By formation of groups/clusters, common space can be converted into gardens	85.86	0.86	4.29
Supply of produce from urban areas can stabilise prices and ensure year round supply	83.79	0.84	4.19
Use of high-tech agricultural practices will conserve and save resources	82.41	0.82	4.12
To mitigate the effect of climate change urban gardening is one of the alternative	83.45	0.83	4.17
In long run it will reduce the amount spent on carbon credit	80.69	0.81	4.03
Urban gardens can facilitate agritourism and recreation	83.45	0.83	4.17
Urban gardens will reduce pressure on rural areas for meeting food demand	79.31	0.79	3.97
Urban gardens don't get any long-term benefits	69.31	0.69	3.47
Growing urban garden is a short term nutritional plan	74.48	0.74	3.72
Sometimes harmful pesticides used in urban gardens have residual effects for decades	73.79	0.74	3.69
Urban gardens increases the green space in cities.	87.93	0.88	4.40
<i>F. Intrinsic rights</i>			
Urban gardens creates micro- bio diversity for many living creatures such as insects and birds	84.83	0.85	4.24
Apart from providing monitory benefits urban gardens also concerned with nutritional values	83.45	0.83	4.17
Can reintroduce ayurvedic culture in urban areas	78.62	0.79	3.93
Urban people get better insights about humans environment interactions	74.48	0.74	3.72
Urban gardens conserve few natural enemies, which are useful for ecosystem	81.38	0.81	4.07
Urban gardens create interest in people by realising the value of nutritional food	82.07	0.82	4.10
Urban gardens improve access to fresh and green vegetables	85.17	0.85	4.26

TABLE 2  
Paired two sample t-test of criterion groups

Statements	Paired two sample	Status
<i>A. Targeting renewable resources</i>		
Urban gardening enhances the efficiency of natural resources.	4.8990	Included
Growing Urban gardening will reduce the dependence on the use of power consuming devices for moderating the room temperature.	3.3141	Included
Urban gardens increases the utilisation of natural resources.	1.9467	Excluded
Urban gardens consume waste water for growing plants which reduces the dependence on fresh water.	3.2615	Included
Urban gardens reduce the no of visits to markets for purchases, so decrease fuel requirements and reduces carbon foot prints.	4.1312	Included
Urban gardening promotes water harvesting in the cities.	3.8490	Included
Cost is more in urban gardening while shifting toward non-renewable energy sources.	6.7937	Included
Usage of non-renewable energy sources in urban areas is inherently different from that of rural areas.	1.5240	Excluded
Proper utilisation of non-renewable energy resources in urban gardening is cost and time effective.	1.2940	Excluded
Urban gardening efficiently uses the natural resources which are otherwise considered as near waste.	4.4836	Included
Crops grown on the rooftops are efficiently absorb and use the sunlight for their metabolism.	4.0505	Included
Soil medium which normally acts as growth media can be replaced by any other alternative media in urban gardens thus saving top soil.	2.0835	Excluded
<i>B. Conservation of ecosystem</i>		
Urban gardens contributes towards increase bio life of the earth	8.8465	Included
Urban gardens contributes towards reducing global warming	4.1952	Included
Urban gardening facilitates in creation of the favourable micro climate of surrounding locality	5.3666	Included
Urban gardens minimise accumulation of heavy metals in the soil.	4.5383	Included
Urban gardens reduce the usage of non-renewable energy sources by decreasing use of agrochemicals	1.5523	Excluded
Urban gardening contributes in mitigating the greenhouses effect and global warming through its ability to sequester carbon in the soil.	8.0333	Included
Urban gardening encourage birds and other natural predators to live happily on gardens which assists in natural pest control.	4.0000	Included
Urban gardening helps in producing food in the locality which intern minimises transportation related greenhouse gas emission (carbon foot prints).	4.5883	Included
Urban gardens provide fresh and healthy food using less energy	4.1284	Included
Urban gardens absorb the greenhouse gases to maximum extent.	3.6600	Included

Table 2 Continued



Statements	Paired two sample	Status
Urban farming and gardening act as a saviour of local flora and fauna thereby maintain local biodiversity	4.6569	Included
Urban gardening is a tool to effectively convert food waste into a nutritive compost.	7.2296	Included
Urban gardening protects and recharges earth's resources like soil and ground water	8.8465	Included
<i>C. Pollution mitigation</i>		
Use of pesticides and chemical spray on plants in urban garden contaminates the soil, water and surrounding environment.	4.0762	Included
Urban garden reduces the possibility of use of plastic bags to carry food items like vegetables and fruits	3.3806	Included
Due to improvement in micro-climate, it reduces the dependence on air-conditioners thereby contributes for low CfC emission	5.7208	Included
Green foliage cover on these gardens acts as a natural sink for common contaminants.	1.6557	Excluded
Plant photosynthesis minimises Co <sub>2</sub> emitted in urban area.	0.9547	Excluded
Urban gardening increases the microbial activity in the soil.	3.0193	Included
Urban gardening helps in recycling waste water for irrigating urban gardens.	5.3079	Included
Some tree species will reduce the noise pollution by reducing frequency of sound waves.	4.6188	Included
<i>D. Health and Welfare</i>		
Biodiversity in urban areas increases lifespan by producing healthy ecosystem such as clean air, water and soil.	2.0966	Excluded
Growing by themselves can also substantially help to minimise food wastage.	4.4921	Included
Green cover in and around residential area helps in preventing runoff.	4.1952	Included
Urban gardening generates employment opportunities.	5.9876	Included
Urban gardening increases the nutritional diversity among households.	1.6749	Excluded
Day to day gardening activities help in burning excess calories.	4.0505	Included
Urban gardening decreases excess waste of manpower at house hold level.	3.6003	Included
Urban gardening reduces the mental stress and provide some sort of relaxation.	4.0000	Included
Threatened and endangered bird species may find suitable habitat on these gardens.	3.3356	Included
Contribute for community gardens by using wastage of urban gardens.	4.9820	Included
<i>E. Intergenerational decisions</i>		
Urban gardens produce which is grown local has high perceived value and less likely to be sorted as trash.	6.7330	Included
Urban gardening helps the children learn about the gardening practices along with their parents.	4.2426	Included
By formation of groups/clusters, common space can be converted into gardens.	7.4833	Included
Supply of produce from urban areas can stabilise prices and ensure year round supply.	3.6924	Included
Use of high-tech agricultural practices will conserve and save resources.	4.1312	Included
To mitigate the effect of climate change urban gardening is one of the alternative.	3.2026	Included
In long run it will reduce the amount spent on carbon credit.	3.0500	Included
Urban gardens can facilitate agritourism and recreation	4.3028	Included

Table 2 Continued

Statements	Paired two sample	Status
Urban gardens will reduce pressure on rural areas for meeting food demand.	3.8490	Included
Urban gardens increases the green space in cities.	3.3466	Included
<i>F. Intrinsic rights</i>		
Urban gardens creates micro- bio diversity for many living creatures such as insects and birds.	6.0000	Included
A part from providing monitory benefits urban gardens also concerned with nutritional values.	2.5621	Included
Can reintroduce ayurvedic culture in urban areas.	3.1704	Included
Urban gardens conserve few natural enemies, which are useful for ecosystem.	1.4660	Excluded
Urban gardens create interest in people by realising the value of nutritional food.	1.5396	Excluded
Urban gardens improve access to fresh and green vegetables.	5.8384	Included

TABLE 3  
Items retained in the final scale

Statements
<i>A. Targeting renewable resources</i>
Urban gardening enhances the efficiency of natural resources.
Growing Urban gardening will reduce the dependence on the use of power consuming devices for moderating the room temperature.
Urban gardens consume waste water for growing plants which reduces the dependence on fresh water.
Urban gardens reduce the no of visits to markets for purchases, so decrease fuel requirements and reduces carbon foot prints.
Urban gardening promotes water harvesting in the cities.
Cost is more in urban gardening while shifting toward non-renewable energy sources.
Urban gardening efficiently uses the natural resources which are otherwise considered as near waste.
Crops grown on the rooftops are efficiently absorb and use the sunlight for their metabolism.
<i>B. Conservation of ecosystem</i>
Urban gardens contributes towards increase bio life of the earth
Urban gardens contributes towards reducing global warming
Urban gardening facilitates in creation of the favourable micro climate of surrounding locality
Urban gardens minimise accumulation of heavy metals in the soil.

Table 3 Continued

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 Statements
 

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Urban gardening contributes in mitigating the greenhouses effect and global warming through its ability to sequester carbon in the soil.

Urban gardening encourage birds and other natural predators to live happily on gardens which assists in natural pest control.

Urban gardening helps in producing food in the locality which intern minimises transportation related greenhouse gas emission (carbon foot prints).

Urban gardens provide fresh and healthy food using less energy

Urban gardens absorb the greenhouse gases to maximum extent.

Urban farming and gardening act as a saviour of local flora and fauna thereby maintain local biodiversity

Urban gardening is a tool to effectively convert food waste into a nutritive compost.

Urban gardening protects and recharges earth's resources like soil and ground water

C. *Pollution mitigation*

Use of pesticides and chemical spray on plants in urban garden contaminates the soil, water and surrounding environment.

Urban garden reduces he possibility of use of plastic bags to carry food items like vegetables and fruits

Due to improvement in micro-climate, it reduces the dependence on air-conditioners thereby contributes for low CfC emission

Urban gardening increases the microbial activity in the soil.

Urban gardening helps in recycling waste water for irrigating urban gardens.

Some tree species will reduce the noise pollution by reducing frequency of sound waves.

D. *Health and welfare*

Growing by themselves can also substantially helps to minimise food wastage.

Green cover in and around residential area helps in preventing runoff.

Urban gardening generates employment opportunities.

Day to day gardening activities helps in burning excess calories.

Urban gardening decreases excess waste of manpower at house hold level.

Urban gardening reduces the mental stress and provide some sort of relaxation.

Threatened and endangered bird species may find suitable habitat on these gardens.

Contribute for community gardens by using wastage of urban gardens.

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 Table 3 Continued

## Statements

E. *Intergenerational decisions*

Urban gardens produce which is grown local has high perceived value and less likely to be sorted as trash.

Urban gardening helps the children learn about the gardening practices along with their parents.

By formation of groups/clusters, common space can be converted into gardens.

Supply of produce from urban areas can stabilise prices and ensure year round supply.

Use of high-tech agricultural practices will conserve and save resources.

To mitigate the effect of climate change urban gardening is one of the alternative.

In long run it will reduce the amount spent on carbon credit.

Urban gardens can facilitate agritourism and recreation

Urban gardens will reduce pressure on rural areas for meeting food demand.

Urban gardens increases the green space in cities.

F. *Intrinsic rights*

Urban gardens creates micro- bio diversity for many living creatures such as insects and birds.

Apart from providing monitory benefits urban gardens also concerned with nutritional values.

Can reintroduce ayurvedic culture in urban areas.

Urban gardens improve access to fresh and green vegetables.

TABLE 4  
Details of construction and standardization of perception scale.

Components/Domains	Total items	Items retains after relevancy test	Items retained after items analysis
Targeting renewable resources	14	12	8
Conservation of ecosystem	13	13	12
Pollution mitigation	13	8	6
Health and welfare	10	10	8
Intergenerational decisions	13	10	10
Intrinsic rights	7	6	4
Total	70	59	48

Table 3 Continued

TABLE 5  
Results of the administered scale

Category	Respondent	
	Score range	Frequency (%)
Very Low environmental sustainability (<Mean -S.D.)	172.28	8 (25 %)
Low environmental sustainability (<Mean -0.425*S.D.)	172.28 to 186.35	2(6.25%)
Medium environmental sustainability (<Mean +0.425*S.D. to >Mean - 0.425*S.D.)	186.35 to 207.14	13 (40.62 %)
High environmental sustainability (>Mean + 0.425S.D.)	207.14 to 221.22	3(9.38)
Very High environmental sustainability (>Mean + S.D.)	221.22	6 (18.75)
Mean=196.75	S.D.= 24.47	

along with the conservation of its scare resources. The results of present study were in consonance with study of Kowsalya and Krishnamurthy, (2017).

The environmental sustainability scale developed is found to be reliable, valid and internally consistent, hence it can be used to analyze the environmental sustainability of urban gardens.

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## Effect of Nano Phosphorus Fertilizers on Growth and Yield of Maize (*Zea mays* L.) in Central Dry Zone of Karnataka

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### ABSTRACT

A field experiment was conducted during *Kharif* 2021 in farmer's field at Halavarthy village of Davanagere district, Karnataka, which comes under Central Dry Zone of Karnataka (Zone 4), using RCBD with eleven treatments and three replications to evaluate the impact of three different nano phosphorus fertilizers on the productivity of maize. Treatments included  $T_1$ : Absolute control,  $T_2$ : 100 per cent recommended package of practice (RPP),  $T_3$  to  $T_5$ : 75 per cent RDP (75% recommended dose of P) through SSP + 5 per cent RDP through NP1(hydroxyapatite nano fertilizer), NP2 (nano rock phosphate) and NP3 (hydroxyapatite nanoparticles coated with CMC),  $T_6$  to  $T_8$ : 75 per cent RDP + 1 per cent RDP through foliar spray of NP1, NP2 and NP3,  $T_9$  to  $T_{11}$ : 75 per cent RDP + 5 per cent RDP through soil application of NP1, NP2 and NP3 and 1 per cent RDP through foliar spray of NP1, NP2 and NP3, respectively. Results revealed that application of phosphorus in nano form had significant effect on growth and yield of maize. Treatment with 75 per cent RDP through SSP + 5 per cent RDP through soil application of NP1 + 1 per cent RDP through foliar spray of NP1 recorded significantly higher plant height, number of leaves plant<sup>-1</sup>, chlorophyll content, dry matter accumulation, cob length (19.01 cm), number of rows cob<sup>-1</sup> (16.27), number of kernels row<sup>-1</sup> (35.27), test weight (33.20 g), stover yield (82.23 q ha<sup>-1</sup>) and kernel yield (73.59 q ha<sup>-1</sup>) over absolute control and were better when compared to  $T_2$  (100 % RPP).

**Keywords :** Hydroxyapatite nanoparticles, Nano rock phosphate, Coated hydroxyapatite nanoparticles, Carboxy methyl cellulose, Maize productivity, Yield attributes

PHOSPHORUS (P) along with other nutrient elements plays a central role in achieving food and nutritional security of ever growing population of the world. In the absence of any one of the essential nutrients makes it impossible to achieve this goal. Among the essential nutrients, potential reserve of P resource is dwindling in the world, if exhausted it spell a dooms day on crop production. On the other hand, only 15 to 20 per cent of the applied P being utilized by crop. The low P use efficiency in crop production reported may be attributed to the faster rate of conversion of applied P into non available forms as a result of its reaction with soil chemical

constituents (Ca, Fe or Al). The efforts are underway to make use of such fixed P by using PSB or mobilizing using VAM. Even under such circumstances the P use efficiency still remains low. With recent advances happened in material science in the synthesis of nano materials/ products for variety of uses, the fertilizer industry too started using the nano technology for the synthesis of nano fertilizers, which have many advantages over conventional nutrient carrier fertilizer materials in crop production. Employing nanotechnology in synthesis and formulations of nano fertilizers and their subsequent use is regarded as a breakthrough in achieving higher

nutrient use efficiency with minimum environmental risk. Nano P fertilizers are those which contain conventional P fertilizers or rock phosphate or hydroxyapatite encapsulated in nanomaterials or coated with a thin protective nano scale polymeric film or delivered as nanoemulsions or nanoparticles. Nano fertilizers release nutrients into the soil gradually in a sustained manner. The controlled release of nutrient may reduce the loss of nutrient from soil. Nano coatings on fertilizer particles can hold the material more strongly when sprayed on the plant due to the higher surface tension. Nano material increases the plant uptake efficiency of nutrients and reduce the adverse impacts of conventional fertilizer application thus, enhances the growth and yield of crops. Thus, realizing the importance of nano fertilizers in crop nutrition, an experiment was conducted with the objective to study the 'Effect of synthesized nano phosphorus fertilizers on growth and yield of maize'

#### MATERIAL AND METHODS

A field experiment was conducted during *Kharif* 2021 in farmer's field at Halavarthy village of Davanagere district, Karnataka, which comes under Agro Climatic Zone-4, Central Dry Zone of Karnataka. It lies between 76°08' E longitude and 14°39' N latitude with an altitude of 690 ± 04 m above mean sea level. The experiment consisted of eleven treatment combinations *viz.*, T<sub>1</sub>: Absolute control, T<sub>2</sub>: 100 per cent recommended package of practice (RPP), T<sub>3</sub> to T<sub>5</sub>: 75 per cent RDP (75% recommended dose of P) through SSP + 5 per cent RDP through NP1 (hydroxyapatite nano fertilizer), NP2 (nano rock phosphate) and NP3 (hydroxyapatite nanoparticles coated with CMC) respectively, T<sub>6</sub> to T<sub>8</sub>: 75 per cent RDP through SSP + 1 per cent RDP through foliar spray of NP1, NP2 and NP3 respectively, T<sub>9</sub> to T<sub>11</sub>: 75 per cent RDP through SSP + 5 per cent RDP through soil application of NP1, NP2 and NP3 + 1 per cent RDP through foliar spray of NP1, NP2 and NP3, respectively.

#### Synthesis of Nano Phosphorus Fertilizers

Three types of nano phosphorus fertilizers *viz.*, hydroxyapatite nanoparticles (NP1), nano rock

phosphate (NP2) and hydroxyapatite nanoparticles coated with carboxy methyl cellulose (NP3) were synthesized in laboratory as per the standard protocol. Hydroxyapatite nanoparticles were synthesized using chemical synthesis method as described by Mateus *et al.* (2007) using aqueous solution of calcium hydroxide (1M) and orthophosphoric acid (85%). Rock phosphate nano fertilizer was prepared by ball-milling the rock phosphate till it reaches nano size. Nano hydroxyapatite particles were coated with CMC as per the method described by Liu and Lal (2014) using nano hydroxyapatite and carboxy methyl cellulose powder and the prepared material was characterized using XRD, SEM, EDS and FTIR. The phosphorus (P) content in these synthesized NP1, NP2 and NP3 was 15.82, 9.48 and 15.03 per cent, respectively.

#### Initial Physico-chemical Properties of the Soil at Experimental Site

The texture of soil was sandy loam with 72.18 per cent sand, 9.86 per cent silt and 17.96 per cent clay. The soil was nearly neutral (pH: 6.97) in reaction, low in organic carbon content (4.34 g kg<sup>-1</sup>) and normal with respect to salt content (0.124 ds m<sup>-1</sup>). The available nitrogen content was low (183.56 kg ha<sup>-1</sup>), phosphorus was medium (27.29 kg P<sub>2</sub>O<sub>5</sub> ha<sup>-1</sup>) and potassium was medium (373.16 kg K<sub>2</sub>O ha<sup>-1</sup>) and medium in micro nutrients.

#### Collection of Growth and Yield Parameters

Five plants from each treatment in all three replications were labelled to collect growth and yield attributes of maize. Growth observations were taken at 30, 60, 90 DAS and at harvest. Plant height was measured from base of plant to the base of fully opened top most leaf and expressed in centimetre. Number of leaves per plant was recorded by counting fully opened leaves, the relative chlorophyll content of maize leaves was measured by placing chlorophyll meter at 20 to 25 cm above on one side of the mid rib of the leaf blade, midway between the leaf base and leaf tip between 8 to 9 AM in morning or between 5 to 6 PM in evening hours. Cob length was

measured from base to tip of cob, from all the five cobs collected from tagged plants. Number of rows per cob and number of kernels per row were recorded by counting them manually. Test weight was recorded by weighing hundred grains from each treatment and expressed in grams.

## RESULTS AND DISCUSSION

### Growth Parameters

Data on growth parameters of maize presented in Tables 1 and 2 indicated that nano phosphorus fertilizers application either through soil and or foliar spray recorded significantly higher growth parameters of maize viz., plant height, number of leaves per plant, chlorophyll meter reading and dry matter production than absolute control. At 30 DAS, significantly higher plant height of 33.55 cm was recorded in T<sub>2</sub> (100% RPP) than absolute control but was on par with all other treatments that received

phosphorus through conventional and nano sources (T<sub>3</sub> to T<sub>11</sub>) 20 per cent less than normal recommended P dose. Significantly maximum number of leaves per plant (7.27), chlorophyll meter reading (461.23) and dry matter production (11.13 g plant<sup>-1</sup>) were recorded in the T<sub>9</sub> treatment that received 75 per cent RDP through SSP + 5 per cent RDP - NP1+ FS of 1 per cent RDP - NP1 over absolute control (T<sub>1</sub>) but it was on par with the treatments T<sub>2</sub> to T<sub>11</sub>.

At 60 DAS, 90 DAS and at harvest, plant height (195.18, 202.72 and 203.98 cm, respectively), number of leaves per plant (13.66, 15.03 and 13.10, respectively), chlorophyll meter reading (496.33, 364.00 and 139.17, respectively) and dry matter production (101.57, 147.64 and 162.72 g plant<sup>-1</sup>, respectively) were significantly higher in treatment T<sub>9</sub> that received 75 per cent RDP through SSP + 5 per cent RDP - NP1+ FS of 1 per cent RDP - NP1 over absolute control (T<sub>1</sub>). However, it was on par with

TABLE 1

Effect of nano P fertilizers on plant height ( cm), number of leaves plant<sup>-1</sup> of maize at different growth stages

Treatments	Plant height (cm)				Number of leaves plant <sup>-1</sup>			
	30 DAS	60 DAS	90 DAS	At Harvest	30 DAS	60 DAS	90 DAS	At Harvest
T <sub>1</sub> : Control	22.55	122.73	125.93	127.19	5.78	8.37	8.87	7.40
T <sub>2</sub> : 100 % RPP	33.55	188.54	194.40	195.66	6.44	12.27	12.87	11.27
T <sub>3</sub> : 75 % RDP + 5 % RDP - NP1	31.85	193.57	198.77	198.36	6.98	12.77	13.83	11.90
T <sub>4</sub> : 75 % RDP + 5 % RDP - NP2	31.39	190.52	196.72	197.32	6.78	12.88	13.80	11.83
T <sub>5</sub> : 75 % RDP + 5 % RDP -NP3	31.71	192.38	195.91	197.51	6.97	12.73	13.67	11.73
T <sub>6</sub> : 75 % RDP + FS of 1 % RDP - NP1	30.93	189.58	196.45	197.71	6.66	12.91	13.80	11.77
T <sub>7</sub> : 75 % RDP + FS of 1 % RDP -NP2	31.08	188.60	195.13	196.39	6.48	12.60	13.53	11.50
T <sub>8</sub> : 75 % RDP + FS of 1 % RDP -NP3	30.30	189.63	195.83	197.09	6.58	12.34	13.60	11.60
T <sub>9</sub> : T <sub>3</sub> + FS of 1 % RDP - NP1	32.78	195.18	202.72	203.98	7.27	13.66	15.03	13.10
T <sub>10</sub> : T <sub>4</sub> + FS of 1 % RDP - NP2	31.45	193.70	201.23	202.49	7.05	13.13	14.93	12.67
T <sub>11</sub> : T <sub>5</sub> + FS of 1 % RDP - NP3	31.45	194.66	201.92	203.18	7.03	13.53	15.00	13.07
S.Em ±	1.39	7.80	9.28	10.49	0.34	0.52	0.76	0.64
CD @ 5%	4.11	23.03	27.38	30.96	NS	1.54	2.24	1.89

T1 : Control  
 T2 : 100 % RPP  
 T3 : 75 % RDP + 5 % RDP - NP1  
 T4 : 75 % RDP + 5 % RDP - NP2  
 T5 : 75 % RDP + 5 % RDP -NP3  
 T6 : 75 % RDP + FS of 1 % RDP - NP1

T7 : 75 % RDP + FS of 1 % RDP -NP2  
 T8 : 75 % RDP + FS of 1 % RDP -NP3  
 T9 : T3 + FS of 1 % RDP - NP1  
 T10: T4 + FS of 1 % RDP - NP2  
 T11: T5 + FS of 1 % RDP - NP3



TABLE 2

Effect of nano P fertilizers on chlorophyll meter reading and dry matter production (g plant<sup>-1</sup>) of maize plant at different growth stages

Treatments	Chlorophyll meter reading				Dry matter production (g plant <sup>-1</sup> )			
	30 DAS	60 DAS	90 DAS	At Harvest	30 DAS	60 DAS	90 DAS	At Harvest
T <sub>1</sub> : Control	368.43	413.42	295.33	103.60	8.33	48.00	69.66	79.24
T <sub>2</sub> : 100 % RPP	424.06	452.54	331.07	125.27	9.85	90.67	129.24	145.83
T <sub>3</sub> : 75 % RDP + 5 % RDP - NP1	445.07	477.94	342.23	132.00	10.85	97.82	137.27	156.54
T <sub>4</sub> : 75 % RDP + 5 % RDP - NP2	442.75	470.38	340.17	130.23	10.46	95.30	135.20	154.10
T <sub>5</sub> : 75 % RDP + 5 % RDP -NP3	445.29	471.96	341.90	131.53	10.65	96.80	135.23	155.19
T <sub>6</sub> : 75 % RDP + FS of 1 % RDP - NP1	442.59	478.52	341.13	131.48	10.04	96.55	134.53	154.77
T <sub>7</sub> : 75 % RDP + FS of 1 % RDP -NP2	440.18	474.45	340.23	130.27	10.00	94.27	133.33	153.30
T <sub>8</sub> : 75 % RDP + FS of 1 % RDP -NP3	441.90	476.22	340.07	131.23	10.02	95.70	133.43	154.20
T <sub>9</sub> : T <sub>3</sub> + FS of 1 % RDP - NP1	461.23	496.33	364.00	139.17	11.13	101.57	147.64	162.72
T <sub>10</sub> : T <sub>4</sub> + FS of 1 % RDP - NP2	459.01	490.00	361.33	136.67	10.93	100.47	144.23	159.83
T <sub>11</sub> : T <sub>5</sub> + FS of 1 % RDP - NP3	460.03	494.00	363.33	138.29	10.97	101.02	146.67	160.13
S.Em ±	13.47	14.93	12.35	6.07	0.47	3.86	6.39	5.97
CD @ 5%	39.75	44.05	36.45	17.92	1.40	11.40	18.85	17.61

T1 : Control

T2 : 100 % RPP

T3 : 75 % RDP + 5 % RDP - NP1

T4 : 75 % RDP + 5 % RDP - NP2

T5 : 75 % RDP + 5 % RDP -NP3

T6 : 75 % RDP + FS of 1 % RDP - NP1

T7 : 75 % RDP + FS of 1 % RDP -NP2

T8 : 75 % RDP + FS of 1 % RDP -NP3

T9 : T3 + FS of 1 % RDP - NP1

T10: T4 + FS of 1 % RDP - NP2

T11: T5 + FS of 1 % RDP - NP3

the treatment that received 100 per cent RPP (T<sub>2</sub>), and with all other treatments that received 75 per cent RDP through SSP along with soil and/or foliar applied nano phosphorus treatments (T<sub>3</sub> to T<sub>11</sub>).

The results indicated that soil and / or foliar application of nano phosphorus fertilizer enhanced the growth parameters of maize compared to control and produced better and at par growth parameters with 100 per cent RPP applied treatment (Tables 1 and 2). The better and / or at par growth parameters observed in maize with the application of nano phosphorus fertilizers even at 20 per cent reduced level might be attributed to slow and continued release of P, which coincided with crop nutrient demand (Manikandan and Subramanian, 2015 in maize; Rajendran *et al.*, 2017 and Babubhai, 2018 in maize). The enhanced and continued P availability from these nano sources at distinct physiological phases, might have increased

metabolic processes in plants (protein formation, photosynthesis, cell division, cell respiration and energy storage, *etc.*) thereby increased the below ground (root growth) and above ground biomass (plant height, number of leaves and dry matter production). Besides, application of P in nano form allows better dissolution and faster absorption and assimilation by the plants compared to P supplied through conventional fertilizers. Liu and Lal (2014) also confirmed that biomass productions were enhanced by 18.2 per cent (above ground) and 41.2 per cent (below ground) in nano hydroxyapatite treated plants compared to di calcium phosphate applied soybean plants and application of nano hydroxyapatite (nHA) increased the growth rate and seed yield by 32.6 per cent and 20.4 per cent, respectively compared to regular P treated soybean plants and these results are in agreement with the research findings of Beeresha and Jayadeva (2020)

in maize; Sohair *et al.* (2018) in Egyptian cotton; Harish and Gowda (2017) in groundnut; Rajendran *et al.* (2017) in greengram and Soliman *et al.* (2016) in Baobab. In comparison with mere soil application or foliar application of nano hydroxyapatite fertilizer, combined soil and foliar application of hydroxyapatite nano fertilizer (NP1) recorded higher plant height, number of leaves per plant, chlorophyll meter reading and dry matter accumulation at all growth stages of maize plants might be due to quicker absorption of applied nutrients in nano form because nano coatings on fertilizers hold nutrient more strongly on plant surface due to its high surface tension, smaller size and higher surface area and thus it get easily penetrated into plant leaves and contributed to increased growth parameters.

### Yield Parameters

The data on yield attributes *viz.*, cob length, number of rows per cob, number of kernels per row and test weight of maize as influenced by the application of P through conventional and nano phosphorus fertilizers are presented in Table 3.

The experimental data indicated that, significantly higher cob length of 19.01 cm was recorded in T<sub>9</sub>,

treatment compared to control (11.45 cm) and was on par with all other treatments. Least number of rows cob<sup>-1</sup> (10.73) was recorded in control which was increased significantly to 16.67 with the application of 75 per cent RDP through SSP + 5 per cent RDP - NP1 + FS of 1 per cent RDP - NP1 (T<sub>9</sub> treatment) and the treatment T<sub>9</sub> was on par with all the treatments that received P either through conventional and or along with nano sources (T<sub>2</sub> to T<sub>11</sub>). Similarly, the number of kernels cob<sup>-1</sup> (35.27) were significantly higher in the treatment T<sub>9</sub> (T<sub>3</sub>+1% RDP - NP1 through foliar spray) which was on par with all other treatments except control (21.53). Compared to control, test weight was significantly increased in all the treatments which received P either through conventional or nano sources.

Compared to control, application of 75 per cent RDP through SSP along with 5 per cent RDP through soil application of hydroxyapatite nano fertilizer (NP1) + 1 per cent RDP through foliar application of NP1 recorded higher values of yield attributes of maize than that was recorded with 100 per cent RPP applied treatment. The observed increase in yield parameters with the application of nano P fertilizers along with

TABLE 3  
Yield attributes of maize as influenced by the application of nano P fertilizers

Treatments	Cob length (cm)	No of rows Cob <sup>-1</sup>	No of kernels Row <sup>-1</sup>	Test Weight (g)
T <sub>1</sub> : Control	11.45	10.73	21.53	26.14
T <sub>2</sub> : 100 % RPP	16.53	13.78	31.22	30.15
T <sub>3</sub> : 75 % RDP + 5 % RDP - NP1	17.67	14.67	33.53	31.83
T <sub>4</sub> : 75 % RDP + 5 % RDP - NP2	17.50	14.00	33.47	31.42
T <sub>5</sub> : 75 % RDP + 5 % RDP - NP3	17.53	14.33	33.47	31.76
T <sub>6</sub> : 75 % RDP + FS of 1 % RDP - NP1	17.50	14.33	33.53	31.64
T <sub>7</sub> : 75 % RDP + FS of 1 % RDP - NP2	17.20	14.06	33.20	31.63
T <sub>8</sub> : 75 % RDP + FS of 1 % RDP - NP3	17.63	14.00	33.53	31.09
T <sub>9</sub> : T <sub>3</sub> + FS of 1 % RDP - NP1	19.01	16.27	35.27	33.20
T <sub>10</sub> : T <sub>4</sub> + FS of 1 % RDP - NP2	18.10	15.33	34.47	32.25
T <sub>11</sub> : T <sub>5</sub> + FS of 1 % RDP - NP3	19.00	15.73	35.00	33.07
S.Em ±	0.86	0.86	1.53	1.15
CD @ 5%	2.53	2.54	4.52	3.41

conventional P fertilizers might be attributed to improvement in growth parameters. The improvement in growth parameters was due to synchronized supply of P (15.82%) and Ca (32%) contained in NP1 and also due to the slow and continued release of P from nano sources throughout the crop period and calcium present in these synthesized materials played a vital role in meristem growth, cell elongation and nutrient uptake like nitrogen which enhanced the vegetative growth and ultimately lead to more number of leaves. The increase in number of leaves enhances the amount of photosynthetic material and contributed to increased root biomass in turn increases nutrient content, uptake, growth and yield attributes of maize. Through several research papers, it was confirmed that supply of nutrients in nano form had positive effect on growth parameters, yield parameters and yield of crops because nano fertilizers influence the physiology of plants by positively increasing the root biomass to efficiently absorb the nutrients from the rhizosphere in turn, contributed to increased growth and yield attributes.

### Kernel and Stover Yield

The kernel and stover yield of maize (Fig.1) varied significantly with the application of P from conventional and nano sources. Significantly higher kernel yield of 73.59 q ha<sup>-1</sup> was recorded in T<sub>9</sub> treatment compared to control (55.56 q ha<sup>-1</sup>), T<sub>2</sub> (62.78 q ha<sup>-1</sup>), T<sub>7</sub> (65.63 q ha<sup>-1</sup>) and T<sub>8</sub> (65.59 q ha<sup>-1</sup>) treatments and was at par with all other remaining treatments tried.

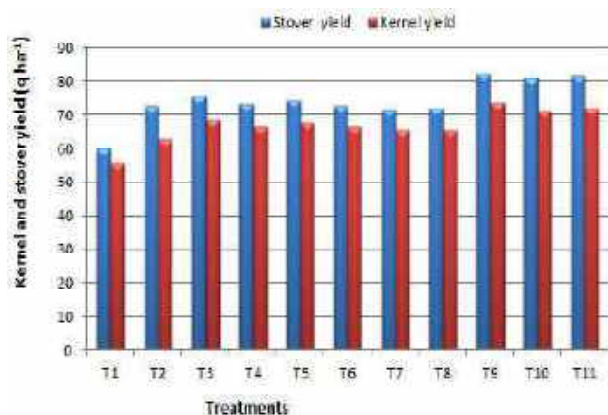


Fig.1 : Stover and kernel yield (q ha<sup>-1</sup>) of maize as influenced by the application of nano P fertilizers

In case of stover yield also, significantly highest value (82.23 q ha<sup>-1</sup>) was recorded in T<sub>10</sub> treatment than all other treatments except T<sub>10</sub> (80.89 q ha<sup>-1</sup>), T<sub>11</sub> (81.73 q ha<sup>-1</sup>) and T<sub>3</sub> (75.61 q ha<sup>-1</sup>) and lowest stover yield value (59.92 q ha<sup>-1</sup>) was recorded in control.

Compared to control, application of 75 per cent RDP through SSP along with 5 per cent RDP through soil application of hydroxyapatite nano fertilizer (NP1) and / or 1 per cent RDP through foliar application of NP1 recorded higher maize yield than that was recorded with 100 per cent RPP applied treatment. So, the yield obtained with soil and / or foliar application of nano P fertilizers along with conventional P sources is equivalent or more than that was obtained with 100 per cent RPP applied treatment. The yield attributes and yield of maize were gradually increased in all the treatments where 25 per cent RDP from conventional sources was substituted with nano P fertilizers in different proportions (5% RDP as soil application and / or 1% RDP as foliar spray) So, application of 5 per cent RDP through soil application nano P fertilizers and / or 1 per cent RDP through foliar spray of nano P fertilizers can replace 20 per cent RDP through conventional P source (SSP). Hence, 25 per cent RDP through conventional sources can be substituted with 5 per cent RDP through nano P fertilizers which saves nearly 25 per cent conventional P sources.

The higher yield in nano phosphorus fertilizers applied treatments might be attributed to the increased cob length, number of rows per cob, number of kernels per cob and test weight. The improvement in yield and yield parameters was due to the improvement in growth parameters (Table 1 and 2). The improvement in growth parameters was due to synchronized supply of P and Ca contained in NP1. Through several research papers, it was confirmed that supply of nutrients in nano form had positive effect on growth and yield parameters because nano fertilizers influence the physiology of plants by positively increasing the root biomass to efficiently absorb the nutrients from the rhizosphere soil (Rajendran *et al.*, 2017). Adhikari *et al.* (2014) also confirmed that application of nano phosphorus

fertilizer had enhanced the maize grain yield by 44.68 per cent and stover yield by 13.17 per cent over the control. These results were corroborated with findings of Ekinçi *et al.* (2014) in cucumber; Liu and Lal (2014) in soybean; Abdel-Aziz *et al.* (2016) in wheat; Harish and Gowda (2017) in groundnut; Babubhai (2018) in maize and Khanm *et al.* (2018) in tomato and Rathnayaka *et al.* (2018) in rice.

Application of 75 per cent recommended dose of phosphorus (RDP) through conventional P fertilizer (SSP) along with soil application of 5 per cent RDP through hydroxyapatite nanoparticles (NP1) and 1 per cent of foliar application of RDP through NP1 recorded highest growth parameters, yield attributes and yield of maize plant compared to 100 per cent RDP applied through conventional fertilizer treatment. This results in saving of nearly 25 per cent of P fertilizer application through conventional sources. From this study, it is confirmed that application of phosphorus in nano form will increase the use efficiency of P, in turn contributed to higher growth and yield of maize.

TABLE 4

Effect of Nano P fertilizers on stover and kernel yield (q ha<sup>-1</sup>) of maize

Treatments	Stover yield	Kernel yield
T <sub>1</sub> : Control	59.92	55.56
T <sub>2</sub> : 100% RPP	72.73	62.78
T <sub>3</sub> : 75% RDP+5 % RDP - NP1	75.61	68.56
T <sub>4</sub> : 75% RDP+5 % RDP - NP2	73.35	66.43
T <sub>5</sub> : 75% RDP+5 % RDP - NP3	74.29	67.41
T <sub>6</sub> : 75% RDP+ FS of 1 % RDP - NP1	72.98	66.52
T <sub>7</sub> : 75% RDP+FS of 1% RDP -NP2	71.53	65.63
T <sub>8</sub> : 75% RDP+FS of 1% RDP -NP3	72.17	65.59
T <sub>9</sub> : T <sub>3</sub> + FS of 1% RDP - NP1	82.23	73.59
T <sub>10</sub> : T <sub>4</sub> +FS of 1% RDP - NP2	80.89	71.11
T <sub>11</sub> : T <sub>5</sub> +FS of 1% RDP - NP3	81.73	71.96
S.Em ±	2.33	2.56
CD @ 5%	6.88	7.57

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## Climate - Resilient Technology to Adapt to Climate Change for Sustainable Livelihood and Production

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### ABSTRACT

In the five most climatically vulnerable districts of Alappuzha in Kerala and Gadag, Kalaburagi, Tumakuru and Chikkaballapura in Karnataka, climate resilient technologies such as the selection of climate resilient varieties, location specific intercropping systems and goat management were demonstrated during 2021. Selection of improved varieties viz., finger millet (ML-365), maize (MAH-14-5), sorghum (SPV 2217) and pigeonpea (BRG-5) performed better with yield of 20.30 q/ha, 27.10 q/ha, 22.75q/ha and 9.30 q/ha, respectively than local varieties. Ground nut + pigeonpea (13.40 q/ha), maize + pigeonpea (71.95 q/ha), green gram + pigeonpea (8.63 q/ha), pigeonpea + black gram (12.25 q/ha) and finger millet + pigeonpea (20.7 q/ha) intercropping systems recorded significantly higher yields compared to their sole crops (12.57 q/ha, 53.97q/ha, 6.25 q/ha, 4.90 q/ha and 20.1 q/ha, respectively). Improved goat shelter with raised platform, reduced the mortality from 40 per cent to 0 and increased number of kids per year (17 kids/ year) with net return of Rs.57345/- and B:C ratio of 1.7 compared to traditional goat rearing shelter.

**Keywords :** Climate change, Climate resilient, Climate vulnerable, Intercropping system

CLIMATE change and its variability have emerged as serious concerns to Indian agriculture in recent years. Global climate change projections include increased extreme events (e.g., heat and cold waves, flooding), increased atmospheric carbon dioxide and ground-level, ozone concentrations and a rise in sea level that will inundate coastal areas, among other things (Raghavan *et al.*, 2020). Climate change can reduce agricultural income by 15 to 25 per cent. Hence, it is right time to value and execute climate-resilient agriculture more rigorously. Planned approaches to adaptation in agriculture and development practices are necessary to cope with climate change and make agri-production resilient to climate changes and shocks. India has a diverse ecology and some regions have evolved and adapted practices over time to tackle vagaries.

Judicious use of some of these practices has the potential to mitigate the effects of climate change. Proper management and implementation of practices that have resulted in an increased agri-produce in unfavourable conditions can also be used to adapt to climate change. These practices lead to increased resilience and consistency in yield despite varying climatic conditions.

Climate-resilient crops and crop varieties have enhanced tolerance to biotic and abiotic stresses. They are intended to maintain or increase crop yields under extreme weather conditions and thereby provide a means of adapting to diminishing crop yields in the face of droughts, higher average temperatures and other climatic conditions (Maricelis Acevedo *et al.*, 2020). Adoption of

climate-resilient crops, such as short duration crop varieties, heat-tolerant varieties, drought tolerant and resistant cereal, legumes or varieties with enhanced salinity tolerance or rice with submergence tolerance, can help farmers to better cope with climate shocks.

Intercropping is an important aspect to combat the crop failure in rainfed agriculture under the situation of climate change and helps in improving productivity and profitability through efficient utilization of natural resources. Intercropping provides insurance against drought, modifies soil environment, improves moisture and radiation use, ensures better weed control, reduces disease and pest incidence and on the whole increases and stabilizes the productivity. Intercropping has been identified as a kind of biological insurance against risks under aberrant rainfall behavior. Crop diversification is also necessary to get higher yield and return besides maintaining soil health apart from other benefits (Siddique *et al.*, 2012)

By adopting new technology and innovative measures for crops and livestock production, farmers are more inclined to adapt their sustainable livelihoods to mitigate the impact of precipitation deficits and climate shocks. It ensures the farmers' income becomes more resilient, not only from the short-term climate shocks but also in the long run by producing crops that are resilient to drought and weather variability (Adiqa Kausar Kiani *et al.* 2021). In this regard, study was undertaken to evaluate different climate resilient technologies *viz.*, improved and

drought resistance varieties, location specific intercropping system and livestock management technology in selected NICRA villages of Karnataka and Kerala.

## MATERIAL AND METHODS

The participatory trials were undertaken in farmers' fields during 2021 under the 'National Innovations in Climate Resilient Agriculture' (NICRA) project which is in operation in five most climatically vulnerable districts namely Alappuzha in Kerala and Gadag, Kalaburagi, Tumkuru, Chikkaballapura in Karnataka. Table 1 shows the villages that were chosen for the study, as well as the soil types, normal rainfall and climatic vulnerabilities.

The demonstration of climate resilient varieties, location specific intercropping system and goat management conducted in selected farmer's field. Fields were selected based on the willingness of farmers to engage in participatory research to evaluate the science based strategy. Selection also ensured trials with all prominent crops in the domain. Capacity building of selected farmers was undertaken through repeated trainings in multi-disciplinary approach. Selected farmers participated in each and every research intervention like soil sampling, input application and yield estimation.

### Climatic Conditions

During 2021, among the climate vulnerable districts the highest annual rainfall was recorded

TABLE 1  
Selected NICRA village information

NICRA village	Taluk and District	Annual rainfall (mm)	Soil type	Climate variability
Edathua	Alappuzha, Kerala	2928.3	Clayey alluvial	Flood/water inundation
Suntanoor	Aland taluk, Kalaburgi	782.9	Medium deep black clayey soil	Drought
Chikkadoddavadi	Korategere taluk, Tumkuru	697	Red sandy soil	Drought
Singatarayanakeri	Mundaragi taluk Gadag	641.6	Red gravel	Drought
Hanumaigarahalli	Chintamani taluk, Chikkaballapura district	703.2	Red loamy soil	Drought

TABLE 2  
Climatic conditions of the NICRA village

NICRA village	Rainfall (mm)		Rainy days	Dry spell	Intensive rain (> 60 mm)
	Normal	Actual			
Edathua, Alappuzha, Kerala	2928.3	3610.2	109	-	8
Suntanoor, Aland taluk, Kalaburgi	782.9	1179.1	53	1	4
Chikkadoddavadi Korategere taluk, Tumkuru	697.0	1097	252	6	3
Singatarayanakeri, Mundaragi taluk Gadag	641.6	612.8	33	5	2
Hanumaigarahalli, Chintamani taluk, Chikkaballapura district	703.2	1322.4	57	-	2

in NICRA village of Alappuzha district of Kerala with 3610.2 mm (109 rainy days) as against normal rainfall of 2928.3 mm. Eight intensive rain spells of more than 60 mm occurred during the months from June to December. In NICRA village of Kalaburgi district received an annual rainfall of 1179.1 mm as against normal rainfall of 782.9 mm and Tumkur district received annual rainfall of 1097 mm with six dry spells and also three intensive rain spells of more than 60 mm occurred in the months of October and November. In Gadag, 5 dry spells with annual rainfall of 612.80 mm (33 rainy days) was recorded as against normal rainfall of 641.60 mm in Singatarayanakeri village. In total it was 4.67 per cent deficit from the normal rainfall. Hunumaigarahalli village, Chikkaballapura district recorded 1322.4 mm with 57 rainy days as against normal rainfall of 703.21mm, with two intensive rain spells of more than 60mm occurred (Table 2).

### Climate Resilient Varieties

Farmers are still growing local and long duration varieties which are low productive and often crop failures are experienced either due to rainfall extremes or disease occurrence. During 2021, high yielding short duration varieties and drought resistant varieties of finger millet, sorghum, pigeonpea and maize were demonstrated in 63 farmer's field covering on area of 32.5 ha at climatically vulnerable districts of Karnataka (Tumkur, Gadag, Kalaburgi and Chikkaballapura).

### Improved Intercropping System

Famer's practice sole cropping but is risky and often results in low yields or sometimes even in crop failure due to erratic monsoon rainfall and skewed distribution. Considering climatic and other risk, during 2021, location specific intercrop cropping system like finger millet + pigeonpea, groundnut + pigeonpea, maize + pigeonpea, green gram + pigeonpea and pigeonpea + blackgram were demonstrated in 66 farmers field covering an area of 33.6 ha at climatically vulnerable districts *viz.*, Tumkur, Gadag, Kalaburgi and Chikkaballapura.

### Goat Shelter with Raised Platform

Kuttanad region of Kerala, a unique ecosystem which lies up to 2 m below Mean Sea Level (MSL), is often susceptible to submergence during the South-West monsoon period (June-Sept). In these conditions if farmers follow traditional goat shelters with a flank constructed of country wood stored at ground level will be submerged during June to September and farmers get only 7-8 months/year for goat rearing. Considering these problems, during 2021-22 goat shelter with raised platform and scientific management practices for goat rearing were introduced as technology demonstrations in 13 farmer's field at Edathua, Alappuzha, Kerala. The data on yield, economics and other parameter were recorded adopting a standard procedure.



## RESULTS AND DISCUSSION

### Climate Resilient Varieties

During 2021, a rainfall of 967 mm was received with six dry spells and three intensive rainfall spell (October and November) during cropping season in NICRA village at Tumkuru district. The results showed that the short duration finger millet variety ML-365 recorded an average yield advantage of 20.30 per cent and a benefit cost ratio of 1.84 as compared to other local varieties in the village and high yielding as well as drought resistant maize hybrid MAH-14-5 recorded higher grain yield, net returns and B:C ratio (27.1q/ha, Rs.18688/ha and 1.77, respectively) as compared to local varieties (20.9q/ha, Rs.9842 and 1.42). The yield and economics of medium duration varieties were higher than other local and long duration varieties due to medium and short duration varieties ability to avoid the heavy rains that occurred in November. Ramachandrappa *et al.* (2016) also reported the similar results (Table 3).

Chikkaballapura NICRA village had 384.9 mm and 275.7 mm of rainfall in October and November of 2021, respectively. Pigeonpea variety BRG-5 performed better in severe rainfall conditions with an average grain yield of 9.13q/ha and higher net returns (Rs.26810/ha) and B:C ratio (7.86) than other local varieties in the NICRA village (Table 3).

NICRA village at Kalaburgi, during the cropping season received 1179.1 mm rainfall with one dry spell (> 20 days) in August and four intensive rainfall spell (> 60mm). In *Rabi* season, sorghum variety SPV 2217 recorded higher grain yield (22.75 q/ha), net returns (Rs.37450/ha) and B:C ratio (2.42) as compared to other local varieties (18.50 q/ha).

### Location Specific Intercropping System

Finger millet, pigeonpea, groundnut, maize and green gram are the main crops cultivated in NICRA village in Chikkaballapura, Tumkuru, Gadag and Kalaburgi districts of Karnataka which are affected due to late onset of monsoon followed by dry spell and intensive rainfall at critical crop growth stages (Table 4). Intercropping has been identified as a kind of biological insurance against risks under aberrant rainfall behavior (Thimmegowda *et al.*, 2016) and intercropping system is best crop diversification for livelihood security and resilience to climate variability

In Chikkaballapura district, groundnut + pigeonpea (8:2) intercropping systems recorded higher groundnut equivalent yield 13.40 q/ha with higher benefit cost ratio (1.61). The increase in yield might be due to no or low competition between main crop and intercrop for growth, development and for above ground and below ground resources as groundnut crop was of shorter duration and non-spreading nature and further, might be due to complementarity

TABLE 3  
Performance of climate resilient varieties in different climate vulnerable districts of Karnataka

Climate vulnerable district	Crop	Treatment	Area (ha)	Farmers (No.)	Yields (q/ha)	Increase (%)	Net return (Rs./ha)	B:C
Tumkur	Finger millet	ML-365	11.0	9	20.3	20.83	20785	1.84
	Local variety		1	2	16.8	-	13670	1.57
Tumkur	Maize	MAH-14-5	2.0	3	27.1	29.67	18688	1.77
	Local variety		0.5	1	20.9	-	9842	1.42
Kalaburgi	Sorghum	SPV 2217	12.0	30	22.75	22.97	37450	2.42
	Local variety		2	10	18.50	-	26750	2.07
Chikkaballapura	Pigeonpea	BRG-5	4.0	8	9.30	14.11	26810	1.86
	Local variety		1	3	8.15	-	20580	1.68

TABLE 4  
Performance of improved intercropping system in different climate vulnerable districts of Karnataka

Climate vulnerable district s	Treatment	Area (ha)	Farmers (No.)	MCEY (q/ha)	Increase (%)	Net return (Rs./ha)	B:C
Chikkaballapura	Groundnut + pigeonpea	8.0	15	13.40	6.60	64356	1.61
	Groundnut	6	10	12.57	-	17372	1.40
Gadag	Maize+ pigeonpea	6.0	15	71.95	33.31	54542	2.18
	Maize	2.0	5	53.97	-	36327	1.93
Gadag	Greengram + pigeonpea	2.0	5	8.63	38.08	32403	2.27
	Greengram	1.0	4	6.25	-	21100	2.01
Kalaburgi	Pigeonpea + blackgram	10.0	25	12.25	150.00	35250	1.81
	Pigeonpea	2.0	6	4.90	-	12850	1.67
Tumkur	Finger millet + pigeonpea	7.6	6	20.7	2.99	21457	1.85
	Finger millet	10	12	20.1	-	20466	1.83

MCEY = Main crop equivalent yield

in resource utilization by groundnut crop (Ramesh and Devasenapathy, 2007).

Diversification of maize and green gram cropping based systems by intercropping with pigeonpea may foster productivity and resilience to adverse weather conditions. Maize + pigeonpea and greengram + pigeonpea recorded higher maize (7.95q/ha) and greengram (8.63 q/ha) equivalent yield with higher net return and B:C ratio compared to their sole crops in NICRA village at Gadag district. Leah L.R. Renwick *et al.*, 2020 reported that, maize + pigeonpea was the only intercrop that consistently required less land than its sole maize to produce the same yield particularly under drought. Despite intercropping systems having greater planting density than sole maize and theoretically greater competition for water, they were not more prone to yield loss with drought.

Pigeonpea + blackgram in 1:3 ratio recorded higher pigeonpea equivalent yield, net return and B:C ratio (12.25 q/ha, 35250/ha and 1.81 respectively) as compared to sole pigeonpea (4.90 q/ha, 12850/ha and 1.67, respectively) in Kalaburgi district NICRA village. Sole pigeonpea growth has been drastically reduced as a result of heavy rainfall (194.6) in October, which caused water stagnation in crop

fields, resulting in a decreased crop yield but inter crop of pigeonpea + blackgram reduced the risk of that extreme weather condition. Kathmale *et al.*, (2014) reported that, the legumes as intercrops act as cover crops in wider row spaced pigeonpea resulting in higher *in-situ* moisture conservation and efficient utilization by both the component crops, further more helping in increased pigeonpea equivalent yields.

Under NICRA villages of Chikkadoddavadi Korategere taluk, Tumkuru, intercropping of finger millet + pigeonpea (8:1) recorded higher finger millet grain equivalent yield of 20.7 q/ha with higher net return (Rs.21457/ha and B:C ratio (1.85) than sole finger millet cropping system (20.1q/ha, Rs.20466/ha and 1.83, respectively). This was attributed to the better performance of small millets even under more than 30 days dryspell and erratic rainfall during crop growth period, both as sole crop and intercrop probably due to their drought tolerance (Shashidhara *et al.*, 2000). Adikant Pradhan *et al.* (2014) and Santosh Nagappa Ningoji *et al.* (2021) reported that finger millet intercropping system recorded the better yield and monetary returns as compared to the solecrop.

TABLE 5  
Performance goat shelter with raised platform in NICRA village Alappuzha district, Kerala

Intervention	Unit	Farmers (No.)	No. kid / year	Increase (%)	Net return (Rs./ha)	B:C
Goat shelter with raised platform	13	13	17	112.50	57345	1.7
Tradition goat shelter	4	4	8	-	4419	1.1

### Goat Shelters with Raised Platforms

Heavy rains followed by an unprecedented flood caused devastating effects in all low-lying areas, with most livestock shelters submerged or destroyed. However, shelter management for small ruminants to tackle flood condition the construction of goat shelters with raised platforms helped to overcome this difficulty. Disease outbreak was reduced by following these practices and improved shelter, reducing the mortality from 40 per cent to 0. This resulted increased number of kids per year (17 kids/year) with net return of Rs.57345 and B:C of 1.7 compared to traditional goat rearing shelter (8 kid/year, Rs.4419 and 1.1, respectively) (Table 5). Ravi *et al.*, (2022) stated that higher body weight gain was observed in the 6 month old male kids of Marwari breed, when reared under closed type improved animal shelter during summer months. Kids gained 9.52 kg body weight in improved shelter compared to 7.52 kg in traditional shelter during 6 months period of experiment from May to October.

Climate resilient technologies for aberrant rainfall situations play a crucial role in agriculture for sustaining the productivity and livelihood of farmers. Selection of variety according to the weather condition and finger millet + pigeonpea, groundnut + pigeonpea, pigeonpea + blackgram, maize + pigeonpea and greengram + pigeonpea inter cropping systems would enhance the productivity and economic benefits to the dryland farmers. Goat shelters with raised platforms aided in overcoming flood-related difficulties.

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## Seed Morphometric Changes Influenced by Accelerated Ageing in Contrast-coloured Maize (*Zea mays* L.) Genotypes

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### ABSTRACT

Accelerated ageing induces many deteriorative changes to seeds which ultimately lead to reduced germination and loss of viability. Taking advantage of the image analysis, the seed morphometric changes after accelerated ageing was investigated. Three contrasting colour maize genotypes *viz.*, African tall (white), MAH 14-5 (orange) and local land race (red) selected were artificially aged for 96 and 120 hours. The aged seeds showed 100 per cent viability but reduced germination per cent due to the increased fraction of abnormal seedlings. The parameters like area, diameter, perimeter, roundness increased, elongation decreased while roughness and compactness remained constant in artificially aged seeds. Area increase in 96 and 120 hours of ageing recorded 18 and 23 per cent in African Tall, 11 and 14 per cent in MAH 14-5 and 9 and 10 per cent in local landrace respectively while the diameter and perimeter showed similar per cent increase. There was a significant difference in morphometric changes between genotypes with white genotype showing a higher increase in size parameters compared to coloured genotypes which may be attributed to impermeability resulting out of oxidation of phenols in coloured genotypes. Lesser the change in morphometric characters after ageing higher was the germination indicating that change in seed morphometric characters during ageing as a deteriorative sign.

**Keywords :** Imbibition, Deterioration, Perimeter, Area

SEED ageing is an irreversible and inexorable process of a progressive decrease in vigour ultimately leading to the loss of seed viability (Stewart & Bewley, 1980 and Lehner *et al.*, 2008). The rate of seed ageing depends upon the genotype/ species, the conditions prevailing during storage like moisture content, temperature, humidity and seed composition (Roberts, 1973). It has been reported that high moisture content and high temperature usually accelerate seed deterioration (Ellis and Hong, 1991; Goel *et al.*, 2003) based on which seeds are accelerated to artificially age by exposing them to high humidity and temperature of about 40 - 45p °C and 100 per cent RH (Delouche and Baskin, 1973). Accelerated ageing is shown to induce many deteriorative changes to seeds during storage like genetic damage, protein

degradation, enzyme inactivation and loss of membrane integrity (Bailly *et al.*, 1996; Merritt *et al.*, 2003; Bailly., 2004; Ratajczak & Pukacka, 2005; Wang *et al.*, 2011 and Ratajczak *et al.*, 2015) which ultimately lead to reduced germination (Walters, 1998) and loss of viability. Though there are numerous studies on physiology, ROS and its mechanisms of membrane degradation during ageing, there is less knowledge about the seed morphometric changes brought about by accelerated ageing.

Ellis *et al.* (1992) reported that the rate of seed deterioration depends upon the moisture content and temperature of storage conditions. Among these two factors, the sensitivity of seeds to high temperatures is strongly dependent on their water content, loss of

viability being faster with increasing moisture content indicating the crucial role of moisture in seed deterioration (McDonald, 1999 and Roberts & Ellis, 1989). In accelerated ageing conditions, the increased humidity (~100% RH) leads to moisture absorption by seeds (ISTA, 2010 and Kapoor *et al.*, 2011). This moisture absorption during ageing would lead to swelling of seeds altering the seed size parameters. So, the morphometric changes like seed perimeter, area, diameter, roundness, roughness, elongation and compactness after ageing can be effectively captured by image analysis which in turn would indicate the rate of seed deterioration and help in varietal identification (Nethra *et al.*, 2005). A strong relation between water absorption and change in seed perimeter measured by an image analyser was established by Satya Srii *et al.* (2020). Previous studies (Renuka *et al.*, 2022) have reported the use of morphological differences between parental lines in hybrid seed production to help characterization, however use of seed morphometric characters to study imbibition during ageig is unexplored.

Seed image analysis works based on the extraction of numerical data from a captured image of seeds and seedlings and their subsequent data processing with the help of suitable computer software (Hemender *et al.*, 2018). The major advantage of image analysis over other conventional methods is the easy determination of dimensional changes in time without any manipulation of the seeds (Tanabata *et al.*, 2012) and its capacity to detect even the smallest changes in seed dimensions (Dell'Aquila *et al.*, 2000; Dell'Aquila, 2005). Taking advantage of the image analysis procedure, in this study we investigated the changes in seeds morphometric parameters like area, diameter, perimeter, roughness, elongation, roundness and compactness after subjecting seeds to accelerated ageing. As the rate of moisture absorption during ageing depends upon the initial moisture content of seeds and the genotype, we studied morphometric changes in three contrast coloured maize genotypes (white, orange and red) in response to accelerated ageing conditions *i.e.*, genotypic difference in moisture absorption ie morphometric changes and ultimately in deterioration.

## MATERIAL AND METHODS

### Seed Material

The three contrasting colour maize genotypes African tall fodder maize (white), MAH 14-5 (Orange) and local landrace from Tamil Nadu, India (red) were selected for the study and the fresh seeds of African fodder maize and MAH 14-5 were received from Seed Stores, National Seed Project, University of Agricultural Sciences, Bangalore, India and local red landrace was collected from Maize Research Station, Vagarai, Tamil Nadu, India. The fresh seeds were checked for optimum moisture content and stored at -20p °C until further use.

### Accelerated Ageing

Artificial ageing was performed according to ISTA guidelines (ISTA, 2010). The moisture content of the samples was determined and those with optimal moisture content between 10-14 per cent moisture was kept for accelerated ageing. For artificial ageing (AA), the plastic AA boxes were first sterilized with 5 per cent sodium hypochlorite and dried then, each AA box was filled with  $40 \pm 1.0$  ml of distilled water. Seeds were placed on the screen one layer deep to ensure an even uptake of moisture from the humid environment and the lid was placed on each plastic AA box. These AA boxes were placed on the shelves of the ageing chamber (Manufacturer: Thermo Scientific, Model: IGS 60/100/180) allowing air space of 2.5 cm between plastic AA boxes to assure temperature uniformity. The temperature was set to  $41 \pm 0.3p$  °C with 100 per cent RH and was monitored at regular intervals. After 96 hours and 120 hours of ageing, the AA boxes were removed from the chamber. The control seeds were stored in sealed, optimum storage conditions for the same duration as of ageing.

### Germination Per cent and Viability

Seed germination per cent and viability of the aged seeds along with control (fresh, non- aged seeds) were measured using standard ISTA protocol (ISTA, 2010) to confirm the process of ageing. Seed germination test was performed for 100 seeds in 4 replicates for each ageing treatment and control by between paper

method. The viability testing of seeds was performed for 50 seeds in 2 replicates for each ageing treatment using Tetrazolium (Tz) staining. Tz staining was performed by soaking the seeds in water for 12 hours followed by cutting the seeds longitudinally through the embryo and  $\frac{3}{4}$  of the endosperm and then soaking the seeds in 1 per cent Tz solution for 2 hours and evaluating the uptake of stains.

### Image Analysis for Seed Morphometric Characters

Seed morphometric studies were performed using the image analysis approach. Two replicates of 25 seeds in each ageing treatment and control were taken for the study. Seed morphometric parameters like area, diameter, perimeter, roundness, compactness, elongation and roughness of the individually labelled seeds in Petri dishes was measured using an image analyser (Manufacturer: Expert Vision Labs Private Limited, Model L-2000 in conjunction with biovis software) and recorded as seed morphometric characters before ageing, then the seeds were subjected to accelerated ageing and storage in case of control for different periods after which the seeds were again analysed through image analysis. The parameters like roughness, compactness, elongation and roughness of seeds were measured by specifically developed macros based on image analysis library and formulas (Varma *et al.*, 2013).

### Statistical Analysis

Descriptive statistics were performed using Microsoft Excel 2010. Significance of values of different parameters before and after ageing within a genotype was performed using Paired t-Test (paired two samples

for mean) in Microsoft Excel 2010. The significance of the difference in values between genotypes and also between two ageing periods was analysed individually using SPSS software (ANOVA with single factor). Correlation between various parameters was confirmed using the  $r^2$  value calculated using Microsoft Excel 2010 in which most parameters showed strong correlation ( $r^2 > 0.7$ ) with few having moderate correlation ( $0.5 < r^2 < 0.7$ ). Per cent increase/decrease in values of each parameter before and after ageing was calculated which was used to plot a radar plot using Microsoft Excel 2010.

## RESULTS AND DISCUSSION

### Seed Physiological Quality After Ageing

The germination per cent, viability per cent of three genotypes of seeds aged for different time intervals along with control evaluated as per ISTA protocol (ISTA, 2010) is given in Table 1. One way ANOVA was used to test the significance of data and there was a significant difference ( $P < 0.01$ ) in seed quality parameters between genotypes and between different ageing treatments.

Ageing treatments of 96 and 120 hours were selected after standardising viability at different periods of ageing, to have seeds that are still viable but less vigorous *i.e.*, at the initial stages of deterioration to study the initial changes brought about by ageing to morphometric characters of seeds which might, in turn, correlate to deteriorative changes. Results of viability and germination tests showed that all three genotypes remained viable after 96 and 120 hours of ageing yet there was a decrease in germination

TABLE 1  
Seed quality parameters measured at different ageing periods for two Maize genotypes

Genotypes	Seed germination(per cent)			Viability (per cent)		
	Control	Ageing (96 h)	Ageing (120 h)	Control	Ageing (96 h)	Ageing (120 h)
African Tall (white)	100 ± 0.00 **	90 ± 0.83 **	84 ± 0.70 **	100 ± 0.00	100 ± 0.00	100 ± 0.00
MAH 14-5 (orange)	100 ± 0.00 **	93 ± 1.22 **	88 ± 0.44 **	100 ± 0.00	100 ± 0.00	100 ± 0.00
Local landrace (red)	100 ± 0.00 **	96 ± 0.54 **	93 ± 0.70 **	100 ± 0.00	100 ± 0.00	100 ± 0.00

Values are expressed as mean ( $\pm$ SD). \*\* indicates significant difference between ageing times at  $p \leq 0.01$ .

per cent as shown in Table 1. This contradiction in results of viability and ageing is due to the fraction of abnormal seedlings produced by aged seeds which would not be considered as germinated in germination tests as per ISTA test guidelines. Though aged seeds had 100 per cent viability even after ageing the deteriorative changes that occurred due to ageing reflected as abnormal seedlings. It was also reported in peas and soybean that accelerated ageing increased the proportion of abnormal seedlings which reduced the germination per cent of aged seeds (Veselova and Veselovsky, 2003 and Rastegar *et al.*, 2011). But there was a significant difference in germination per cent between genotypes after ageing where African Tall showed the least germination per cent after ageing followed by MAH 14-5 and Local landrace which might be due to varying genetic potential between genotypes. An interesting point is that the darker the colour, the greater the resistance to ageing *i.e.*, coloured genotypes incurred lesser damage compared to colourless genotype due to ageing. The reason for the increased germination after ageing in coloured genotypes could be attributed to the presence of proanthocyanidins with free radical scavenging activity in the seedcoat (Takahata *et al.*, 2001) which would in turn help in cell repair mechanisms preventing membrane damage (Bailly, 2004).

### Seed Morphometric Analysis

Seed morphometric parameters like area, diameter, perimeter, roundness, compactness, elongation and roughness measured for different seed groups revealed that the control (fresh, non-aged) seeds had no change in morphometric characters before and after the storage period while the seeds subjected to ageing showed a significant difference in values after ageing. The parameters like area, diameter, perimeter, roundness increased, elongation decreased while roughness and compactness remained constant when the seeds were artificially aged. Data recorded for various parameters before and after ageing along with percent change in parameter after ageing for three genotypes are presented in Table 2, 3 and 4. This data (Fig. 1) shows that there was a

TABLE 2  
Seed morphometric parameters of white maize (African Tall- fodder maize) measured before and after ageing in T1 and T2 along with percent change in parameters

Parameters	Before ageing	After ageing	Per cent change
<i>T1 (white maize)</i>			
Area square (cm)	0.73831	0.86961	18%
Diameter (cm)	0.87641	0.959068	9%
Perimeter (cm)	2.98482	3.257892	9%
Roundness	1.03697	1.111904	7%
Roughness	1.03261	1.033192	0%
Elongation	1.25762	1.225448	-3%
Compactness	12.1804	12.35156	1%
<i>T2 (white maize)</i>			
Area square cm	0.78291	0.962952	23%
Diameter cm	0.875407	0.986072	13%
Perimeter cm	2.87533	3.245132	13%
Roundness	1.02521	1.150136	12%
Roughness	1.03253	1.033884	0%
Elongation	1.240747	1.17448	-5%
Compactness	12.29995	12.47616	1%

TABLE 3  
Seed morphometric parameters of orange maize (MAH 14-5) measured before and after ageing in T1 and T2 along with percent change in parameters

Parameters	Before ageing	After ageing	Per cent change
<i>T1 (Orange maize)</i>			
Area square cm	0.51764	0.575841	11%
Diameter cm	0.7081	0.759013	7%
Perimeter cm	2.36762	2.5442125	7%
Roundness	1.17417	1.218283	4%
Roughness	1.02265	1.023017	0%
Elongation	1.12385	1.113441667	-1%
Compactness	10.88918	10.90508	0%
<i>T2 (Orange maize)</i>			
Area square cm	0.55045	0.625274	14%
Diameter cm	0.739703	0.802678	9%
Perimeter cm	2.48067	2.708144	9%
Roundness	1.122687	1.196872	7%
Roughness	1.023893	1.023856	0%
Elongation	1.115287	1.079171722	-3%
Compactness	11.22109	11.16935	0%



TABLE 4

Seed morphometric parameters of red maize (local landrace) measured before and after ageing in T1 and T2 along with percent change in parameters

Parameters	Before ageing	After ageing	Per cent change
<i>T1 (Red maize)</i>			
Area square cm	0.501706	0.546908	9%
Diameter cm	0.707217	0.738156	4%
Perimeter cm	2.39521	2.490916	4%
Roundness	1.099223	1.100836	0%
Roughness	1.03078	1.02716	0%
Elongation	1.119803333	1.119312	0%
Compactness	11.49296	11.45215	0%
<i>T2 (Red maize)</i>			
Area square cm	0.514243	0.5644548	10%
Diameter cm	0.714353	0.759934	6%
Perimeter cm	2.41172	2.549356	6%
Roundness	1.105403	1.109316	0%
Roughness	1.02853	1.02934	0%
Elongation	1.11161	1.1124916	0%
Compactness	11.40051	11.40193	0%

difference in seed size of different genotypes even before subjecting to ageing where the African Tall (white) were larger followed by MAH 14-5 (Orange) while Local landrace (red) was the smallest among three. Among various parameters, roundness and elongation were seen to be negatively correlated with  $r^2 > 0.5$ . All genotypes recorded similar roughness values which were cross-verified by physical examination of seed surface.

To eliminate exaggeration of increase/decrease in parameters between genotypes, per cent increase was calculated taking into account the initial value and the change in value after ageing for all genotypes. These results of per cent change in morphometric parameters at different periods of ageing for three genotypes are presented in form of a radar chart in Fig. 2, which shows that the increase/ decrease per cent for all parameters was higher in African Tall (white genotype) followed by MAH 14-5 (orange genotype) and least in Local landrace (red). Between ageing periods, 120 hours of ageing recorded more increase/ decrease per cent than 96 hours of ageing in all three genotypes. Area increase in 96 and 120 hours

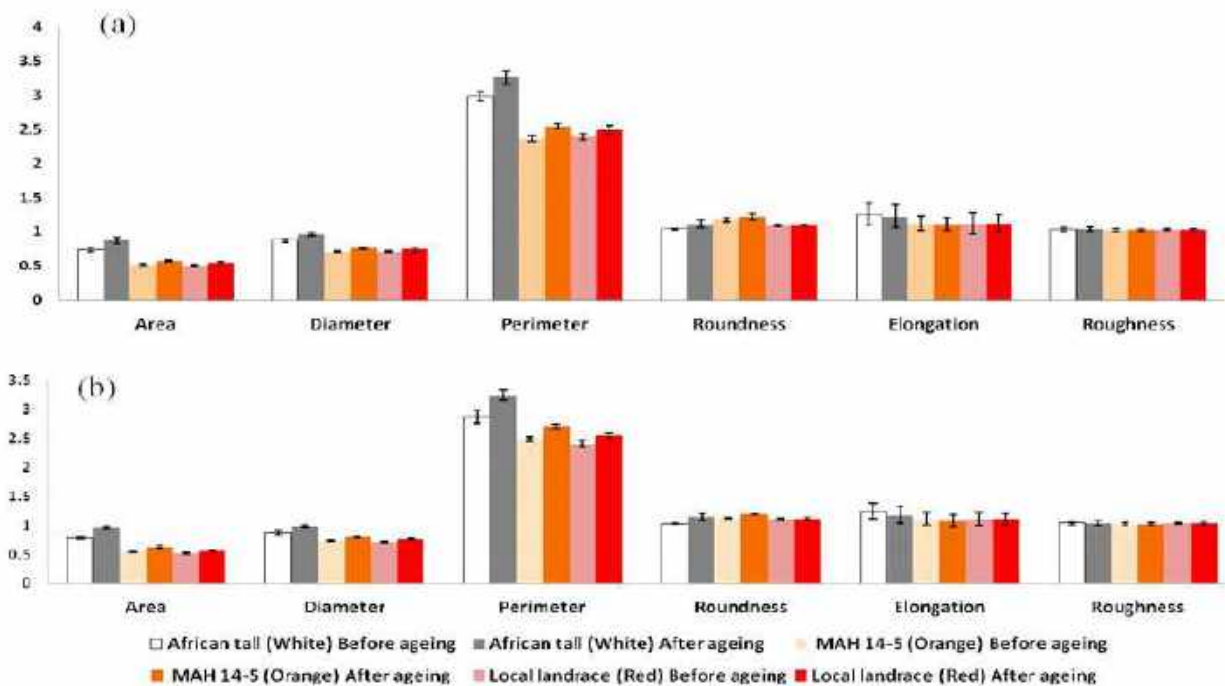


Fig. 1: Morphometric parameters of maize seeds of three genotypes after ageing for (a) 96 hours and (b) 120 hours. Black bar indicates standard error

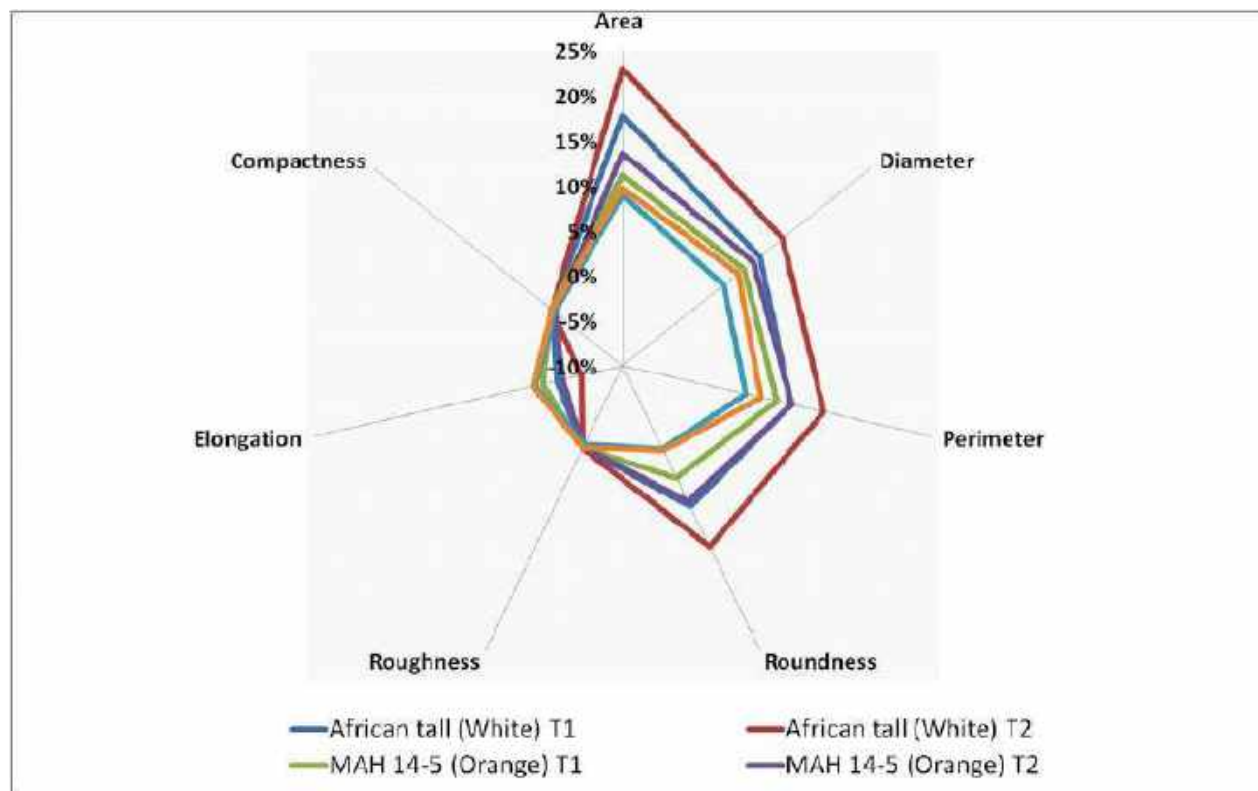


Fig. 2: Per cent change in seed morphometric characters brought about by different periods of ageing in three genotypes

of ageing recorded 18 and 23 per cent in African Tall, 11 and 14 per cent in MAH 14-5 and 9 and 10 per cent in Local landrace respectively. Diameter and perimeter showed the same per cent increase confirming the accuracy of measurements made by image analysis where they increased by 9 and 13 per cent in African Tall, 7 and 9 per cent in MAH 14-5 and 4 and 6 per cent in Local landrace at 96 and 120 hours of ageing respectively.

The paired t-test showed a high level of significance for both single and two-tail tests ( $P < 0.001$ ) for all the parameters measured before and after ageing. Also, there was a significant difference ( $P < 0.01$ ) for values between genotypes tested using ANOVA (single factor). Significant correlations between parameters ( $r^2 > 0.5$ ) showed the accuracy and relevance of values measured by the image analyser.

The increase in size parameters like area, diameter, perimeter in aged seeds is due to the absorption of moisture by seeds in the ageing chamber which is

maintained at 100 per cent RH. There was an increase in area, perimeter and diameter, in turn, reflecting an increase in roundness while the decrease in elongation. This absorption of moisture leads to swelling/ increased size of seeds after ageing and it is seen that seeds absorb more moisture if they are exposed to the ageing chamber for a longer duration which is reflected as the increased per cent of the size increase in seeds that was aged for a longer duration. Absorption of moisture increases the moisture content of seeds which in turn leads to faster deterioration. It is reported that higher moisture content in seeds leads to faster deterioration (Ellis *et al.*, 1992). Results from germination also correlate to the above fact that the genotype which showed a higher increase in size after ageing *i.e.*, more absorption of water had lesser germination per cent. African tall which showed 18 and 23 per cent increase in the area after 96 and 120 hours of ageing had only 90 and 84 per cent germination respectively, on the other hand, local landrace which showed only 9 and 10 per cent increase in the area after 96 and 120 hours

had 96 and 93 per cent germination while MAH 14-5 which had an intermediate increase of about 11 and 14 per cent showed 93 and 88 per cent germination after 95 and 120 hours respectively.

This differential ability of genotypes to absorb moisture may be due to its differential permeability properties. The primary line of protection to seeds against ageing conditions is offered by seed coats as they are the main interface between seed and external environment. It is reported the cracks, cleavages, fissures and scratches resulting either from ageing or genetics or handling procedures (*e.g.*, harvesting, drying, processing and sowing operations) in seed coats could lead to deteriorative changes affecting the embryo and ultimately decreasing the seed longevity. Thus the primary resistance of seeds against deterioration can be attributed to thick impermeable seed coats that could guard the embryo from adverse external conditions (Black & Halmer, 2006 and 205 Brooker *et al.*, 2007). The impermeability of seed coat is proposed to be created by oxidation of phenolic compounds by polyphenol oxidase or peroxidase which could, in turn, provide impermeability to seed coats (Pourcell *et al.*, 2005 and Rajjou & Isabelle, 2008). Also, studies show that phenolic compounds in seed coats contribute to seed longevity by limiting the permeability to oxygen and moisture (Pourcell *et al.*, 2007). This may be one of the reasons why the cultivars with dark seed coats in chickpea (Gvozdeva and Zhukove, 1971), soybean (Shahi and Pandey, 1982), snap bean (*Phaseolus vulgaris*) (Prasad and Weigle, 1976), french bean (Powell, 1986) were found to be less permeable than the light seeded cultivars. Studies show that coloured seed coats are impermeable compared to colourless ones in legumes with pigmentation negatively correlating to permeability (Souza and Marcos-Filho, 2001). The results were again proven in our study where the white genotype showed more moisture absorption compared to orange and red with red absorbing the least moisture. Thus the impermeability resulted from oxidation of phenolic compounds may be one of the reasons for reduced damage and deterioration reflected as higher germination per cent

in local landrace (red) and MAH14-5 (Orange) compared to African Tall (white) after ageing.

Though numerous studies on the effect of artificial ageing on seed physiology, ROS and its mechanisms of membrane degradation, there are no reports of difference in morphometric characters of seeds brought about by artificial ageing. The study reveals that besides physiological, chemical and physical changes, seed morphometric characters also change due to ageing. The increase in size parameters of seeds shows that an increase in moisture content under imposed ageing conditions *i.e.*,  $41 \pm 0.3$  °C with 100 per cent RH is the crucial deteriorating agent in artificial ageing while under natural conditions, there may be many other factors playing role in deterioration. Further studies on the comparison of changes in morphometric characters of natural and artificial ageing would provide deep insight into the mechanisms of the two process.

This study indicates the potential of image analysis to estimate moisture increase in aged seeds by capturing minute changes in seed dimensions. Thus, image analysis can be a better alternative for measuring moisture changes in seeds during storage as conventional moisture estimation methods are either destructive or time taking and are practically impossible for larger lots. Further study and standardisation of the utility of image analysis in moisture estimation would help in the efficient storage and handling of seeds.

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## Effect of Green Synthesized and Chemical Nanoparticles on Seed Quality Parameters in Pigeonpea [*Cajanus cajan* (L.) Millsp.]

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### ABSTRACT

An experiment was conducted to standardize green and chemical nanoparticle concentrations and their effect on seed quality in pigeonpea. Among the green nanoparticles, green zinc oxide @ 1250 ppm recorded maximum seed germination (96.25%), speed of germination (28.64), mean shoot length (29.23 cm), mean root length (24.32 cm), mean seedling length (53.55 cm), seedling dry weight (60.5 mg), seedling vigour index-I (5153), seedling vigour index-II (5823), field emergence (94.50%), total dehydrogenase activity (2.353  $A_{480\text{nm}}$ ) and lowest electrical conductivity (26.49  $\mu\text{S/cm/g}$ ). Among chemical nanoparticles, zinc oxide @ 500 ppm recorded maximum seed germination (95.50%) speed of germination (24.28), mean shoot length (23.48 cm), mean root length (20.24 cm), mean seedling length (43.71 cm), seedling dry weight (56.3 mg/seedling), seedling vigour index-I (4174), seedling vigour index-II (5371), field emergence (92.5%), total dehydrogenase activity (2.021  $A_{480\text{nm}}$ ) and lowest electrical conductivity (31.88  $\mu\text{S/cm/g}$ ), whereas in control lowest seed germination (92.75%), speed of germination (21.63), mean shoot length (19.72 cm), mean root length (19.14 cm), mean seedling length (38.86 cm), seedling dry weight (47.0 mg/seedling), seedling vigour index-I (3575), seedling vigour index-II (4324), field emergence (90%), total dehydrogenase activity (1.773  $A_{480\text{nm}}$ ) and highest electrical conductivity (35.87  $\mu\text{S/cm/g}$ ) was observed. These findings suggest that, seed treatment with green zinc oxide @ 1250 ppm and chemical zinc oxide @ 500 ppm nanoparticles will be helpful to maintain seed quality in pigeonpea.

Keywords : Pigeonpea, Nanoparticle, Seed quality

**P**IGEONPEA [*Cajanus cajan* (L.) Millsp.] is an important pulse crop in India and it belongs to the family *Fabaceae*, which is the major source of dietary protein for most of the vegetarian population and it is backbone of nutritional security of our country. Seed quality is affected by both biotic and abiotic factors, which influence germination, as well as other measures of seed quality which affect the ability of seeds to produce seedlings which can emerge and establish. Pulses are highly susceptible to infestation by bruchids, thus causing severe deterioration in seed quality other than storage losses. Generally, management of stored product pest is done through fumigation and also controlled by synthetic

insecticides, which have many limitations and undesirable side effects. In this regard, nanotechnology has contributed maximum to agro-technological revolution. Treating seeds with green nanoparticles act as a best seed quality enhancement technique, where we can observe faster germination, healthy and vigorous seedlings.

In the 21<sup>st</sup> century green nanotechnology regarded as the upcoming industrial revolution and 4<sup>th</sup> generation seed treatment for improving seed quality traits. Green nanotechnology is use of biological routes such as involving microorganisms and plants for the synthesis of nanoparticles (NPs).

Biosynthesis of plant based support materials has gained much importance as compared to conventional adsorbents due to their plentiful existence, low cost, nontoxic nature, high efficiency as well as environmental friendly in nature, which are considered as green nanoparticles. Green nanoparticles are effectively utilized to control pests and diseases, to promote plant growth and seed storage (Balogun *et al.*, 2020).

The problem of various biosafety issues with respect to the environment, plants, animals and human beings can be minimized to a great extent by utilizing the nano particles derived from biological sources such as proteins and carbohydrates which have low impact on the same system. Several green NPs (ZnO, Ag, MgO, Fe and CuO) have been applied as seed pre-treatment agents. It can internalize the seed coat and support water uptake inside the seeds, could possibly interact with  $\alpha$ -amylase enzyme or act as nanocatalyst; thereby enhancing seed starch degradation for seed germination and seedling growth, also mitigating the detrimental effects of seed ageing and in helps elevated levels of antioxidant enzymes (Khan *et al.*, 2020). Metal NPs have an important role in potentiality to affect the physiological condition and also it can modulate the innate immune system (Sarkar *et al.*, 2020). In view of the above, current research has been undertaken with the objective to evaluate the effect of green synthesized and chemical nanoparticles on seed quality parameters in pigeonpea.

### MATERIAL AND METHODS

The current experiment was conducted by using the freshly harvested seeds from NSP Bangalore during 2020-2021. The seeds are used to study the seed storability experiment by using green synthesized and chemical NP *i.e.*, green zinc oxide (500, 750, 1000 & 1250 ppm), green silica (250, 500, 750 & 1000 ppm), chemical zinc (250 & 500 ppm), chemical silica (250 & 500 ppm) along with spinosad as (4.4 mg/kg seed) and control (without any seed treatment) with 4 replications by using CRD. Nanoparticles concentrations were standardized after one month of seed treatment and also to study

the nanoparticle concentration to increase the seed storability.

### Seed Treatment

Seed treatment was done through artificial seed treater in seed processing unit at Nongwoo Seed India Private Limited, Yelahanka New Town, Bangalore. Stock solutions were prepared in 50 ml falcon tubes as per treatments scheduled and solution was poured in seed bin of seed treater for uniform distribution of solution to all seeds in the form of dry treatment. After the seed treatment, the seeds were incubated for a short period to achieve equilibration and treated seed were kept in cloth bag under ambient room temperature for further observation. After one month, all the seed quality parameters were evaluated as per ISTA (2013).

The following parameters were studied from the experiment to standardize the nanoparticle based seed treatment *viz.*, Seed germination (%), Speed of germination, Mean shoot length (cm), Mean root length (cm), Mean seedling length (cm), Mean seedling dry weight (mg/seedling), Seedling Vigour Index-I (SVI-I), Seedling Vigour Index-II (SVI-II), Field emergence (%), Electrical conductivity of seed leachate ( $\mu\text{S}/\text{cm}/\text{g}$ ) and Total dehydrogenase activity ( $A_{480\text{nm}}$ ).

### RESULTS AND DISCUSSION

Standardization of nano particle concentrations and their effect on seed quality parameters were studied.

TABLE 1  
Initial seed quality parameters of Pigeonpea cv. BRG-5

Seed Quality Parameters	
Initial seed moisture content (%)	9.16
Seed Germination (%)	96.00
Seedling shoot length (cm)	19.38
Seedling root length (cm)	20.50
Mean seedling length (cm)	39.88
Seedling vigour index-I	3868
Seedling vigour index-II	4462
Electrical conductivity ( $\mu\text{S}/\text{cm}/\text{g}$ )	25.66
Total dehydrogenase activity ( $A_{480\text{nm}}$ )	2.013

Immediately after collection of seed material the seed quality parameters were analyzed without any seed treatment (Table 1). One month after seed treatment (Table 2), seed moisture content does not vary much (9.10-9.14%) among concentrations treated compared to initial moisture content.

Seed germination was recorded at 96.00 per cent immediately after harvest. During standardization of concentrations green zinc recorded better (96.25%) in maintaining the seed germination after one month of seed treatment compared to control (92.00%) which was significantly different (Table 2). Among chemical nanoparticles higher germination was

recorded with seeds treated with zinc oxide @ 500 ppm (95.50%) followed by silicon dioxide @ 250 ppm (93.50%).

Field emergence significantly differed among concentrations used. Green zinc oxide @ 1250 ppm (94.50 %) is on par with green silicon dioxide @ 750 ppm (94.00 %), which is followed by green zinc oxide @ 1000 ppm (93.25 %) in recording field emergence. Among chemical NPs highest field emergence recorded in chemical zinc oxide @ 500 ppm (92.50 %) compared to the spinosad (91%) and control (90 %) (Table 2).

TABLE 2

Influence of seed treatment with nanoparticles on seed moisture content, germination, field emergence, speed of germination, seedling shoot length, seedling root length and mean seedling length in pigeonpea cv. BRG-5

	Seed Moisture Content (%)	Germination (%)	Field Emergence (%)	Speed of Germination	Seedling Shoot Length (cm)	Seedling Root Length (cm)	Mean Seedling Length (cm)
C <sub>0</sub>	9.14	92.75	90.00	21.63	19.72	19.14	38.86
C <sub>1</sub>	9.12	93.25	90.75	22.97	22.02	18.02	40.04
C <sub>2</sub>	9.13	93.50	92.25	23.71	23.80	20.00	43.80
C <sub>3</sub>	9.11	94.75	93.25	27.51	27.03	22.48	49.51
C <sub>4</sub>	9.11	96.25	94.50	28.64	29.23	24.32	53.55
C <sub>5</sub>	9.13	93.00	90.50	21.26	22.10	19.39	41.49
C <sub>6</sub>	9.10	96.00	93.25	25.98	22.30	22.11	44.41
C <sub>7</sub>	9.13	95.50	94.00	26.17	26.03	21.74	47.77
C <sub>8</sub>	9.14	93.50	91.50	23.51	21.83	20.47	42.29
C <sub>9</sub>	9.13	93.00	92.25	22.71	22.00	19.63	41.63
C <sub>10</sub>	9.11	95.50	92.50	24.28	23.48	20.24	43.71
C <sub>11</sub>	9.12	93.50	90.50	22.61	23.19	19.88	43.07
C <sub>12</sub>	9.14	94.00	90.75	24.29	22.14	18.86	41.00
C <sub>13</sub>	9.12	93.25	91.00	24.91	22.80	18.18	40.98
Mean	9.12	94.13	91.93	24.30	23.41	20.32	43.72
SEm (±)	0.03	0.78	0.79	0.56	0.58	0.50	0.83
CD@0.01	0.10	2.97	3.02	2.13	2.21	1.91	3.17
CV (%)	0.59	1.66	1.72	4.58	4.95	4.93	3.80

C<sub>0</sub> - ControlC<sub>1</sub> - Green ZnO 500 ppmC<sub>2</sub> - Green ZnO 750 ppmC<sub>3</sub> - Green ZnO 1000 ppmC<sub>4</sub> - Green ZnO 1250 ppmC<sub>5</sub> - Green SiO<sub>2</sub> 250 ppmC<sub>6</sub> - Green SiO<sub>2</sub> 500 ppmC<sub>7</sub> - Green SiO<sub>2</sub> 750 ppmC<sub>8</sub> - Green SiO<sub>2</sub> 1000 ppmC<sub>9</sub> - Chemical ZnO 250 ppmC<sub>10</sub> - Chemical ZnO 250 ppmC<sub>11</sub> - Chemical SiO<sub>2</sub> 500 ppmC<sub>12</sub> - Chemical SiO<sub>2</sub> 500 ppmC<sub>13</sub> - Spinosad 4.4 mg/kg seed



Higher precursor activity of nanoscale Zn in production of essential biomolecules that activates various enzymes which are responsible for driving many metabolic reactions in seeds and might mediate the ROS production leads to activation of pre-germinative metabolism in seed, Whereas, silica nanoparticles could increase cell extension by formatting complexes of Si polyphenol or lignin, which facilitate cell wall loosening to increase the seed germination, results are in conformity with Dragisic *et al.* (2007); Shyla & Natarajan (2014) and Korishettar *et al.* (2016).

Speed of germination significantly differed among concentrations, highest speed in germination of seedlings were recorded in green zinc oxide @ 1250 ppm (28.64) and it is on par with green zinc oxide @ 1000 ppm (27.51), which is immediately followed by green silicon dioxide @ 750 ppm (26.17), green silicon dioxide @ 500 ppm (25.98). Among chemical NPs, speed of germination in chemical zinc oxide @ 500 ppm (24.28), chemical silicon dioxide @ 250 ppm (22.61) compared to the spinosad (24.91) and control (21.63) were recorded (Table 2). The reason for rapid germination could be NPs may form new pores on seed coat during penetration facilitating the influx of water inside the

seed thereby enhanced the speed of germination, similar findings were also reported by Sridhar (2012) and Mahakam *et al.* (2016).

Seedling shoot length significantly differed among concentrations used (Fig. 1). Highest shoot length was recorded in seeds treated with green zinc oxide @ 1250 ppm (29.23 cm) and it is on par with green zinc oxide @ 1000ppm (27.03 cm). Among chemical NPs, shoot length recorded in chemical zinc oxide @ 500 ppm (23.48 cm) followed by chemical silicon dioxide @ 250 ppm (23.19 cm) compared to the spinosad (22.80 cm) and control (19.72 cm). Seedling root length significantly differed among green and chemical NPS (Fig. 1). green zinc oxide @ 1250 ppm (24.32 cm) recorded highest root length and it is on par with green zinc oxide @ 1000ppm (22.48 cm). Among chemical NPs, chemical zinc oxide @ 500 ppm (20.24 cm), chemical silicon dioxide @ 250 ppm (19.88 cm) compared to the spinosad (18.18 cm) and control (19.14 cm) was recorded.

Green zinc oxide @ 1250 ppm (53.55 cm) better in showing seedling length and it is on par with green zinc oxide @ 1000 ppm (49.51 cm), which is immediately followed by green silicon dioxide @ 750 ppm (47.77 cm), chemical zinc oxide @ 500 ppm



Fig. 1: Influence of seed treatment with nanoparticles on mean seedling length in pigeonpea cv. BRG-5

C<sub>0</sub> - Control

C<sub>1</sub> - Green ZnO 500 ppm

C<sub>2</sub> -Green ZnO 750 ppm

C<sub>3</sub> -Green ZnO 1000 ppm

C<sub>4</sub> -Green ZnO 1250 ppm

C<sub>13</sub> -Spinosad 4.4 mg/kg seed

C<sub>5</sub> -Green SiO<sub>2</sub> 250 ppm

C<sub>6</sub> -Green SiO<sub>2</sub> 500 ppm

C<sub>7</sub> -Green SiO<sub>2</sub> 750 ppm

C<sub>8</sub> -Green SiO<sub>2</sub> 1000 ppm

C<sub>9</sub> -Chemical ZnO 250 ppm

C<sub>10</sub> -Chemical ZnO 500 ppm

C<sub>11</sub> -Chemical SiO<sub>2</sub> 500 ppm

C<sub>12</sub> -Chemical SiO<sub>2</sub> 250 ppm

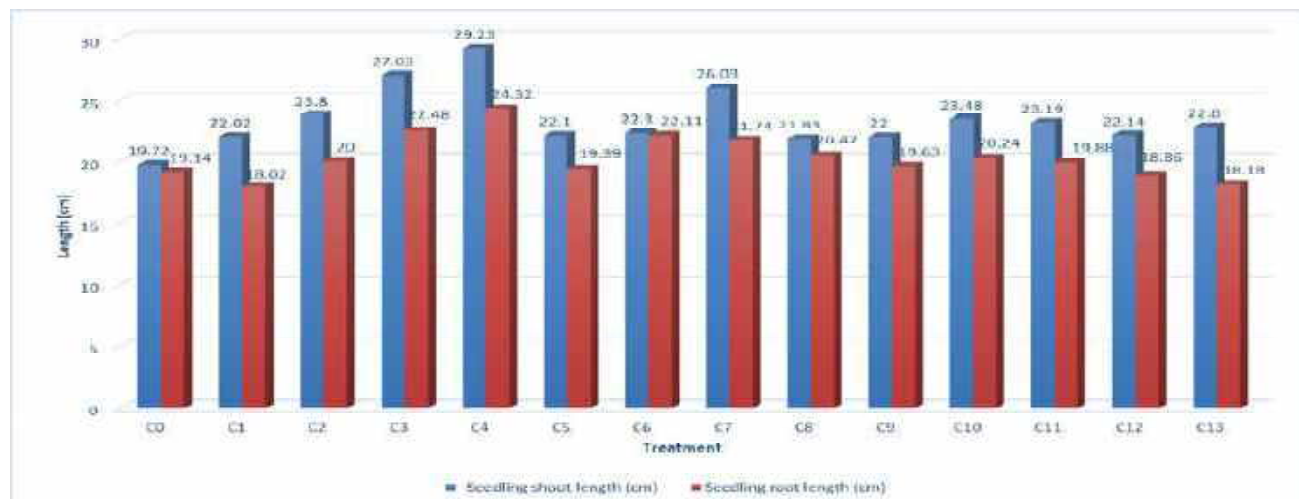


Fig. 2: Influence of seed treatment with nanoparticles on seedling shoot and root length (cm) in pigeonpea cv. BRG-5

C<sub>0</sub>- Control

C<sub>1</sub>- Green ZnO 500 ppm

C<sub>2</sub>-Green ZnO 750 ppm

C<sub>3</sub>-Green ZnO 1000 ppm

C<sub>4</sub>-Green ZnO 1250 ppm

C<sub>5</sub>-Green SiO<sub>2</sub> 250 ppm

C<sub>6</sub>-Green SiO<sub>2</sub> 500 ppm

C<sub>7</sub>-Green SiO<sub>2</sub> 750 ppm

C<sub>8</sub>-Green SiO<sub>2</sub> 1000 ppm

C<sub>9</sub>- Chemical ZnO 250 ppm

C<sub>10</sub>-Chemical ZnO 500 ppm

C<sub>11</sub>- Chemical SiO<sub>2</sub> 500 ppm

C<sub>12</sub>-Chemical SiO<sub>2</sub> 500 ppm

C<sub>13</sub>-Spinosad 4.4 mg/kg seed

(43.71 cm) compared to the spinosad (40.98 cm) and control (38.86 cm) in Fig. 2.

The probable reason could be due to the excess absorption at higher concentration resulted in penetration of NPs in to cell wall and plasma membrane of epidermal layers in shoot, root and accumulation in vascular tissues thereby enhancement in cell division and cell elongation in pigeonpea, similar findings were reported by Korishettar *et al.* (2016) in pigeonpea.

Mean seedling dry weight significantly differed among concentrations of Zn and Si NPs (Table 3). Green zinc oxide @ 1250 ppm (60.5 mg) recorded highest dry weight of seedling and it is on par with green zinc oxide @ 1000 ppm (60.5 mg), which is followed by green silicon dioxide @ 750 ppm (59.0 mg). Among chemical NPs, dry weight of seedlings were recorded in chemical zinc oxide @ 500 ppm (56.3 mg) followed by spinosad (53.0 mg) compared to the control (47.0 mg). During seed germination and early seedling growth, soluble sugars are mobilized from seeds resulted in higher biomass of the plants, similar findings were reported by Mahakam *et al.* (2016).

A significant variation for seedling vigour index-I and II was observed among concentrations used. Highest SVI-I recorded in green zinc oxide @ 1250 ppm (5153) and is on par with green zinc oxide @ 1000 ppm (4692). Among chemical NPs, chemical zinc oxide recorded @ 500 ppm (4174), chemical silicon dioxide @ 500 ppm (4026) compared with spinosad (3821) and control (3575) (Table 3). The highest SVI-II recorded in seeds treated with green zinc oxide @ 1250 ppm (5823) and it on par with green silicon dioxide @ 750 ppm (5635). Among chemical NPs, chemical zinc oxide @ 500 ppm (5371), followed by green silicon dioxide @ 1000 ppm (5071) compared to the spinosad (4942) and control (4324) (Table 3). Vigour enhancement by incorporation of nanoparticles (SNPs) increased cell division within the apical meristem of seedling, similar results were reported by Harish and Rame Gowda (2017).

Electrical conductivity significantly differed among concentrations used. Among NPs, green zinc oxide @ 1250 ppm (26.49  $\mu\text{S}/\text{cm}/\text{g}$ ) recorded lowest electrical conductivity and it is on par with green silicon dioxide @ 500 ppm (28.05  $\mu\text{S}/\text{cm}/\text{g}$ ). Among chemical NPs chemical zinc oxide @ 500 ppm

TABLE 3

Influence of seed treatment with nanoparticles on mean seedling dry weight, seedling vigour index- I, seedling vigour index- II, Electrical conductivity and total dehydrogenase activity in Pigeonpea cv. BRG-5

	Mean Seedling Dry Weight (mg)	Seedling Vigour Index- I	Seedling Vigour Index- II	Electrical Conductivity ( $\mu\text{S}/\text{cm}/\text{g}$ )	Total Dehydrogenase Activity (Absorbance at 480nm)
C <sub>0</sub>	47.0	3575	4324	35.87	1.773
C <sub>1</sub>	52.1	3734	4856	30.93	1.956
C <sub>2</sub>	52.5	4095	4912	29.90	2.037
C <sub>3</sub>	57.8	4692	5472	29.14	2.187
C <sub>4</sub>	60.5	5153	5823	26.49	2.353
C <sub>5</sub>	49.4	3858	4597	30.30	1.885
C <sub>6</sub>	49.8	4262	4776	28.05	1.849
C <sub>7</sub>	59.0	4563	5635	28.35	2.163
C <sub>8</sub>	54.2	3955	5071	29.20	2.188
C <sub>9</sub>	53.4	3872	4963	32.93	2.028
C <sub>10</sub>	56.3	4174	5371	31.88	2.021
C <sub>11</sub>	52.0	4026	4862	34.71	1.915
C <sub>12</sub>	51.9	3853	4875	35.79	1.839
C <sub>13</sub>	53.50	3821	4942	34.22	2.004
Mean	53.53	4116.64	5034.21	31.27	2.014
SEm ( $\pm$ )	8.36	84.21	85.46	0.54	0.05
CD@0.01	3.18	321.30	326.08	2.04	0.19
CV (%)	3.12	4.09	3.40	3.42	4.96

C<sub>0</sub> - Control  
C<sub>1</sub> - Green ZnO 500 ppm  
C<sub>2</sub> - Green ZnO 750 ppm  
C<sub>3</sub> - Green ZnO 1000 ppm

C<sub>4</sub> - Green ZnO 1250 ppm  
C<sub>5</sub> - Green SiO<sub>2</sub> 250 ppm  
C<sub>6</sub> - Green SiO<sub>2</sub> 500 ppm  
C<sub>7</sub> - Green SiO<sub>2</sub> 750 ppm

C<sub>8</sub> - Green SiO<sub>2</sub> 1000 ppm  
C<sub>9</sub> - Chemical ZnO 250 ppm  
C<sub>10</sub> - Chemical ZnO 250 ppm  
C<sub>11</sub> - Chemical SiO<sub>2</sub> 500 ppm

C<sub>12</sub> - Chemical SiO<sub>2</sub> 500 ppm  
C<sub>13</sub> - Spinosad 4.4 mg/kg seed

(31.88  $\mu\text{S}/\text{cm}/\text{g}$ ) followed by chemical silicon dioxide 250 ppm (34.71  $\mu\text{S}/\text{cm}/\text{g}$ ) compared to the spinosad (34.22  $\mu\text{S}/\text{cm}/\text{g}$ ) and control (35.87  $\mu\text{S}/\text{cm}/\text{g}$ ) were recorded (Table 3). Electrical conductivity of seed leachate indicates the seed coat or membrane integrity, nanoparticles at lower concentration had no negative effect on the seed surface thereby reduced leakage of solute or electrolytes from the seeds by maintaining the seed coat integrity, these results are in conformity with study of Surabhi *et al.* (2021).

Total dehydrogenase activity (Table 3) of seeds treated with green zinc oxide @ 1250 ppm was highest (2.353

$A_{480\text{nm}}$ ) and it is on par with green silicon dioxide 1000 ppm (2.188  $A_{480\text{nm}}$ ) followed by chemical zinc oxide 500 ppm (2.021  $A_{480\text{nm}}$ ) compared to the spinosad (2.004  $A_{480\text{nm}}$ ) and control (1.773  $A_{480\text{nm}}$ ). The increased availability of these micronutrients at nanoscale with increased chemical reactivity resulted in the increase in synthesis and activity of the dehydrogenase enzymes, similar results were reported by Vijayalaxmi *et al.* (2013).

Among fourteen concentrations of zinc and silica nanoparticles, green synthesized nanoparticles found better in all seed quality parameters. Among green nanoparticles, zinc oxide @ 1250 ppm found better

in seed germination (96.25%), mean seedling length (53.55cm), seedling vigour index I (5153) and field emergence (94.50%). Among chemical nanoparticles, zinc oxide @ 500 ppm found better in seed germination (95.50%), mean seedling length (43.71), seedling vigour index (4174) and field emergence (92.50%). It was clearly concluded that, seed treatment with nanoparticles is promising technology which improves the seed quality in pigeonpea.

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## Cloning of *White* Gene of the Melon Fly, *Zeugodacus cucurbitae* (Coquillett) (Diptera : Tephritidae) and *in vitro* Restriction Analysis of Different Single Guide RNAs (sgRNAs)

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### ABSTRACT

*Zeugodacus cucurbitae* (Coquillett) is a major pest affecting cucurbit crops. CRISPR/Cas9 mediated mutagenesis of target genes in *Z. cucurbitae* by generating a series of frame-shift mutation will help in changing physiology and behaviour of the insect. *White* gene is an important eye pigmentation gene widely used as marker gene in *Drosophila melanogaster*. PCR and Cloning of *Z. cucurbitae white* gene (2051bp) was performed. Off-target minimized gRNAs were designed by using bioinformatics online software CHOPCHOP by giving *white* coding sequences as input. Efficiency of the designed sgRNA was confirmed by *in vitro* restriction assay.

Keywords : CRISPR/Cas9, Guide RNA, *White* gene, *Zeugodacus cucurbitae*

FRUIT flies (Diptera : Tephritidae) are highly invasive and damaging pest species affecting the international trade of fruits and vegetables. More than 4500 species from Tephritidae family flies were described till date, which represents one of the most diverse group of acalyptrate Dipterans from superfamily Tephritoidea (Freidberg, 2006 and David *et al.*, 2016). Direct losses in a large variety of agricultural produce occur due to feeding and development of the maggots in fruits and vegetables (Virgilio *et al.*, 2015).

The melon fly, *Zeugodacus cucurbitae* (Coquillett) (Diptera: Tephritidae) is presumed to be from India (Bezzi, 1913), but now this pest has worldwide distribution (Virgilio *et al.*, 2010 and Li *et al.*, 2012) infesting more than 125 host plant species, most of which belong to the family Cucurbitaceae like cucumber (*Cucumis sativus*), bitter melon (*Momordica charantia*), pumpkins (*Cucurbita moschata*), watermelon (*Citrullus lanatus*), muskmelon (*Cucumis melo*) etc. (White, 2006; Vayssieres *et al.*, 2008). *Z. cucurbitae* adults respond

positively to preferred cucurbitaceae host fruit volatiles (Subhash *et al.*, 2018).

Existing management practices for the melon fly are destruction of infested fruits and vegetables, raking of soil, biological control and chemical control. Sterile insect technique (SIT) based area wide management practices helped to eradicate the pest from all the islands of the Okinawa archipelago in Japan (Shimizu *et al.*, 2007). Conventional management practices are associated with many disadvantages related to efficiency, can be time-consuming and come with environmental, animal and human hazards as well as side effects. By considering drawbacks, researchers explored new strategies and recent advancements in genetic control

CRISPR-Cas9 system is considered as a revolutionary technology with high efficiency and precision that can be applied to a wide range of species. A single guide RNA (sgRNA) finds target site in a genome, which can be implemented by scanning for protospacer adjacent motif (PAM) sequences (like 5'-NGG-3'

for SpCas9). The guide RNA (gRNA) domain of the sgRNA determines both the efficacy and specificity of the genome editing activities by Cas9 (Jinek *et al.*, 2012).

Researches on the basis of genetic control of the melon fly, *Z. cucurbitae* are lacking. *White* gene regulating pigmentation in *Z. cucurbitae* will modify the visual behaviour of the target insect, which will help in devising suitable genetic control for the area wide pest management of the same. Optimization of sgRNA design is important for the success of gene editing experiments. The aim of this study is to identify and clone the *white* gene in *Z. cucurbitae* and validate sgRNAs by *in vitro* restriction assay to confirm the efficiency of restriction of the target gene, so that we can proceed for further micro injection studies.

## MATERIAL AND METHODS

### Mass Rearing of the Insect

Stock culture of *Zeugodacus cucurbitae* was maintained on cucumber (*Cucumis sativus* L.) at Division of Basic Sciences, ICAR-IIHR, Bengaluru, India at 25±1 °C, 75±1 per cent relative humidity with 14h:10h L:D photoperiod. The infested cucumber was placed in a container containing thin layer of sieved sand at the bottom for pupation. After pupation, the pupae were collected from sand and transferred to acrylic cages for adult emergence. Grinded sugar along with yeast powder and water was provided as a food source for adults. Insects obtained from this starter culture were used in further experiments.

### White Gene Identification and Primer Designing

The pigmentation gene, *white* was selected from the annotated NCBI genome database. Sequence similarity of *white* gene of melon fly with other tephritid species was determined by sequence alignment through ClustalW multiple sequence alignment in BioEdit (version 7.2.6.1) software.

Gene-specific primers were designed manually by using OligoAnalyzer Tool at Integrated DNA Technologies (IDT) site (Table 1). Then the specificity

of the primers was confirmed by using the database of NCBI Primer BLAST (<https://www.ncbi.nlm.nih.gov/tools/primer-blast/>).

TABLE 1

### *Zeugodacus cucurbitae* white gene-specific primers

Primers	Sequences (5'-3')
Forward Primer	AAATGGGTCAGGAGGATCAG
Reverse Primer	CTCCTCATTTTACTCCTTGCG

### RNA Isolation and Complementary DNA (cDNA) Synthesis

Male adult flies (5 nos.) were taken from stock culture and kept in a 10 ml falcon tube. Then the falcon tube was kept in a container containing liquid nitrogen for 2 minutes. After that the tube was shaken vigorously to detach different body parts. Then five heads were collected by using fine camel brush and transferred to one 1.5 ml eppendorf tube. Total RNA was extracted by using Trizol Reagent (Sigma Aldrich, USA) according to the manufacturer's protocol. Integrity of RNA was checked on 1 per cent agarose gel and further quantified with Nanodrop spectrophotometer (Nanodrop Lite, Thermo Scientific, USA).

Complementary DNA (cDNA) was synthesized from the total RNA (2µg) by Revert Aid First Strand cDNA synthesis kit (Thermo Scientific, USA) according to the manufacturer's protocol. cDNA synthesis was confirmed by PCR amplification of *RPL60* internal control gene and confirmed on 2 per cent agarose gel.

### PCR Amplification of *White* Gene and Gel Elution

The cDNA was diluted by using autoclaved milliQ water (1:10::cDNA:Water) and further used as a template to amplify the entire coding region of the *white* gene using gene-specific primers in PCR (Table 1, 2 and 3). Amplicon products were separated to determine size *via* electrophoresis on 1 per cent agarose gel. The desired amplicon band was eluted by using NucleoSpin Extract II kit (Machery Nagel, Germany) by adopting manufacturer's protocol and further used for cloning.

TABLE 2  
PCR amplification of white gene

Chemicals for White Gene Amplification	Working Concentration	Quantities ( $\mu$ l)
Autoclaved milliQ water	-	12.6 $\mu$ l
10X Mg <sup>+2</sup> free buffer	1X	2.5 $\mu$ l
25mM MgCl <sub>2</sub>	2.5mM	2.5 $\mu$ l
2.5mM dNTPs mix	0.4mM	4.0 $\mu$ l
Template (cDNA)	1:10 diluted	1.0 $\mu$ l
Forward Primer (white gene) 5'-AAATGGGTCAGGAGGATCAG-3'	0.2 $\mu$ M	1.0 $\mu$ l
Reverse Primer (white gene) 5'-CTCCTCATTCTTACTCCTTGCG-3'	0.2 $\mu$ M	1.0 $\mu$ l
LA Taq polymerase	1 unit/ $\mu$ l	0.4 $\mu$ l
	Total Volume	25 $\mu$ l

TABLE 3  
PCR conditions

Steps	Temperature	Time	Cycles
Initial denaturation	95°C	1 minutes	1x cycle
Final denaturation	95°C	10 seconds	35x cycles
Annealing	56°C	40 seconds	
Extension	68°C	2 minutes 10 seconds	
Final extension	68°C	10 minutes	1x cycle
Store	4°C	Forever	

### Ligation of eluted *white* gene amplicon to cloning vector and transformation

The eluted *white* gene amplicon was ligated into general purpose cloning vector, pTZ57R/T vector (Thermo Scientific, Lithuania) (Table 4). The main features of the vector are the blue and white colony selection, the presence of ampicillin resistance

marker gene and the integrated sequence of M13 primers for easy sequencing etc. Recombinants were distinguished from non-recombinants by blue-white selection of colony. Blue colony indicates non-recombinant colony, whereas white colony indicates recombinant colony.

The ligated products were used for transforming *Escherichia coli* DH5- $\alpha$  by standard protocols. The transformed cells were spread on LB agar plates containing X-gal (20 mg/ml), IPTG (100 mM) and ampicillin (100  $\mu$ g/ml). The plates were then incubated at 37 °C overnight to screen blue and white colonies and all the white colonies (colonies harboring the insert) were inoculated in LB broth containing ampicillin, incubated at 37 °C overnight and stored at 4 °C until further use.

### Plasmid Isolation and Sequencing

Plasmids were isolated from the overnight culture of the transformed white colonies cultured in LB broth

TABLE 4

#### Ligation of *white* gene into cloning vector

Chemicals Added	Working Concentration	Quantities ( $\mu$ l)
Autoclaved milliQ water	-	8.5 $\mu$ l
5X Ligase buffer	1X	4.0 $\mu$ l
pTZ57R/T vector	25 ng	0.5 $\mu$ l
Template gene ( <i>white</i> )	111.8 ng	6.0 $\mu$ l
T4 DNA ligase	1 unit/ $\mu$ l	1.0 $\mu$ l
	Total Volume	20 $\mu$ l

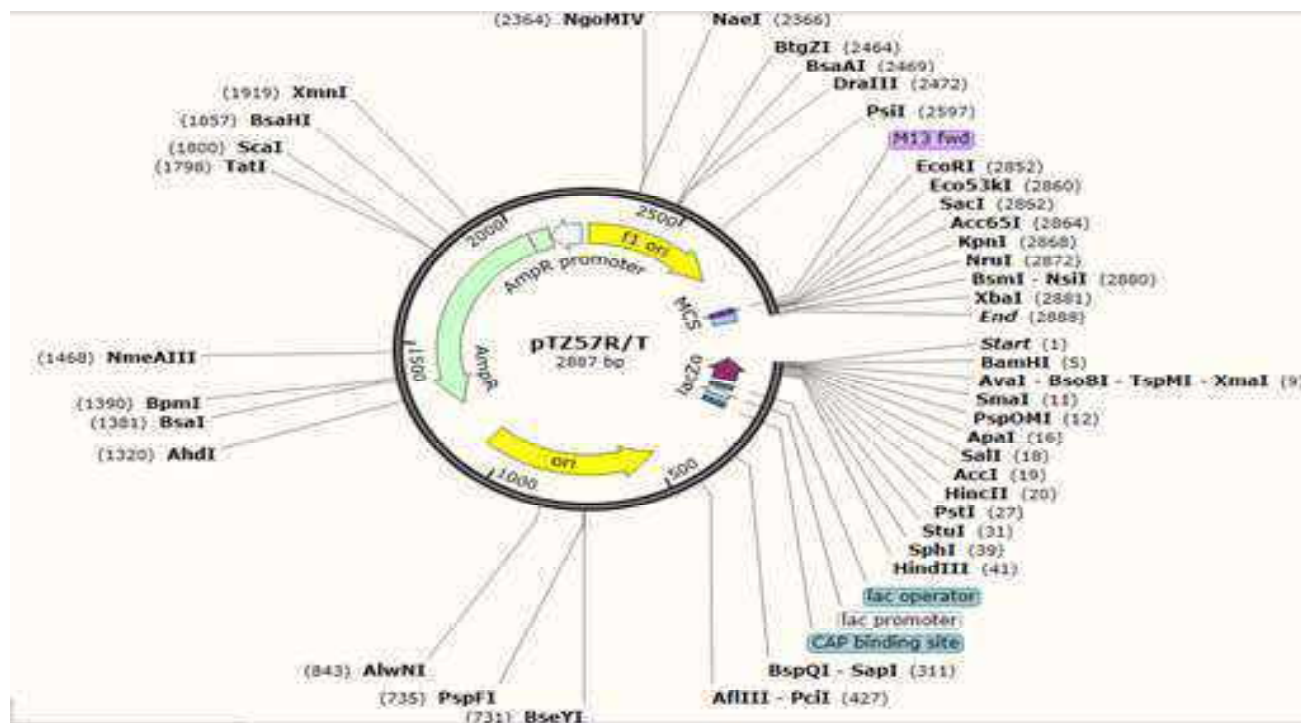


Fig. 1 : pTZ57R/T vector map

using Gene JET™ Plasmid Miniprep Kit (Thermo Scientific, Fermentas, Lithuania) according to manufacturer's protocol and the recombinant plasmid was confirmed in 1 per cent agarose gel electrophoresis with reference plasmid (control DNA1).

Sequencing was carried out in triplicates (three biological replicates and three technical replicates) of the above clones in an automated sequencer (ABI prism ® 3730 XL DNA Analyzer; Medauxin, Bengaluru) using M13 universal primers both in forward and reverse directions.

### Sequence Analysis and Data Interpretation

Multiple sequence alignments of the *white* gene clone sequences and database *white* gene reference sequences were performed with ClustalW multiple alignment tool and the results were displayed using 'BioEdit (version 7.2.6.1). Percentage query similarity between the *white* gene sequenced clones and database *white* gene reference sequences was noted.

### Identification of Off-Target Minimized gRNA

CHOPCHOP (version 3) (<https://chopchop.cbu.uib.no/>) is a web tool for selecting target sites for

CRISPR/Cas9, CRISPR/Cpf1, CRISPR/Cas13 or NICKASE/TALEN-directed mutagenesis. It uses different alignment algorithms to predict off-target binding of sgRNAs and TALENs within short search time (Montague *et al.*, 2014).

The sequenced *Z. cucurbitae* genome set was imported into the public site of the CHOPCHOP (version 3) tools (Labun *et al.*, 2016). The gRNA region was identified from functional domain of the concerned protein by using CHOPCHOP (version 3) tools. Two sgRNA target sites, in exon2 (5'-AGATTATCCGTGGTGAGCGTAGG-3') and exon 7 (5'-ACCGAATGAAGTCGACACATTGG-3') were selected by performing NCBI-BLAST to check off-target effect. Reverse complement of the gRNA was also designed (Table 5).

### Hybridisation of gRNA and Cloning

The designed gRNA and its reverse complement were hybridized by following manufacturer's protocol (Thermo Scientific sgRNA hybridization kit). Then the hybridized gRNA was ligated to a lab modified linearized IVT cloning vector and incubated at 16 °C



TABLE 5  
*Zeugodacus cucurbitae* white gene sgRNA primers and its reverse complement (RC)

Whsg1	Whsg7
>Wh sg1-	>Wh sg7-
AGATTATCCGTGGTGAGCGTAGG	ACCGAATGAAGTCGACACATTGG
>Wh sg1 RC-	>Wh sg7 RC-
CCTACGCTCACCACGGATAATCT	CCAATGTGTCGACTTCATTTCGGT

overnight. Initially, the IVT cloning vector was digested by using endonuclease enzyme (BbsI), the site where sgRNA has to be ligated. Then the ligated product was transformed into *E. coli DH5-α* cell and incubated over night at 37 °C. The recombinant colony was picked for inoculation into fresh LB media and plasmid was isolated from overnight inoculated product. Plasmids were further sequenced in order to identify the insert.

**PCR Amplification of sgRNA Cassette and *in vitro* Transcription**

For *in-vitro* single guide RNA synthesis, sgRNA cassettes (T7 promoter + sgRNA+ scaffold+ terminator) was amplified by using M13 forward and

reverse primers. The amplicon size was determined by gel electrophoresis in 1.5 per cent agarose gel.

TABLE 6  
 Ligation protocol for sgRNA mobilization into IVT vector

Chemicals added	Working Concentration	Volume (μl)
Autoclaved MilliQ water	-	8.5 μl
5X Ligase buffer	1X	4.0 μl
Linearised IVT vector	25 ng	3 μl
ds Oligos (Hybridized gRNA)	2.75 ng	3.5 μl
T4 DNA ligase	1 unit/ μl	1.0 μl
Total Volume		20 μl

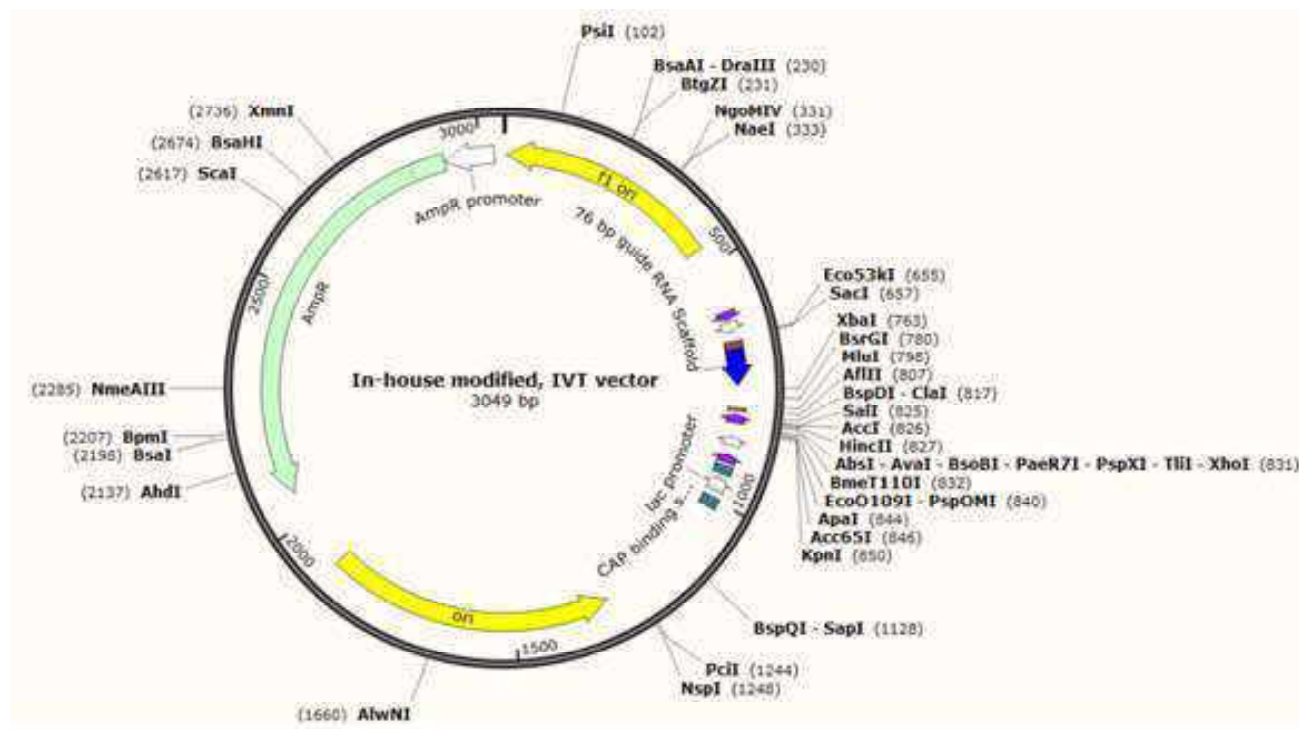


Fig. 2 : IVT vector map

The sgRNAs (24 nucleotides) were synthesized through *in vitro* transcription by using T7 promoter by following the manufactures protocol (NEW ENGLAND Biolab ® Inco) (Table 7). The reaction mixture was incubated 37 °C overnight. Next day the sample was treated with DNase I (4 µl) enzyme and incubated at 37 °C for 1 hour and then the samples were treated with 0.5 M EDTA (5 µl) and incubated at 65 °C for 10 minutes to denature DNase I enzyme. Then the sample was purified by Phenol: Chloroform: Isoamyl alcohol (P:C:I) method to yield good quantity of sgRNAs.

TABLE 7  
*In vitro* transcription reaction mixture

Components	Working Concentration	Volume
Autoclaved MilliQ water	-	12.5 µl
5X Transcription Buffer	1X	20 µl
NTP Mix	10 mM	20 µl
Template	5 µg	40 µl
T7 RNA Polymerase	20 units/ µl	5 µl
Ribolock RNase Inhibitor	40 units/ µl	2.5 µl
Total Volume		100 µl

The concentration of sgRNAs were quantified using Nanodrop spectrophotometer (Nanodrop Lite, Thermo Scientific, USA) and further assayed on 2 per cent agarose gel with RNA ladder that has 100-base band. Concentration of the sgRNAs were estimated by using ImageJ software (v 1.53s) by comparing the intensity of band between ladder and sgRNAs.

### *In vitro* Restriction Assay

*In vitro* restriction of *white* CDS with Cas9 nuclease and synthesized sgRNAs is quintessential to validate the efficiency of sgRNAs. The assay was performed for two sgRNAs (Whsg1, Whsg7) along with a negative control (Table 8). The components of CRISPR/Cas9 system were added in fixed quantities by following manufactures protocol (NEW ENGLAND Biolab ® Inco) with slight modifications (Table 9). Then *in vitro* restriction was confirmed by gel electrophoresis in 2 per cent agarose gel.

TABLE 8  
The CRISPR/Cas9 system of two sgRNAs for *in vitro* restriction assay

Sample No.	Components of CRISPR/Cas9 System
1.	<i>White</i> gene CDS + Cas9 only
2.	<i>White</i> gene CDS + Cas9 + Whsg1
3.	<i>White</i> gene CDS + Cas9 + Whsg7

TABLE 9  
The CRISPR/Cas9 components for *in vitro* restriction assay

Components	Working Concentration	Volume
Autoclaved MilliQ water	-	11.34 µl
NEBuffer r3.1	1X	2 µl
150 mM KCl	5 mM	0.66 µl
sgRNA	150 ng/ µl	0.4 µl
Cas9 diluent (0.25 µl + 4.75 µl NEBuffer r3.1)	300 ng/ µl	0.6 µl
Reaction Volume		15 µl
Pre-incubate for 30 minutes at 25°C		
<i>White</i> gene CDS	150 ng/ µl	5 µl
Total Reaction Volume		20 µl
Incubate for 1 hour at 37°C		

## RESULTS AND DISCUSSION

### RNA Isolation and cDNA Synthesis

Integrity of total RNA isolated from head region was confirmed by gel electrophoresis in 1 per cent agarose gel (Fig. 3). In nanodrop spectrophotometer (Nanodrop Lite, Thermo Scientific, USA), the RNA concentration was 1.3 µg/ µl with A260/280 value of 1.64. cDNA synthesis was confirmed by PCR amplification of *RPL60* internal control gene. The amplicon band size of 119bp was separated by gel electrophoresis on 2 per cent agarose gel (Fig. 4). *RPL60* gene was the most stable reference gene (119bp) found in *Zeugodacus cucurbitae* (Zhang *et al.*, 2018).

### PCR Amplification of *White* Gene and Cloning

PCR amplification of *white* gene CDS with gene-specific primers was confirmed by gel

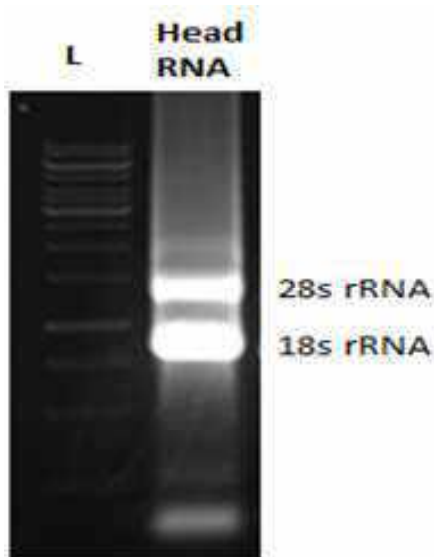


Fig. 3: Total RNA isolated from head region of *Zeugodacus cucurbitae*

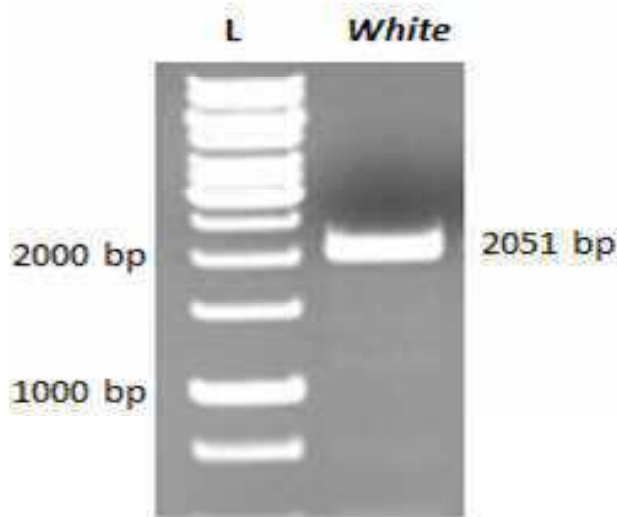


Fig. 5 : PCR amplification of *Zeugodacus cucurbitae* white gene CDS

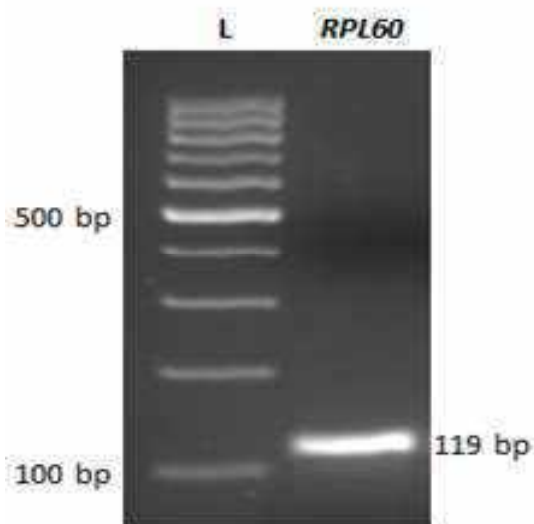


Fig. 4 : PCR amplification of *RPL60* internal control gene

electrophoresis on 1 per cent agarose gel with amplicon band size of 2051bp (Fig. 5). Further the band was eluted from the gel and quantified on Nanodrop spectrophotometer (Nanodrop Lite, Thermo Scientific, USA). The concentration was 34.5 ng/  $\mu$ l. This eluted product was further used for cloning. The isolated plasmids were checked by gel electrophoresis on 1 per cent agarose gel along with reference control DNA1 plasmid to observe the raise in band size. In all the clones, insert was observed (all bands were raised compared to reference plasmid) (Fig. 6).

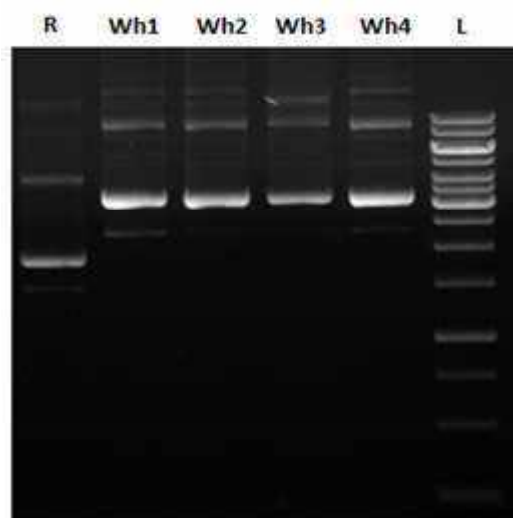


Fig. 6 : *Zeugodacus cucurbitae* white gene clones compared with reference plasmid (R)

NCBI-BLAST of the cloned sequences showed 98.09 per cent sequence similarity with predicted *Z. cucurbitae* protein white (W) (XM\_011189498.2). It showed high sequence similarity with predicted white gene sequences of *Bactrocera dorsalis* (XM\_011202225.3) (90.44%), *B. tryoni* (XM\_040106531.1) (90.20%), *B. latifrons* (XM\_018942980.1) (89.61%) and *B. oleae* (XM\_036371618.1) (89.51%) (Fig.7). This revealed that the predicted domains are conserved in all the related tephritid fruit flies.

Descriptions Graphic Summary Alignments Taxonomy

Sequences producing significant alignments Download Select columns Show 100

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Description	Scientific Name	Max Score	Total Score	Query Cover	E value	Per. Ident	Acc. Len	Accession
<input checked="" type="checkbox"/> <a href="#">Zeugodacus cucurbitae isolate Cucifan white-eyes mRNA, complete cds</a>	<a href="#">Zeugodacus cucurbitae</a>	3768	3768	100%	0.0	100.00%	2040	<a href="#">OQ612786.1</a>
<input checked="" type="checkbox"/> <a href="#">PREDICTED: Zeugodacus cucurbitae protein white (W) mRNA</a>	<a href="#">Zeugodacus cucurbitae</a>	3541	3541	100%	0.0	97.99%	3253	<a href="#">XM_011189498.3</a>
<input checked="" type="checkbox"/> <a href="#">PREDICTED: Bactrocera dorsalis protein white (LOC105224215) mRNA</a>	<a href="#">Bactrocera dorsalis</a>	2710	2710	100%	0.0	90.64%	3392	<a href="#">XM_011202225.4</a>
<input checked="" type="checkbox"/> <a href="#">PREDICTED: Bactrocera neohumeralis protein white (LOC126750950) mRNA</a>	<a href="#">Bactrocera neohumeralis</a>	2599	2599	100%	0.0	90.54%	3326	<a href="#">XM_050473441.1</a>
<input checked="" type="checkbox"/> <a href="#">PREDICTED: Bactrocera tryoni protein white (LOC120776067) mRNA</a>	<a href="#">Bactrocera tryoni</a>	2660	2660	100%	0.0	90.20%	2640	<a href="#">XM_040106531.1</a>
<input checked="" type="checkbox"/> <a href="#">Bactrocera dorsalis clone IHR_BO_WG1 white protein (w) mRNA, complete cds</a>	<a href="#">Bactrocera dorsalis</a>	2595	2595	100%	0.0	88.66%	2037	<a href="#">MT895645.1</a>
<input checked="" type="checkbox"/> <a href="#">PREDICTED: Bactrocera latitrons protein white (LOC18974879) mRNA</a>	<a href="#">Bactrocera latitrons</a>	2593	2593	100%	0.0	89.61%	2142	<a href="#">XM_010942900.1</a>
<input checked="" type="checkbox"/> <a href="#">PREDICTED: Bactrocera oleae protein white (LOC106620505) mRNA</a>	<a href="#">Bactrocera oleae</a>	2582	2582	100%	0.0	89.51%	3349	<a href="#">XM_038371438.1</a>
<input checked="" type="checkbox"/> <a href="#">Ceratitis capitata protein white (W) mRNA</a>	<a href="#">Ceratitis capitata</a>	2043	2043	99%	0.0	84.78%	2250	<a href="#">NM_101273155.1</a>
<input checked="" type="checkbox"/> <a href="#">C. capitata white gene mRNA</a>	<a href="#">Ceratitis capitata</a>	2043	2043	99%	0.0	84.78%	2352	<a href="#">X89533.1</a>
<input checked="" type="checkbox"/> <a href="#">PREDICTED: Anastrepha obliqua protein white (LOC129736220) mRNA</a>	<a href="#">Anastrepha obliqua</a>	1921	1921	98%	0.0	83.94%	2987	<a href="#">XM_054074570.1</a>
<input checked="" type="checkbox"/> <a href="#">PREDICTED: Anastrepha ludens protein white (LOC128558475) transcript variant X4, mRNA</a>	<a href="#">Anastrepha ludens</a>	1905	1905	98%	0.0	83.81%	2590	<a href="#">XM_053084783.1</a>

**gRNAs Cloning, PCR Amplification of gRNAs Cassette and *In vitro* Transcription**

Designed gRNAs had no potential off-target sites, which were confirmed by NCBI-BLAST. The seed region (final 12 nucleotide of the target sequence within the sgRNA) (Wu, 2014) and protospacer adjacent motif (PAM) sequences were matching perfectly to the target sequence. Studies by Cong *et al.* (2013) revealed that the seed region of sgRNA and PAM sequences play vital role in initiating efficient restriction in the target region. Mismatch in these seed region and absence of PAM site effects in non-recognition of target site. gRNA was hybridized and cloned into IVT cloning vector. Plasmids isolated were confirmed by gel electrophoresis in 1 per cent agarose gel against reference IVT vector (Fig. 8). It confirmed insertion of gRNA into the vector in all the gRNAs. Further the clones were confirmed by sequencing.

Guide RNA cassette (T7 promoter + gRNA + scaffold) was PCR amplified with M13 forward and reverse primer. The PCR product was confirmed by gel electrophoresis on 1 per cent agarose gel with amplicon band size of 317bp (Fig. 9). The bands were eluted from the gel and used for *in vitro* transcription. The *in vitro* transcribed sgRNAs were quantified by gel electrophoresis on 2 per cent agarose gel with

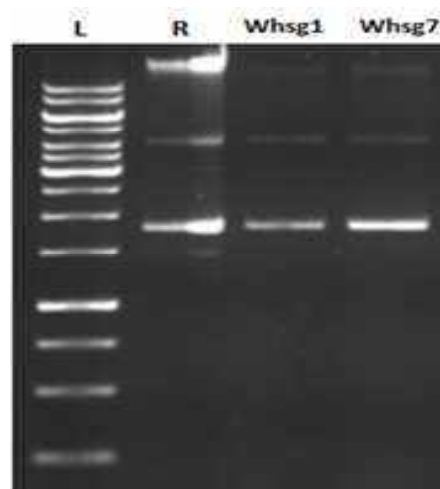


Fig. 8 : White gRNA clones compared with reference IVT vector

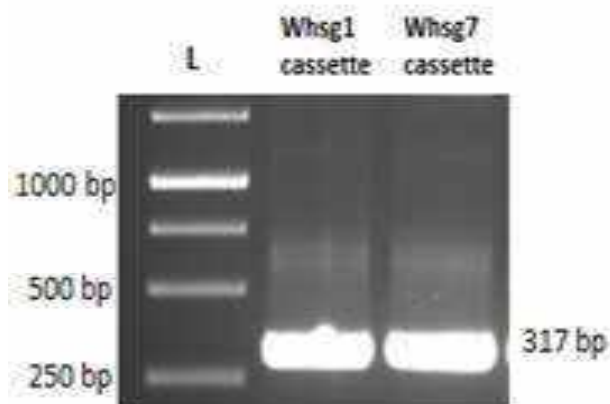


Fig. 9 : PCR amplification of gRNA cassette

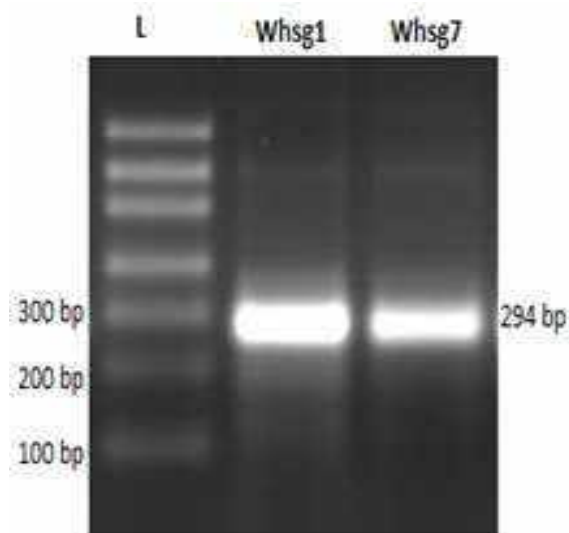


Fig. 10 : Quantification of Wh sgRNA with RNA ladder

RNA ladder (Fig. 10) and processed by using ImageJ software (v1.53s). Concentration of Whsg1 and Whsg7 was 548 ng/  $\mu$ l and 465 ng/  $\mu$ l respectively.

### ***In vitro* Restriction Assay**

*In vitro* restriction assay confirmed the potency of Cas9 protein and sgRNAs to cleave the target site of double stranded DNA. The *in vitro* complex mix was loaded on 1.5 per cent agarose gel. First lane was 1kb ladder, second lane was *white* gene CDS + Cas9, third lane was *white* gene CDS + Cas9 + Whsg1 and fourth lane was *white* gene CDS + Cas9 + Whsg7. Visualisation on agarose gel revealed one solid band of 2051bp in second lane. Third lane showed multiple fragments of bands of size 1890bp and 161bp, which were cut released bands from 2051bp *white* gene CDS band. Fourth lane showed multiple fragments of bands of size 1641bp and 410bp, which were cut released bands from 2051bp *white* gene CDS band (Fig. 11). These sgRNAs were further used for microinjection of ribonucleoprotein (RNP) complex into embryos of *Z. cucurbitae*.

*White* gene was abundantly expressed in the compound eyes of *Bactrocera*, *Anastrepha* and *Ceratitis*. Mutation of this gene leads to loss of eye pigmentation (Bai *et al.*, 2019 and Sim *et al.*, 2019). Complete loss of *white* gene impairs formation of dimers with Brown and Scarlet proteins, which blocks

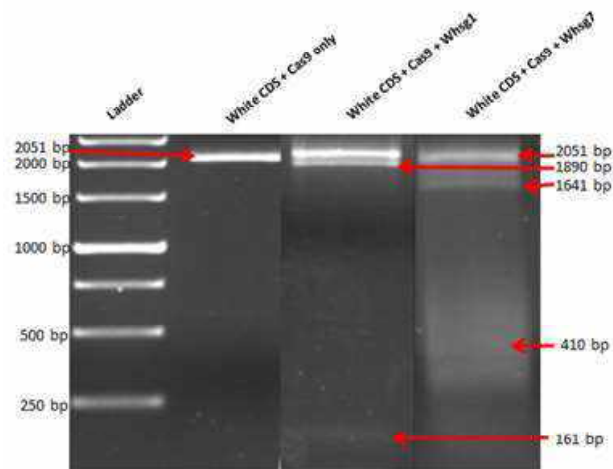


Fig. 11: *In vitro* restriction assay of *white* sgRNAs

the transport of precursor of ommochromes, responsible for eye pigmentations. So, the eyes appear as white as a consequence (Choo *et al.*, 2018).

*White* mutants in *Ceratitis capitata* exhibited unsuccessful mating due to reduced courtship behaviour (Briceno, 2003).

Cloning and sequencing of *white* gene in *Z. cucurbitae* paved way for further characterization and functional analysis. As there is non-availability of published database of *white* gene of *Z. cucurbitae*, functional annotation of this gene following microinjection and phenotypic and molecular validation will enrich the knowledge about its functionality. *In vitro* restriction assay confirmed the restriction efficiency of the designed sgRNAs. Further, it can be proceeded for microinjection of *Z. cucurbitae* embryos. Post mutagenesis studies can be carried out to check physiological and behavioural changes in the flies.

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## Assessment of Soil Fertility Status of Appanahalli Sub Watershed of Gubbi Taluk, Karnataka

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SAYANTIKA BHATTACHARYA :  
Carried out the experiment,  
drafted the manuscript and  
performed the statistical  
analysis;

T. CHIKKARAMAPPA :  
Conceived the study, final  
approval of version to be  
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### ABSTRACT

A study was conducted to assess the fertility status of soils in Appanahalli sub watershed of Gubbi taluk, Karnataka. A total of six micro watersheds under Appanahalli sub watershed with three major landforms viz., undulating upland or Ridge, Midland and Valley under Ragi cropping system were selected. At 300 m grid interval, 180 representative soil samples were collected at 0-20 cm and 20-40 cm depth during the year 2020. Results of study indicated that soil reaction of Appanahalli sub watershed was very acidic to moderately alkaline in nature (4.99 to 8.13) with the mean value of 6.50. The highest mean pH value was observed in Singadahalli micro watershed (6.86). Among the three major landforms studied, pH values in lowland were better than midland and upland physiography. The soils of sub-watershed were non saline and ranged from 0.02 dSm<sup>-1</sup> to 0.48 dSm<sup>-1</sup> in all the micro watersheds. Soil pH and EC were increased with depth. Soil Organic carbon (SOC) content was ranged from 0.13 to 0.78 per cent with a mean value of 0.50 per cent. Haradagere micro watershed recorded highest organic carbon with a mean value 0.58 per cent. However, the values of available N ranged from 123.15 kg ha<sup>-1</sup> to 345.61 kg ha<sup>-1</sup>. The highest average (46.04 and 279.44 kg ha<sup>-1</sup>) available P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O was recorded in Singadahalli and Galigkere-3 micro watershed, respectively. Available Fe, Mn, Cu, Zn and microbial biomass carbon and nitrogen contents were found in sufficiency range in surface soils of all micro watersheds. Soil organic carbon, available N, P, K, Fe, Mn, Cu and Zn contents were higher in surface soils and valley portion of watershed. Nutrient availability in a soil is governed by its pH and better nutrient availability can be expected only at neutral soil reaction. Hence, proper and adequate measures like amelioration of soil acidity/alkalinity need to be adopted for bringing the soils to favourable soil reactions to a better fertility and productivity conditions.

**Keywords :** Watersheds, Soil fertility status, Available nutrients

LAND resources of the country are its most precious and sacred endowment. Land and water are the most important natural resources which play a vital role in agricultural development. But the land is continuously under threat of degradation through various erosional activities. As per the desertification and land degradation atlas of India (2015-2018), 96.4 million hectares *i.e.*, 29.32 per cent of the total geographical area of the country and approximately

6.35 per cent of land in Karnataka is undergoing the process of desertification / land degradation. As per the special report on climate change and land of Intergovernmental panel for climate change released during August, 2019, land use change, land-use intensification and climate change have contributed to desertification and land degradation. The soil productivity and sustainability of soil depends on dynamic equilibrium among its physical, chemical and

biological properties (Ahmed *et al.*, 2012).

Therefore, an imperative stage has come where suitable soil and water conservation measures on watershed basis are immediately warranted to reduce soil erosion, restore land productivity and improve the socio-economic status of the area. With the general acceptance of watershed as the principal unit of planning, many developmental activities based on suitable utilization of locally available natural resources need to be taken which requires detailed characterization of natural resources (Manchanda *et al.*, 2002). Soil resource mapping by using geospatial techniques, identification of constraints/potentials, delineation of erosion-prone areas is pre-requisite for suggesting conservation measures (Surya *et al.*, 2008) and several studies reported potential use of remote sensing for characterization and management of land resources at watershed level (Srinivasa *et al.*, 2008).

Intensive cultivation practices without giving adequate consideration to quality of land resources are noted with ending up in numerous problems like yield stagnation, nutrient mining/depletion and soil degradation. Majumdar *et al.* (2016) quoting evidences from experiments opined that it is a much costlier affair to restore fertility status of a soil denuded of its native fertility through external fertilizer application. Maintenance of food security in a sustainable manner strongly demands management and conservation of land / soil resources by up keeping its quality in a fertile state. In this context, a study namely 'Assessment of soil fertility status of Appanahalli sub watershed of Gubbi taluk, Karnataka' was conducted to know the fertility status of soils of Appanahalli sub watershed.

## MATERIAL AND METHODS

### Study Area

Appanahalli sub-watershed is located in central Karnataka plateau with hot, moist, semi-arid eco sub region, Southern plateau and hill region which belongs to the sub region 8.2 of Karnataka. The sub-watershed (Gubbi taluk, Tumkuru district) is located in between

13°29' 36.94" and 13°25'48.015" North latitude and 76°43'27.767" and 76°48'58.762" East longitudes covering an area of 3484 ha bounded by Anantapur on the north, Kolar and Bangalore on the east, Mandya on south, Hassan and Chitradurga on west. This sub watershed consists of 6 micro watersheds- a) Singadahalli, b) Galigerkere-1, c) Galigerkere-2, d) Galigerkere-3, e) Haradagere-2 and f) Appanahalli-1. The area receives an average annual rainfall of 679.1-888.9 mm, 50 per cent of which is received mainly during kharif season. The elevation of the sub-watershed is 800-900 m above mean sea level. The relief of the study area is very gently sloping to gently sloping, where very gently sloping land covers an area of 1956 ha (56.2%) and gently sloping land occupy 820 ha (23.5%) area. The major crop cultivated in the watershed is Ragi.

### Soil Sampling and Analysis

Considering the uniformity of soil sample distribution in the study area, soil samples were collected from 6 micro watersheds under 3 major landforms (ridge, midland and valley) having ragi based cropping system from 5 farmers plot after harvest. The soil samples were collected at a depth of 0-20 cm and 20-40 cm having approximate grid interval of 300 meters. A total of 180 samples (90 surface and 90 subsurface) were collected from all three major land forms. In the laboratory, the soil samples were air dried under shade and were grounded with a wooden pestle and mortar and passed through 2 mm sieve to separate coarse fragments (> 2 mm). For estimation of chemical properties, a small quantity of 2 mm sieved soil sample was passed through 80 mesh sieves after fine grinding the sample in agate pestle and mortar. The processed soil samples were stored in plastic bags and used for various analysis.

### Chemical Analysis of the Soils

Soil samples collected were analysed for its chemical properties using standard procedures. Soil pH was measured in water at 1:2.5 soil : water ratio as per the method outlined by Jackson (1973). Electrical conductivity (EC) was measured in 1:2.5 soil : water ratio. Soil organic carbon (OC) was determined by



wet digestion method as described by Walkley and Black (1934). Available N, P, K were determined by alkaline potassium permanganate (Subbiah and Asija, 1956), Bray's-1 extraction (Bray and Kurtz, 1945) and Olsen method, neutral ammonium acetate (Jackson, 1973) method respectively. Soil micronutrients were extracted by DTPA at pH 7.3 using 1 : 2 soil : solution ratio as outlined by Lindsay and Norvell (1978). The extractable Fe, Mn, Cu and Zn were estimated by atomic absorption spectrometer.

## RESULTS AND DISCUSSION

### Chemical Properties of Soils under different Landforms

#### Soil Reaction and Electrical Conductivity

Analytical results revealed that pH of Appanahalli sub watershed is very acidic to moderately alkaline in nature (4.99 to 8.13) with a mean value of 6.50. The highest mean value of pH was observed in Singadahalli micro watershed (6.86) followed by Galigkere-3 (6.47) and the lowest mean value was observed in Haradagere micro watershed (6.21) (Table 1). Among the three major landforms studied, pH values in lowland were better than midland and upland physiography. The soils of sub-watershed were non saline and ranged from 0.02 dSm<sup>-1</sup> to 0.48 dSm<sup>-1</sup> with a mean value of 0.16 dSm<sup>-1</sup> (Table 1). In all the micro watersheds, Soil pH and EC of soil increased with depth. This might be due to accumulation of leached bases in the lower horizons. The surface soil accumulated more bases and salts due to their fineness and associated poor drainage conditions owing to low rainfall received in these areas. Thangasamy *et al.* (2005) reported that the variation in soil pH is associated with parent material, rainfall and topography. Pillai and Natarajan (2004) also reported similar low EC values indicating the non-saline nature of soils of Garakahalli watershed.

#### Organic Carbon

Organic carbon (OC) content of Appanahalli sub watershed ranged from 0.13 to 0.78 per cent with a mean value of 0.50 per cent. In Haradagere micro watershed highest OC was recorded with a mean value 0.58 per cent (Table 1). Organic carbon content was

noticed higher in the surface soils compared to that of sub-surface soils for all the studied watersheds. Rajeshwar *et al.* (2009) in a similar study reported that higher values of organic carbon on the surface can be attributed to the addition of farmyard manure and plant residues to surface horizons. This finding is in agreement with findings of Chibsa and Taa (2009), in which they reported that the SOC decrease with increasing soil depth, with more accumulation on the upper surface soil layer. The surface samples of the study area ranged from low to high but most of the samples recorded medium organic carbon content and this may be due to the favourable arid / semi-arid climatic conditions prevailing in these sites for a higher decomposition of the organic matter.

#### Available Nitrogen

The available nitrogen (N) content of soils of Appanahalli sub watershed varied from low to medium (123.15 to 345.61 kg ha<sup>-1</sup>) with a mean value of 228.00 kg ha<sup>-1</sup> (Table 2). In accordance with studies conducted by Chikkaramappa *et al.* (2021) and Sathish *et al.* (2018), low organic matter content in these areas could be due to low rainfall and high temperature which facilitate faster degradation and removal of organic matter led to nitrogen deficiency. Similar nitrogen status was observed by Krishna *et al.* (2017) in Arjunagi sub-watershed under northern dry zone of Karnataka. Their concentration was high on the surface layers compared to sub-surface for all the micro watersheds. Among the three different landforms, available nitrogen content was highest in lowland or valley and least amount was recorded in ridges. This is related to loss of nutrients due to erosion and runoff from upper land of watershed and their deposition in the lowland of watershed. Higher nitrogen value on the surface soils might be due to the high organic carbon content and also external application of fertilizers (Satish Kumar and Naidu, 2012).

#### Available Phosphorous

The available phosphorous content in the soils of sub watershed ranged from 10.12 to 78.34 kg ha<sup>-1</sup> with a mean value of 33.81 kg ha<sup>-1</sup> (Table 2). Available

TABLE I  
Chemical properties of soils under different landforms of Appanahalli sub watershed of Tumkur district

Watershed	Depth (Cm)	Sample no	pH			EC (dS m <sup>-1</sup> )			OC (g/kg)			
			V	M	R	V	M	R	V	M	R	
Appanahalli	0-20	1	6.59	6.63	5.87	0.15	0.16	0.17	3.00	4.20	3.50	
		2	7.65	6.29	7.97	0.18	0.10	0.10	6.60	7.70	3.70	
		3	7.79	6.33	6.44	0.16	0.13	0.17	2.70	6.60	6.70	
		4	7.22	6.14	6.31	0.15	0.09	0.16	3.30	5.10	2.90	
		5	6.49	6.38	6.26	0.15	0.14	0.09	6.40	2.30	7.00	
	20-40	1	6.73	6.65	5.93	0.17	0.18	0.19	2.70	4.00	3.10	
		2	7.80	6.33	7.41	0.23	0.12	0.11	6.10	7.50	3.60	
		3	7.82	6.37	6.50	0.18	0.15	0.19	2.50	6.40	6.10	
		4	7.23	6.30	6.36	0.20	0.10	0.20	3.10	4.40	2.40	
		5	6.53	6.42	6.30	0.17	0.16	0.11	5.90	2.10	6.50	
		Range		5.87-7.97			0.09-0.23			2.10-7.70		
		Mean		6.70			0.15			4.60		
	Galigkere-1	0-20	1	7.08	5.34	6.04	0.12	0.07	0.07	6.00	7.10	2.80
			2	7.13	6.58	6.11	0.04	0.17	0.05	6.40	3.70	6.20
			3	6.85	6.03	5.52	0.09	0.14	0.11	7.80	6.30	4.90
4			6.72	6.22	6.16	0.12	0.12	0.04	6.80	5.60	2.40	
5			7.23	6.35	5.77	0.11	0.05	0.09	6.70	4.10	3.30	
20-40		1	7.13	5.42	6.11	0.15	0.11	0.11	4.60	6.80	2.10	
		2	7.20	6.65	6.14	0.07	0.19	0.08	5.90	3.30	5.60	
		3	7.02	6.16	5.71	0.12	0.16	0.13	7.50	5.80	4.40	
		4	6.84	6.29	6.19	0.14	0.15	0.06	6.30	5.20	2.10	
		5	7.50	6.42	5.85	0.15	0.09	0.12	6.20	3.30	2.60	
		Range		5.34-7.50			0.04-0.19			2.10-7.80		
		Mean		6.39			0.11			5.10		
Galigkere-2		0-20	1	7.18	5.55	6.12	0.15	0.07	0.10	3.90	6.90	2.70
			2	7.37	6.33	6.33	0.10	0.15	0.09	7.20	3.80	4.90
			3	5.42	5.87	6.18	0.11	0.17	0.13	7.10	5.70	5.00
	4		6.39	6.18	6.06	0.14	0.11	0.15	6.30	5.10	4.60	
	5		7.41	5.90	6.98	0.13	0.05	0.03	7.00	3.30	3.10	
	20-40	1	7.18	5.55	6.12	0.15	0.07	0.10	3.30	6.10	1.90	
		2	7.37	6.33	6.33	0.10	0.15	0.09	6.50	3.40	4.40	
		3	5.42	5.87	6.18	0.11	0.17	0.13	5.80	4.90	4.30	
		4	6.39	6.18	6.06	0.14	0.11	0.15	5.70	4.30	4.10	
		5	7.41	5.90	6.98	0.13	0.05	0.03	6.40	2.60	1.90	
		Range		5.42-7.41			0.03-0.17			1.90-7.20		
		Mean		6.35			0.11			4.70		
	Galigkere-3	0-20	1	6.72	7.43	5.59	0.19	0.31	0.07	5.50	4.20	2.80
			2	7.33	6.41	6.11	0.18	0.26	0.10	6.10	3.80	3.30
			3	7.13	6.02	5.18	0.28	0.08	0.038	4.90	3.10	1.90
4			8.02	6.19	5.83	0.43	0.17	0.12	7.60	4.40	2.40	
5			7.16	6.31	4.99	0.26	0.12	0.07	6.80	4.00	1.60	

Watershed	Depth (Cm)	Sample no	pH			EC (dS m <sup>-1</sup> )			OC (g/kg)		
			V	M	R	V	M	R	V	M	R
	20-40	1	6.90	7.68	5.63	0.21	0.32	0.09	5.20	4.10	2.60
		2	7.41	6.45	6.14	0.19	0.28	0.12	5.70	3.60	3.10
		3	7.19	6.19	5.21	0.31	0.11	0.05	4.80	2.70	1.80
		4	8.13	6.27	5.87	0.45	0.21	0.16	7.10	4.10	2.20
		5	7.21	6.35	5.04	0.28	0.14	0.11	6.60	3.90	1.30
	Range			4.99-8.13			0.038-0.45			1.30-7.60	
	Mean			6.47			0.19			4.00	
Haradagere	0-20	1	6.87	6.06	5.31	0.09	0.09	0.08	4.60	6.80	7.10
		2	7.13	6.08	5.23	0.21	0.02	0.04	7.00	6.70	1.80
		3	7.29	6.15	5.14	0.36	0.13	0.10	6.90	7.00	6.60
		4	7.13	6.22	5.05	0.11	0.04	0.07	4.05	7.40	5.80
		5	7.10	5.81	5.11	0.22	0.05	0.12	6.60	6.60	6.60
	20-40	1	7.02	6.29	5.44	0.11	0.17	0.11	4.30	6.50	6.80
		2	7.39	6.30	5.32	0.27	0.09	0.07	4.000	6.60	1.60
		3	7.45	6.39	5.29	0.48	0.16	0.10	6.20	5.00	6.50
		4	7.26	6.35	5.26	0.17	0.07	0.11	3.80	7.10	5.50
		5	7.70	5.98	5.17	0.27	0.09	0.16	6.40	6.40	6.30
Range			5.05-7.7			0.02-0.48			1.60-7.40		
Mean			6.21			0.14			5.80		
Singadahalli	0-20	1	7.83	6.21	5.53	0.34	0.33	0.13	3.40	7.00	5.70
		2	7.89	7.39	7.57	0.27	0.28	0.25	7.00	5.10	3.10
		3	6.78	6.33	5.87	0.36	0.14	0.17	7.70	6.50	6.60
		4	6.88	6.41	5.91	0.31	0.19	0.07	6.20	5.70	6.90
		5	7.54	7.79	6.01	0.26	0.26	0.09	5.40	6.60	4.00
	20-40	1	7.91	6.26	5.80	0.37	0.37	0.15	2.80	4.00	5.20
		2	7.95	7.65	7.65	0.30	0.32	0.28	6.50	4.50	2.60
		3	7.01	6.51	5.97	0.37	0.16	0.19	7.20	6.30	6.20
		4	6.98	6.45	6.01	0.35	0.21	0.10	5.80	5.50	6.30
		5	7.65	7.95	6.08	0.30	0.30	0.11	5.10	5.90	3.20
Range			5.53-7.95			0.07-0.37			2.60-7.70		
Mean			6.86			0.24			5.50		
APPANAHALLI	Range			4.99-8.13			0.02-0.48			1.30-7.80	
	Mean			6.50			0.16			5.00	

V=Valley, M=Midland, R=Ridge

phosphorus status of Appanahalli, Galigkere-1, Galigkere-2 and Galigkere-3 micro watershed areas was low to medium. This might be due to variation in soil properties like clay content, CEC and P fixation capacity. In addition to this, it was observed that the farmers were using only DAP as the source of nutrients in adequate quantity. Haradagere and Singadahalli watershed recorded exceptionally high amount of available phosphorus due to heavy use of complex fertilizers. Application of excess dosage without

having knowledge of the crop requirement and soil fertility status might have led to slight increase in the availability of phosphorous and also variations in available P<sub>2</sub>O<sub>5</sub> content in soils were related with the intensity of soil weathering or soil disturbance, the degree of P-fixation and continuous application of mineral Phosphorous fertilizer sources as indicated by Satish *et al.* (2018). Available phosphorus content was higher in surface soil than subsurface soils and their content were more in valley of watershed when

TABLE 2  
Available macro nutrient status of soils under different landforms of Appanhalli sub watershed of Tumkur district

Watershed	Depth (Cm)	Sample no	Available N (kg ha <sup>-1</sup> )			Available P <sub>2</sub> O <sub>5</sub> (kg ha <sup>-1</sup> )			Available K <sub>2</sub> O (kg ha <sup>-1</sup> )			
			V	M	R	V	M	R	V	M	R	
Appanhalli	0-20	1	282	205.23	249	19.12	15.89	14.24	153.7	148.5	183.43	
		2	193.23	260.1	161.17	11.78	19.12	13.77	330.19	234.1	143.12	
		3	260.7	230.87	196.1	11.66	17.1	14.26	179.98	155.64	197.23	
		4	171.09	220.19	134.1	18.02	14.98	27.12	160.02	140.12	177.16	
		5	206.76	247.17	271.66	16.09	13	55.98	145.76	171.98	168.54	
	20-40	1	280.09	201.12	243.5	17.86	13.45	13.65	151.76	141.76	181.13	
		2	185.77	249.87	150.19	10.12	18.87	11.34	325.67	230.12	140.65	
		3	245.12	222.98	192.45	11.43	15.65	12.76	177.12	148.98	195.78	
		4	166.23	213.56	127.56	17.79	13.29	26.98	154.52	136.15	170.12	
		5	200.78	241.54	255.19	14.99	12.13	50.16	140.98	166.18	166.23	
		Range		127.56-282.12			10.12-55.98			136.15-330.19		
		Mean		215.51			18.09			177.22		
	Galigkere-1	0-20	1	213.43	231.12	185.67	12.34	51.67	49.18	185.12	202.43	140.09
			2	132.87	265.76	323.97	43.97	12.65	19.77	201.32	185.23	142.33
			3	150.32	176.34	224.5	32.88	20.77	29.02	110.86	189.12	127.88
4			156.13	185.76	178.96	47.01	18.02	35.91	281.12	151.12	133.45	
5			156.78	151.43	145.67	15.98	20.88	43.12	156.98	143.87	157.81	
20-40		1	209.87	229.98	179.86	12.19	51.19	49.07	176.65	193.4	131.6	
		2	125.76	261.99	318.23	42.19	12.34	19.68	191.23	168.9	135.2	
		3	141.23	169.87	221.34	32.65	20.68	28.83	100.04	174.65	116.8	
		4	153.42	175.99	172.65	46.92	18.01	35.67	272.43	145.9	128.41	
		5	151.88	143.21	138.23	15.53	20.76	43.02	151.6	133.5	154.21	
		Range		125.76-323.97			12.19-51.67			100.04-281.12		
		Mean		189.07			30.06			162.77		
Galigkere-2		0-20	1	205.76	250.2	181.9	11.89	55.87	51.12	189.5	211.3	150.12
			2	131.6	267.8	323.4	45.71	12.08	19.89	212.43	182.3	142.32
			3	155.12	171.56	212.16	34.67	22.09	30.12	119.12	193.32	129.65
	4		155.87	181.23	220.52	48.02	19.12	28.92	285.65	155.76	147.46	
	5		150.88	156.12	145.6	17.54	20.98	45.65	167.2	145.6	361.43	
	20-40	1	198.16	246.87	176.98	11.76	55.14	50.02	177.98	201.32	145.98	
		2	123.43	260.09	318.98	44.65	11.78	19.21	210.12	176.98	139.12	
		3	149.32	165.43	217.98	33.98	21.9	30.09	111.34	186.78	120.09	
		4	151.77	178.65	213.94	47.09	18.85	28.44	282.73	151.43	139.92	
		5	145.67	151.18	141.54	17.21	20.33	45.12	164.14	139.23	356.99	
		Range		123.43-323.40			11.76-55.87			111.34-361.43		
		Mean		191.65			30.64			183.24		
	Galigkere-3	0-20	1	345.6	231.19	189.5	45.57	44.93	25.53	367.2	350	195.6
			2	342.15	241.87	231	57.19	50.04	24.65	394.8	320.4	169.6
			3	265.3	234.66	212.98	45.95	42.89	17.87	347.6	357.6	153.36
4			285.6	271.14	224.17	58.21	40.12	18.38	386.4	291.6	100.8	
5			278.5	285.65	204.65	41.39	43.29	14.3	357.6	278.4	163.41	

Watershed	Depth (Cm)	Sample no	pH			EC (dS m <sup>-1</sup> )			OC (g/kg)		
			V	M	R	V	M	R	V	M	R
	20-40	1	341.65	224.98	182.43	45.21	44.78	25.31	361.23	344.23	191.32
		2	335.78	235.15	224.13	57.02	49.97	24.19	390.18	313.98	164.89
		3	260.23	227.32	216.24	45.48	42.75	17.78	342.78	352.97	147.12
		4	279.87	264.18	220.13	58.12	40.03	18.32	381.12	276.96	97.65
		5	272.31	280.19	193.24	41.23	43.16	14.12	352.64	272.9	158.74
	Range			182.43-345.60			14.12-58.21			97.65-394.8	
	Mean		253.39			37.93			279.44		
Haradagere	0-20	1	282.34	161.78	345.61	65.01	26.89	31.14	265.78	211.14	230.9
		2	290.12	314.65	320.98	73.99	29.15	29.01	191.17	229.87	213.4
		3	256.17	245.65	275.67	64.86	38.41	20.43	350.98	145.6	119.87
		4	265.78	224.32	140.12	69.12	40.02	26.18	275.68	167.8	295.7
		5	284.6	193.6	211.34	27.72	29.12	33.41	367.87	312.43	181.3
	20-40	1	275.43	156.98	340.04	64.87	26.16	30.98	262.43	201.65	222.65
		2	284.5	208.87	325.76	73.32	28.83	28.65	187.65	224.14	211.2
		3	253.87	241.7	271.8	64.16	38.04	20.14	343.7	141.98	109.8
		4	261.43	219.67	123.15	69.03	39.93	25.77	270.13	160.5	291.13
		5	280.09	187.56	201.87	27.22	27.86	33.05	263.8	301.07	176.87
Range			123.15-345.61			20.14-73.99			109.8-367.87		
	Mean		248.18			40.08			230.94		
Singadahalli	0-20	1	225.12	280.01	220.09	36.65	17.36	38.21	237.12	330.4	161.87
		2	340.09	221.32	225.43	51.98	39.04	38.78	290.02	241.56	177.87
		3	319.12	281.76	281.3	42.65	78.34	33.12	275.43	231.43	181.98
		4	298.87	272.54	265.76	51.87	74.23	20.01	370.4	225.77	185.01
		5	321.77	323.14	215.98	59.02	77.06	39.43	321.5	240.98	121.32
	20-40	1	218.9	276.18	215.43	36.14	16.88	37.97	265.42	323.5	156.78
		2	331.23	215.98	220.12	49.83	38.54	38.18	287.13	235.17	170.16
		3	312.98	275.67	275.54	40.12	78.01	32.76	270.09	227.98	176.98
		4	294.56	268.43	260.13	48.98	74.02	19.12	265.43	221.32	181.13
		5	317.34	319.23	211.76	58.24	76.66	38.01	319.23	232.19	115.98
Range			211.76-340.09			16.88-78.34			115.98-370.4		
	Mean		270.19			46.04			234.71		
APPANAHALLI	Range		123.15-345.61			10.12-78.34			97.65-394.8		
	Mean		228.00			33.81			211.39		

V = Valley M = Midland R = Ridge

compared to upland.

### Available Potassium

Available potassium content of Appanahalli sub-watershed varied from 97.65 to 394.8 kg ha<sup>-1</sup> with a mean value of 211.39 kg ha<sup>-1</sup> (Table 2). In all the six micro watersheds low to medium status of available potassium was recorded with the highest average value of 279.43 kg ha<sup>-1</sup> in Galigkere-3 micro-watershed.

Red soils have lesser fine fractions, in addition, kaolinite types of clay minerals are the causes for medium and low rating of available K<sub>2</sub>O. Application of organic manures to soil which contains various organic acids might have aided in release of non-exchangeable K to water soluble forms and the results were in accordance with Chitra and Janaki (1999). available potassium content was higher in surface soil than subsurface soils and their content were more in

TABLE 3  
Available micro nutrient status in soils of Appanahalli sub watershed of Tumkur district

Watershed	Depth (Cm)	Sample no	Fe (mg kg <sup>-1</sup> )			Mn (mg kg <sup>-1</sup> )			Cu (mg kg <sup>-1</sup> )			Zn (mg kg <sup>-1</sup> )			
			V	M	R	V	M	R	V	M	R	V	M	R	
Appanahalli	0-20	1	17.54	7.65	8.98	9.37	10.6	12.33	1.1	1.6	1.18	3.06	1.6	0.66	
		2	20.76	10.43	5.22	10.67	20.18	9.5	1.22	1.9	1.38	0.5	1.65	0.85	
		3	21.33	17.23	10.75	11.64	12.33	14.5	1.15	0.78	0.61	0.63	0.48	0.76	
		4	7	14.45	14.79	11.3	11.3	6.5	12.76	1.1	0.7	1.03	1.1	0.5	0.8
		5	5.72	11.32	6.93	9.66	11.49	6.99	12.76	1.29	1.32	2.56	0.82	0.56	1.12
20-40		1	16.89	7.45	8.77	9.35	10.4	12.31	0.98	1.41	1.15	2.96	1.34	0.63	
		2	20.73	10.38	5.12	10.54	20.14	9.2	1.16	1.65	1.31	0.32	1.59	0.81	
		3	21.13	17.14	10.56	11.56	12.28	13.98	1.1	0.71	0.57	0.49	0.44	0.72	
		4	5.98	14.38	14.67	11.19	6.21	11.78	0.77	0.67	1.01	1.05	0.41	0.77	
		5	5.66	10.79	6.85	9.63	11.42	6.85	1.15	1.27	2.48	0.76	0.52	1.11	
Range			5.12-21.33			6.21-20.18			0.57-2.56			0.32-3.06			
Mean			11.89			11.22			1.21			0.97			
Galigkere-1	0-20	1	1.78	10.03	12.21	3.41	3.04	5.21	0.31	0.14	0.28	0.15	0.21	0.28	
		2	11.72	2.12	11.84	11.82	1.95	6.93	0.55	0.54	0.51	1.47	0.19	0.61	
		3	13.52	4.17	2.55	11.85	3.49	2.11	0.38	0.11	0.39	0.41	0.16	0.72	
		4	6.18	13.12	2.98	15.39	2.63	4.85	0.26	0.18	0.43	0.23	0.24	0.56	
		5	12.86	4.55	4.74	15.89	9.36	5.26	0.55	0.44	0.15	0.36	0.14	0.22	
20-40		1	1.76	10.01	12.18	3.39	3.01	5.19	0.29	0.11	0.26	0.12	0.18	0.25	
		2	11.69	2.02	11.81	11.81	1.92	6.91	0.51	0.52	0.49	1.45	0.17	0.58	
		3	13.51	3.99	2.49	11.82	3.47	2.02	0.33	0.08	0.35	0.38	0.12	0.7	
		4	6.13	13.05	2.94	15.36	2.62	4.81	0.19	0.15	0.39	0.22	0.21	0.54	
		5	12.84	4.51	4.72	15.86	9.33	5.18	0.51	0.41	0.12	0.31	0.13	0.17	
Range			1.76-13.52			1.92-15.89			0.08-0.55			0.12-1.47			
Mean			7.60			6.86			0.33			0.38			
Galigkere-2	0-20	1	1.75	10.09	13.41	3.32	3.01	5.23	0.32	0.18	0.29	0.16	0.23	0.32	
		2	11.65	2.07	11.87	11.89	1.97	6.89	0.55	0.55	0.55	1.5	0.24	0.8	
		3	13.66	4.12	2.52	11.85	3.49	2.06	0.4	0.12	0.48	0.43	0.14	0.81	
		4	6.23	13.25	10.23	16.02	2.6	3.14	0.23	0.18	0.39	0.31	0.27	0.78	
		5	13.17	4.68	4.68	16.89	9.31	5.23	0.42	0.3	0.18	0.38	0.21	0.36	
20-40		1	1.71	9.98	13.35	3.28	3	5.21	0.28	0.14	0.25	0.15	0.21	0.26	
		2	11.52	2.02	11.81	11.85	1.92	6.83	0.51	0.51	0.51	0.13	0.22	0.5	
		3	13.47	4.05	2.46	11.81	3.47	2.01	0.38	0.11	0.44	0.38	0.11	0.79	
		4	6.21	13.19	10.19	15.93	2.43	3.05	0.21	0.15	0.37	0.26	0.23	0.74	
		5	13.12	4.66	4.62	16.78	9.14	5.13	0.38	0.26	0.15	0.31	0.17	0.33	
Range			1.71-13.66			1.92-16.89			0.11-0.55			0.11-1.5			
Mean			8.19			6.82			0.33			0.39			

Watershed	Depth (Cm)	Sample no	Fe (mg kg <sup>-1</sup> )			Mn (mg kg <sup>-1</sup> )			Cu (mg kg <sup>-1</sup> )			Zn (mg kg <sup>-1</sup> )		
			V	M	R	V	M	R	V	M	R	V	M	R
Galigkere-3	0-20	1	6.46	5.63	5.13	20.86	13.6	5.27	2.81	1.14	0.75	1.15	0.67	0.34
		2	6.36	4.38	3.52	20.22	17.4	3.44	2.25	1.29	0.34	1.64	0.58	0.29
		3	6.48	5.16	2.73	13.84	13.38	5.15	1.96	1.33	0.54	1.3	0.77	0.19
		4	6.08	4.67	2.6	16.8	8.26	2.68	2.37	0.98	1.02	2.01	0.81	0.45
		5	5.86	4.83	2.83	14.28	7.99	1.69	1.63	1.19	0.19	0.82	1.03	0.67
	20-40	1	6.32	5.58	5.06	20.81	13.25	5.52	2.75	1.04	0.72	1.04	0.65	0.29
		2	6.28	4.27	3.35	20.13	17.13	3.28	2.18	1.21	0.28	1.59	0.51	0.21
		3	6.44	5.02	2.66	13.65	13.29	5.03	1.88	1.19	0.53	1.21	0.73	0.11
		4	5.98	4.48	2.45	16.65	8.04	2.61	2.28	0.04	1	1.95	0.73	0.39
		5	5.78	4.77	2.71	14.19	7.83	1.55	1.56	1.03	0.17	0.77	1.01	0.64
	Range	2.45-6.48			1.55-20.86			0.04-2.81			0.11-2.01			
	Mean	4.80			10.93			1.25			0.82			
Haradagere	0-20	1	8.5	13.41	22.87	5.26	5.56	11.16	2.07	1.42	7.63	2.08	0.9	3.23
		2	12.38	16.19	25.67	6.45	7.34	12.78	1.85	1.7	4.12	1.17	0.5	0.94
		3	12.17	29.66	22.89	7.15	2.45	10.44	2.41	0.78	2.1	1.27	0.36	0.49
		4	5.28	23.01	18.45	7.1	4.66	1.76	1.4	2.6	1.7	2.11	0.54	0.78
		5	16.17	21.75	16.55	7.04	10.32	8.32	5.53	1.67	1.61	0.93	0.58	0.65
	20-40	1	8.3	13.29	22.76	5.24	5.53	11.14	2.02	1.41	7.59	2.05	0.7	3.21
		2	12.16	16.12	25.46	6.34	7.31	12.75	1.82	1.5	4.08	1.14	0.2	0.91
		3	12.08	29.61	22.76	7.03	2.42	10.41	2.38	0.76	1.6	1.24	0.34	0.44
		4	5.26	22.98	18.33	6.89	4.64	1.74	1.1	2.4	1.4	2.06	0.51	0.75
		5	16.12	21.56	16.43	6.99	10.27	8.3	5.49	1.65	1.56	0.89	0.55	0.63
	Range	5.26-29.66			1.74-12.78			0.76-7.63			0.2-3.23			
	Mean	17.61			7.16			2.52			1.07			
Singadahalli	0-20	1	2.56	4.21	5.89	2.76	13.59	17.89	1.01	1.1	1.02	0.36	0.58	0.6
		2	3.65	1.89	2.65	4.73	5.49	3.29	1.91	1.01	0.6	1.01	1.79	0.49
		3	2.8	2.75	4.48	4.01	15.28	10.49	1.03	0.47	0.56	1.23	0.41	1.29
		4	2.78	4.2	5.49	8.4	13.19	22.4	1.5	0.58	0.71	0.8	1.23	0.65
		5	4.4	3.59	6.37	4.77	4.01	20.29	1.28	1.64	0.93	0.55	3.01	0.76
	20-40	1	2.51	4.17	5.87	2.74	13.48	17.83	0.98	0.8	0.97	0.33	0.55	0.42
		2	3.6	1.85	2.62	4.68	5.44	3.23	1.85	0.78	0.42	0.88	1.72	0.43
		3	2.62	2.73	4.44	3.99	15.21	10.46	1.01	0.42	0.51	1.18	0.33	1.25
		4	2.74	4.17	5.47	8.2	13.08	22.12	1.32	0.56	0.67	0.6	1.21	0.61
		5	4.1	3.55	6.32	4.71	3.99	20.14	1.19	1.61	0.88	0.53	2.88	0.74
	Range	1.85-6.37			2.74-22.4			0.42-1.91			0.33-3.01			
	Mean	3.81			9.99			0.97			0.94			
Appanahalli	Range	1.71-29.66			1.55-22.4			0.04-7.63			0.11-3.23			
	Mean	8.98			8.83			1.10			0.76			

V = Valley, M= Midland, R = Ridge

TABLE 4

Status of soil microbial biomass carbon and nitrogen in soils of Appanahalli sub watershed of Tumkur district

Watershed	Depth (Cm)	Sample no	SMBC ( $\mu\text{g g}^{-1}$ )			SMBN ( $\mu\text{g g}^{-1}$ )		
			V	M	R	V	M	R
Appanahalli	0-20	1	556.70	476.42	390.00	64.51	53.21	39.02
		2	495.70	425.70	156.20	56.01	49.67	18.22
		3	524.53	425.60	286.51	61.20	49.65	32.54
		4	320.19	305.06	108.90	34.71	35.12	12.71
		5	505.90	407.20	445.02	59.02	45.25	52.15
	Range		305.06-556.70	12.71-64.51				
	Mean		388.64	44.19				
Galigkere-1	0-20	1	527.81	482.13	365.18	62.13	52.81	35.84
		2	518.98	416.98	287.23	60.08	49.87	32.67
		3	488.19	420.81	141.13	55.02	49.63	17.83
		4	239.89	365.18	129.32	31.16	32.41	14.31
		5	423.15	333.54	165.42	49.63	30.08	20.19
	Range		129.32-527.81	14.31-62.13				
	Mean		353.66	39.57				
Galigkere-2	0-20	1	445.02	356.71	285.12	52.15	37.57	32.06
		2	157.92	425.70	505.90	19.53	49.67	59.02
		3	205.00	284.50	325.60	22.55	31.96	36.01
		4	215.00	300.84	300.84	25.13	34.21	34.21
		5	195.06	220.00	108.90	24.01	27.00	12.71
	Range		108.9-505.9	12.71-59.02				
	Mean		288.80	33.18				
Galigkere-3	0-20	1	497.70	300.84	157.92	57.01	34.21	19.53
		2	556.67	280.50	125.00	64.51	32.03	17.51
		3	505.90	290.06	106.80	59.02	32.15	12.46
		4	519.23	476.42	170.60	62.32	53.21	19.60
		5	480.12	268.80	171.57	54.28	31.36	20.02
	Range		106.80-556.67	12.46-64.51				
	Mean		327.20	37.94				
Haradagere	0-20	1	524.53	213.45	567.99	61.20	24.90	66.27
		2	505.90	586.23	476.98	59.02	68.39	55.64
		3	515.24	476.98	312.45	60.11	66.64	36.45
		4	498.76	432.69	215.60	58.19	50.48	25.15
		5	556.78	129.87	345.98	64.96	15.15	40.36
	Range		129.87-586.23	15.15-68.39				
	Mean		423.96	50.19				



Watershed	Depth (Cm)	Sample no	SMBC ( $\mu\text{g g}^{-1}$ )			SMBN ( $\mu\text{g g}^{-1}$ )		
			V	M	R	V	M	R
Singadahalli	0-20	1	495.70	586.23	324.15	56.01	68.39	35.61
		2	556.70	407.20	321.05	64.51	45.25	34.98
		3	505.90	524.53	325.60	59.02	61.20	36.01
		4	524.53	241.77	320.19	61.20	28.21	34.71
		5	476.42	356.71	305.06	53.21	37.57	35.12
	Range	241.77-586.23	28.21-68.39					
	Mean	418.11	47.40					
Appanahalli	Range	106.80-586.23	12.46-68.39					
	Mean	366.73	42.08					

V = Valley, M = Midland, R= Ridge

lowland or valley of watershed when compared to upland.

#### Available Micronutrients

Available Fe, Mn, Cu and Zn content in the soil of Appanahalli sub-watershed ranged from 1.36-29.66 mg kg<sup>-1</sup>, 1.55-22.4 mg kg<sup>-1</sup>, 0.04-7.63 mg kg<sup>-1</sup> and 0.11 to 3.23 mg kg<sup>-1</sup>, respectively (Table 3). Available Fe, Mn, Cu and Zn were in sufficiency range for surface soils of all micro watersheds. The availability of micronutrients content was higher on the surface soils with increase in organic matter because organic matter acts as a chelating agent for complexation of these micronutrients, which reduces their adsorption, oxidation and precipitation into unavailable forms (Mahesh Kumar *et al.*, 2011). Due to higher microbial activity in the surface soil Mn content were also observed sufficient range in the study area. Results were in line with the findings of Murthy *et al.* (1997). Srikanth *et al.* (2008) reported higher available manganese content in soils originated from granite gneiss parent material with semi-arid climate. The content of Zn increases with high organic carbon content but decreases with increase in pH. Since, some of the grid points show alkaline soil reaction, low in OC and dominated by CaCO<sub>3</sub>, zinc may be precipitated as hydroxides and carbonates and as a result, their solubility and mobility might have decreased and reduced the availability. Similar results were reported by Patil *et al.* (2019).

#### Soil Microbial Biomass Carbon and Nitrogen

In Appanahalli sub watershed, microbial biomass carbon and nitrogen ranged from 106.8 to 586.23  $\mu\text{g g}^{-1}$  and 12.46 to 68.29  $\mu\text{g g}^{-1}$ , respectively (Table 4). Haradagere micro watershed recorded highest microbial biomass carbon and nitrogen with a mean value of 423.96 and 50.19  $\mu\text{g g}^{-1}$ , respectively among all six micro watersheds. Valley portion of watershed has higher microbial biomass carbon and nitrogen than ridge because of nutrient enrichment in valley portion, in particular, carbon and nitrogen. Crops in intercropping, crop rotation and various cropping systems possess unique exudate deposition near its roots. These organic and inorganic substances contribute key nutrients to microbial community. This interpretation is in accordance with the study of Duchene *et al.* (2017).

A thorough understanding on the fertility status of soil is essential for planning of better management for the resource. It is evident from the results that soils of Appanahalli sub watershed were very acidic to moderately alkaline in nature. Among the three major landforms studied, pH values in lowland were better than midland and upland physiography. The soils of sub-watershed were non saline. In all the micro watersheds, soil pH and EC of soil increased with depth. The higher SOC and available N was observed on the surface soil layer and it is decreasing with increasing soil depth. The available P<sub>2</sub>O<sub>5</sub> and K<sub>2</sub>O

contents were low to medium in Appanahalli sub watershed. Available Fe, Mn, Cu, Zn and soil microbial biomass carbon and nitrogen were in sufficiency range for surface soils of all micro watersheds. The availability of micronutrients was increased on the surface soils with increase in organic matter. Since nutrients are expected to be available at a favourable soil reaction, proper and adequate measures like amelioration of soil acidity / alkalinity need to be adopted for bringing these soils to a better fertility and productivity conditions. Balanced fertilization and correcting soil reaction while maintaining soil health is a key to soil quality and thereby sustainable crop production.

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## Influence of Seed Treatment and Antioxidant Spray on Crop Growth, Seed Yield and Quality in White Cowpea [*Vigna unguiculata* (L.) Walp] cv. IT - 38956 - 1

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### ABSTRACT

An experiment was conducted at Department of Seed Science and Technology, UAS, GKVK, Bengaluru during *khariif*-2017 to study the influence of seed treatment and antioxidant spray on crop growth and seed yield in white cowpea. The seed treatment with micronutrient like FeSO<sub>4</sub> 1 per cent, CaCl<sub>2</sub> 1 per cent, Sulphur 1 per cent and antioxidant spray *viz.*, salicylic acid, ascorbic acid, @ 100 ppm and @ 150 ppm with their combinations with a control includes 15 treatments replicated three times under RCBD. The results revealed that seed treatment with (FeSO<sub>4</sub> 1 % + CaCl<sub>2</sub> 1 % + Sulphur 1 %) and foliar spray of (salicylic acid @ 100 ppm + ascorbic acid @ 100 ppm) has registered higher crop growth and seed yield *viz.*, plant height at 30 DAS (15.41 cm), 60 DAS (27.76 cm) and at harvest (35.06 cm), number of leaves plant<sup>-1</sup> at 60 DAS (19.20), number of branches plant<sup>-1</sup> at 60 DAS (11.07) and at harvest (12.15), numbers of effective nodules at 60 DAS (16.68), seed size (5.86 cm<sup>3</sup>), number of pods plant<sup>-1</sup> (10.47), no of pods cluster<sup>-1</sup> (3.27), pod length (14.86 cm), number of seeds pod<sup>-1</sup> (10.00), seed yield plot<sup>-1</sup> (0.83 kg), seed germination (92.50 %), test weight (151.90 g), mean seedling length (38.42 cm), mean seedling dry weight (52.83 mg), SVI-I (3553), SVI-II (4252), TDH (4.00 A<sub>480</sub> nm) and lower electrical conductivity (515.70 μS cm<sup>-1</sup>) compared to control and other treatments.

*Keywords* : Cowpea, Seed treatment, Antioxidant spray

THE cowpea has been grown in India, since ancient times. It was noticed in old ethics and vedic times. It is one of species of the widely cultivated genus *Vigna* and belongs to family Leguminosae and native of Central Africa which is drought tolerant and warm weather crop. The cowpeas are well adopted to the drier regions of the tropics and known by many vernacular names like *Lobia* (Hindi), *Alasande* (Kannada) and *Karamani* (Tamil and Telugu) (Javeeda, 2001).

Cowpea is an important crop of the world covering an area of about 143.5 lakh ha with a production of 72.6 lakh tonnes and productivity of 585.6 kg ha<sup>-1</sup>. In India, cowpea is grown in an area of 13 lakh ha with

a production of 31 lakh tonnes and an average productivity of 238 kg ha<sup>-1</sup>. The major cowpea growing states are Gujarat, Uttar Pradesh, Rajasthan, Tamil Nadu, Andhra Pradesh and Karnataka. In Karnataka, cowpea is grown in an area of 0.99 lakh ha with a production of 0.46 lakh tonnes and productivity of 426 kg ha<sup>-1</sup> (Anonymous, 2015).

Pro-anthocyanidins and tannins are the major compounds involved in seed coat pigmentation. Cowpea has a relatively low cost and high quality source of protein. The crop growth, seed yield and quality is generally reduced due to presence of anti-nutrients such as phytates, fibres, trypsin inhibitors, lectins, tannins and polyphenols. These compounds

reduces digestibility and quality of protein (Gatehouse and Boulter, 1989). Phytic acid blocks absorption of micronutrients such as P, Ca, Mg, Fe and Zn and negatively affects the absorption of lipids, proteins and also inhibits important digestive enzymes such as amylase, pepsin and trypsin.

Seed treatment with micronutrient promote the strong, steady growth of crops that produce higher seed yields and increase harvest quality maximizing a plant's genetic potential (Prajapati *et al.*, 2017) and also antioxidant has shown many important function in plant and can change physiological behaviour of plant (Kiran and Channakeshava, 2017). Foliar application with relative low concentration of antioxidant also promoted and influenced the growth, development, differentiation of plants and enhanced the plant growth, seed yield parameters (Siamak *et al.*, 2014) and quality parameters (Hashmi *et al.*, 2012).

#### MATERIAL AND METHODS

An experiment was conducted to study the influence of seed treatment and antioxidant spray on crop growth, seed yield and quality in white cowpea [*Vigna unguiculata* (L.) Walp] cv. IT-38956-1 during Kharif, 2017 at E6-Block, GKVK campus, University of Agricultural Sciences, Bangalore which is situated between 13° 15' N latitude and 77° 32' East longitudes, at 930 m altitude above Mean Sea Level (MSL), which represents the Agro-climate of Eastern Dry Zone of Karnataka. The soil of the experimental sites was red sandy loam in texture. The moisture content at field capacity was 18.63 per cent with a bulk density of 1.43 g cc<sup>-1</sup>. The soil of the site was slightly acidic in reaction (pH 5.8 to 6.1) and electrical conductivity was medium (0.32 to 0.36 dS m<sup>-1</sup>). The organic carbon content was low (0.42 to 0.48%). The available nitrogen was low (228.2 kg ha<sup>-1</sup>). Phosphorus was high (62.5 kg ha<sup>-1</sup>) and potassium was also high (256.1 kg ha<sup>-1</sup>). Cowpea variety cv. IT-38956-1 was used in the experimentation. There were fifteen treatment combination of seed treatment with micronutrient (FeSO<sub>4</sub> 1%, CaCl<sub>2</sub> 1% and Sulphur 1%) and antioxidant spray (Salicylic acid + Ascorbic acid) @ 100 ppm and @ 150 ppm replicated thrice in randomised complete block design. Treatments are

T<sub>0</sub> - Control (no seed treatment and no foliar spray of antioxidants), T<sub>1</sub> - Seed treatment with FeSO<sub>4</sub> 1% and foliar spray of (salicylic acid @ 100 ppm + ascorbic acid @ 100 ppm), T<sub>2</sub> - Seed treatment with FeSO<sub>4</sub> 1 per cent and foliar spray of (salicylic acid @ 150 ppm + ascorbic acid @ 150 ppm), T<sub>3</sub> - Seed treatment with CaCl<sub>2</sub> 1 per cent and foliar spray of (salicylic acid @ 100 ppm + ascorbic acid @ 100 ppm), T<sub>4</sub> - Seed treatment with CaCl<sub>2</sub> 1 per cent and foliar spray of (salicylic acid @ 150 ppm + ascorbic acid @ 150 ppm), T<sub>5</sub> - Seed treatment with Sulphur 1 per cent and foliar spray of (salicylic acid @ 100 ppm + ascorbic acid @ 100 ppm), T<sub>6</sub> - Seed treatment with Sulphur 1 per cent and foliar spray of (salicylic acid @ 150 ppm + ascorbic acid @ 150 ppm), T<sub>7</sub> - Seed treatment with (FeSO<sub>4</sub> 1 per cent + CaCl<sub>2</sub> 1 per cent) and foliar spray of (salicylic acid @ 100 ppm + ascorbic acid @ 100 ppm), T<sub>8</sub> - Seed treatment with (FeSO<sub>4</sub> 1% + CaCl<sub>2</sub> 1%) and foliar spray of (salicylic acid @ 150 ppm + ascorbic acid @ 150 ppm), T<sub>9</sub> - Seed treatment with (CaCl<sub>2</sub> 1% + Sulphur 1%) and foliar spray of (salicylic acid @ 100 ppm + ascorbic acid @ 100 ppm), T<sub>10</sub> - Seed treatment with (CaCl<sub>2</sub> 1% + Sulphur 1%) and foliar spray of (salicylic acid @ 150 ppm + ascorbic acid @ 150 ppm), T<sub>11</sub> - Seed treatment with (FeSO<sub>4</sub> 1% + Sulphur 1%) and foliar spray of (salicylic acid @ 100 ppm + ascorbic acid @ 100 ppm), T<sub>12</sub> - Seed treatment with (FeSO<sub>4</sub> 1% + Sulphur 1%) and foliar spray of (salicylic acid @ 150 ppm + ascorbic acid @ 150 ppm), T<sub>13</sub> - Seed treatment with (FeSO<sub>4</sub> 1% + CaCl<sub>2</sub> 1% + Sulphur 1%) and foliar spray of (salicylic acid @ 100 ppm + ascorbic acid @ 100 ppm), T<sub>14</sub> - Seed treatment with (FeSO<sub>4</sub> 1% + CaCl<sub>2</sub> 1% + Sulphur 1%) and foliar spray of (salicylic acid @ 150 ppm + ascorbic acid @ 150 ppm). Sowing and cultural practices were carried out as per the package of practice except seed treatment and antioxidants was sprayed at 40 DAS. Crop growth, seed yield and quality observations were recorded in field.

#### RESULTS AND DISCUSSION

##### Crop Growth

Crop growth parameters differed significantly among the treatments (Table 1). The seed treatment with

TABLE 1

Influence of seed treatment and antioxidant spray on crop growth attributes in white cowpea cv. IT-38956-1

Treatments	Plant height (cm)			Number of leaves plant <sup>-1</sup>	Number of branches plant <sup>-1</sup>		Number of effective nodules
	30 DAS	60 DAS	At harvest	60 DAS	60 DAS	At harvest	60 DAS
T <sub>0</sub>	10.21	21.21	27.00	11.60	8.60	9.00	9.00
T <sub>1</sub>	14.25	21.98	30.49	12.60	8.80	10.07	15.10
T <sub>2</sub>	13.52	24.26	29.85	13.67	9.33	9.73	10.00
T <sub>3</sub>	12.63	21.83	29.50	13.20	9.33	9.60	13.00
T <sub>4</sub>	14.30	24.01	30.00	15.00	9.47	10.13	13.33
T <sub>5</sub>	12.56	21.33	30.65	12.87	9.73	9.75	11.00
T <sub>6</sub>	13.95	22.57	28.79	15.00	9.37	10.53	9.00
T <sub>7</sub>	15.18	26.23	31.29	13.07	9.47	10.00	15.33
T <sub>8</sub>	15.03	25.99	31.18	13.73	8.67	9.88	12.30
T <sub>9</sub>	14.73	24.97	31.81	12.40	9.33	10.84	9.60
T <sub>10</sub>	14.65	24.65	31.52	14.47	10.40	10.43	9.00
T <sub>11</sub>	15.28	27.23	34.97	15.40	10.33	11.29	15.60
T <sub>12</sub>	15.16	26.51	33.90	14.60	8.93	10.25	11.33
T <sub>13</sub>	15.41	27.76	35.06	19.20	11.07	12.15	16.68
T <sub>14</sub>	14.41	25.55	30.01	13.93	9.53	10.50	15.33
S.Em±	0.75	1.51	1.47	0.90	0.40	0.48	0.85
CD (P= 0.05)	2.17	4.38	4.27	2.62	1.17	1.39	2.59
CV (%)	9.24	10.75	8.21	11.14	7.41	8.09	9.78

(FeSO<sub>4</sub> 1% + CaCl<sub>2</sub> 1% + Sulphur 1%) and foliar spray of (salicylic acid @ 100 ppm + ascorbic acid @ 100 ppm) (T<sub>13</sub>) recorded significant higher plant height of (15.41 cm @ 30 DAS, 27.76 cm @ 60 DAS and 35.06 cm at harvest), number of leaves plant<sup>-1</sup> (19.20 @ 60 DAS), number of branches plant<sup>-1</sup> (11.07 @ 30 DAS and 12.15 at harvest) and number of effective nodules (16.68 @ 60 DAS) as compared to control (T<sub>0</sub>) (10.21 cm, 21.21 cm, 27.00 cm, 11.60, 8.60, 9.00 and 9.00 respectively) Fig. 1.

It is evident from the results that seed treatment with micronutrient which modifies the physiological and biochemical nature of seeds and antioxidant spray with salicylic acid and ascorbic acid enhance the accumulation of chlorophyll, betacyanin, total phenols and antioxidant activity which would likely to increase as indicated by Manjunath *et al.* (2011) in chickpea

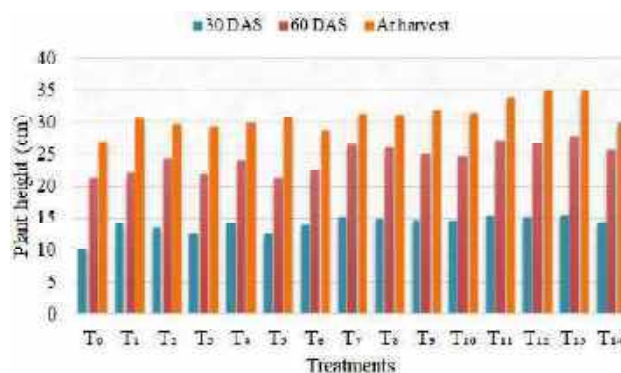


Fig. 1 : Influence of seed treatment and antioxidant spray on plant height at 30 DAS, 60 DAS and at harvest in white seeded cowpea cv. IT-38956-1

and Khandakar *et al.* (2011) in red amaranth. The results are in same line with studies made by Prajapati *et al.* (2017) in blackgram and Rehman *et al.* (2011) in rice. More number of leaves per plant might be

TABLE 2  
Effect of seed treatment and antioxidant spray on seed yield attributes in white cowpea cv. IT 38956-1

Treatments	Seed size (cm <sup>3</sup> )	No. of pods plant <sup>-1</sup>	No. of pods cluster <sup>-1</sup>	Pod length (cm)	No of seeds pod <sup>-1</sup>	Seed yield plot <sup>-1</sup> (kg)
T <sub>0</sub>	5.00	5.05	2.20	8.46	4.20	0.61
T <sub>1</sub>	5.43	5.60	2.40	10.42	8.20	0.72
T <sub>2</sub>	5.13	5.60	2.33	10.20	8.20	0.66
T <sub>3</sub>	5.31	5.30	2.40	9.86	6.60	0.63
T <sub>4</sub>	5.46	5.40	2.33	10.64	7.20	0.75
T <sub>5</sub>	5.10	5.40	2.45	9.48	6.00	0.63
T <sub>6</sub>	5.38	5.87	2.53	10.32	6.80	0.71
T <sub>7</sub>	5.63	6.80	2.47	11.58	8.40	0.75
T <sub>8</sub>	5.30	7.27	2.53	11.36	7.40	0.80
T <sub>9</sub>	5.59	5.93	2.40	11.28	7.60	0.78
T <sub>10</sub>	5.56	6.60	2.53	11.02	8.60	0.81
T <sub>11</sub>	5.73	9.40	3.20	14.34	9.80	0.82
T <sub>12</sub>	5.68	7.93	2.80	13.76	9.20	0.81
T <sub>13</sub>	5.86	10.47	3.27	14.86	10.00	0.83
T <sub>14</sub>	5.54	7.67	2.80	10.94	8.20	0.80
S.Em±	0.12	0.45	0.13	0.90	0.68	0.048
CD (P= 0.05)	0.374	1.31	0.40	2.749	2.08	0.145
CV (%)	3.22	11.73	9.35	11.409	12.50	10.54

due to cell division and cell elongation by antioxidants. Similar finding were reported by Mohsen (2014) in tomato plants, Seadh and Metwally (2015) in wheat. More number of branches plant<sup>-1</sup> might be due to cell division and cell elongation by seed treatment with micronutrient and antioxidant spray. Similar findings were reported by Seadh and Metwally (2015) in wheat. More number of effective nodules might be due to seed treatment with micronutrient which enhances the symbiotic relationship between rhizobia and plant. Thus the development of nodules, while depend on rhizobia, is a well-coordinated development process of the plant which is reported by Mylona *et al.* (1995).

### Seed Yield

Seed yield parameters differed significantly among the treatments (Table 2). The seed treatment with

(FeSO<sub>4</sub> 1% + CaCl<sub>2</sub> 1% + Sulphur 1%) and foliar spray of (salicylic acid @ 100 ppm + ascorbic acid @ 100 ppm) (T<sub>13</sub>). Registered significantly higher seed size of (5.86 cm<sup>3</sup>), number of pods plant<sup>-1</sup> (10.47), number of pods cluster<sup>-1</sup> (3.27), pod length (14.86 cm), number of seeds pod<sup>-1</sup> (10.00) and seed yield plot<sup>-1</sup> (0.83 kg) compared to control (T<sub>0</sub>) which recorded the lowest seed yield attributes (5.00 cm<sup>3</sup>, 5.05, 2.20, 8.46, 4.20 and 0.61 kg respectively) Fig. 2.

The increase in seed yield attributes like seed size is due to presence of higher amount of stored food material which reflected in higher seed size indicated by Manjunath *et al.* (2011) in chickpea, Prajapati *et al.* (2017) in blackgram and The results are in same line with studies made by Chavan and Tagad. (2013) in soybean and Abbas *et al.* (2009) in wheat. Increase in number of pods plant<sup>-1</sup>, number of pods cluster<sup>-1</sup>,

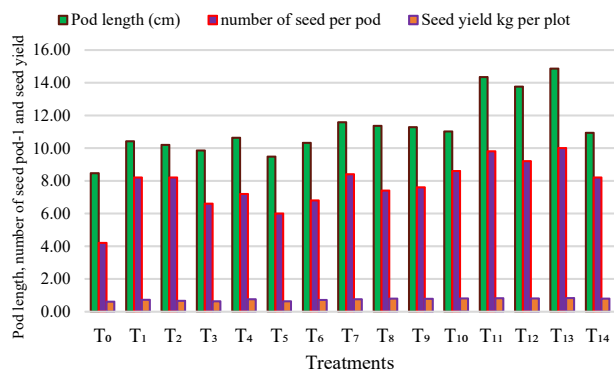


Fig. 2. Influence of seed treatment and antioxidant spray on pod length (cm), number of seeds pod<sup>-1</sup> and seed yield plot<sup>-1</sup> in white seeded cowpea cv. IT 38956-1

pod length and number of seeds pod<sup>-1</sup> might be attributed to bio regulator effect of ascorbic acid and salicylic acid on physiological and biochemical process in plants such as ion uptake, cell elongation, cell division, cell differentiation, sink and source regulation, enzymatic activities, protein synthesis and photosynthetic activity, as well as, increase in antioxidant capacity of plant.

The results are in agreement with research findings of Amal *et al.* (2009) in pea, Abido *et al.* (2015) in sugar beet, Bhingarde *et al.* (2015) in groundnut and Gad el-hak *et al.* (2012) in peas. Increase in seed yield is mainly dependent on source sink relation. As the reproductive get more photosynthetic assimilate, an increase in seed yield is resulted. Improvement in seed yield components might be due to improved vegetative growth. The overall improvement in growth and yield components may be due to synergistic effect of combined use of antioxidants. Similar results were reported by Shabana *et al.* (2015) in sweet pepper, Amal *et al.* (2009) in pea, Bharati *et al.* (2010) in soyabean, Prajapati *et al.* (2017) in blackgram and Tabatabaei (2013) in sorghum and Nagraj *et al.*, 2017 in pigeonpea.

### Seed Quality

Seed quality parameters differed significantly among the treatments (Table 3 and 4). The seed quality parameters were significantly higher in seed treatment with (FeSO<sub>4</sub> 1% + CaCl<sub>2</sub> 1% + Sulphur 1%) and foliar spray of (salicylic acid @ 100 ppm +

TABLE 3

Influence of seed treatment and antioxidant spray on seed quality attributes in white cowpea cv. IT-38956-1

Treatments	Germination (%)	Test weight (g)	Mean seedling length (cm)	SVI-I
T <sub>0</sub>	80.50	127.45	23.12	1861
T <sub>1</sub>	81.50	137.05	28.82	2348
T <sub>2</sub>	81.50	132.45	32.51	2649
T <sub>3</sub>	84.00	135.65	31.75	2667
T <sub>4</sub>	83.00	138.15	28.38	2355
T <sub>5</sub>	84.50	133.35	29.00	2450
T <sub>6</sub>	83.00	130.05	27.00	2241
T <sub>7</sub>	84.00	141.10	31.45	2766
T <sub>8</sub>	84.00	140.45	32.16	2701
T <sub>9</sub>	87.00	138.95	28.14	2448
T <sub>10</sub>	88.50	142.80	34.79	3078
T <sub>11</sub>	92.00	147.15	35.00	3220
T <sub>12</sub>	89.50	132.15	34.28	3068
T <sub>13</sub>	92.50	151.90	38.42	3553
T <sub>14</sub>	90.00	139.30	33.37	3003
S.Em±	1.89	24.814	1.06	71.20
CD (P= 0.05)	5.70	74.796	3.20	214.64
CV (%)	3.12	2.545	4.81	3.74

ascorbic acid @ 100 ppm) (T<sub>13</sub>). Seed germination of (92.50%), test weight (151.90 g), mean seedling length (38.42 cm), mean seedling dry weight (80.78 mg), SVI-I (3553), SVI-II (7472), total dehydrogenase activity (4 A<sub>480</sub> nm) and lowest electrical conductivity (515.70 μS cm<sup>-1</sup>) compared to control (T<sub>0</sub>) (80.50%, 127.45 g, 23.12 cm, 52.83 mg, 1861, 4252, 2.52 A<sub>480</sub> nm and 876.00 μS cm<sup>-1</sup> respectively) Fig. 3 & 4.

Germination percentage differed significantly among treatments. Maximum germination was noticed in T<sub>13</sub>. This might be due to enhanced source to sink relation. Seeds contain greater metabolites for resumption of embryonic growth during germination and better accumulation of food reserves like protein and carbohydrates as reported by Bharati, *et al.* (2013) in soybean, Chavan and Tagad (2015) in soybean and Tabatabaei (2013) in sorghum seed. Higher test weight was recorded in T<sub>13</sub>. This might be due to presence of higher amount of stored food material in seed as indicated by Manjunath *et al.* (2011) in chickpea,



TABLE 4

Influence of seed treatment and antioxidant spray on seed quality attributes in white cowpea cv. IT-38956-1.

Treatments	Mean seedling dry weight (mg)	SVI-II	TDH (A <sub>480</sub> nm)	EC (μS cm <sup>-1</sup> )
T <sub>0</sub>	52.83	4252	2.52	876.00
T <sub>1</sub>	56.48	4603	2.57	707.40
T <sub>2</sub>	61.55	5016	2.80	744.30
T <sub>3</sub>	64.40	5409	2.97	757.10
T <sub>4</sub>	63.03	5231	2.71	713.15
T <sub>5</sub>	56.50	4773	2.80	800.65
T <sub>6</sub>	57.17	4744	2.68	754.80
T <sub>7</sub>	60.27	5061	3.38	823.15
T <sub>8</sub>	63.40	5324	3.48	661.00
T <sub>9</sub>	71.62	6230	3.67	674.70
T <sub>10</sub>	68.09	6025	3.10	729.30
T <sub>11</sub>	74.63	6865	3.74	571.40
T <sub>12</sub>	73.84	6608	3.59	634.20
T <sub>13</sub>	80.78	7472	4.00	515.70
T <sub>14</sub>	73.85	6615	3.21	710.05
S.Em±	1.37	86.78	0.06	20.61
CD (P= 0.05)	4.15	261.60	0.19	62.14
CV (%)	2.98	2.18	2.85	4.09

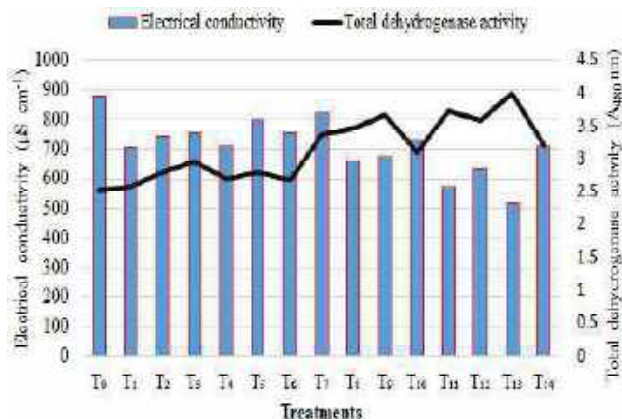


Fig. 3: Influence of seed treatment and antioxidant spray on electrical conductivity (μS cm<sup>-1</sup>) and total dehydrogenase activity (A<sub>480</sub> nm) in white seeded cowpea cv. IT 38956-1



Fig. 4: Influence of seed treatment and antioxidant spray on seedling vigour index- II and mean seedling dry weight (mg) in white seeded cowpea cv. IT 38956-1

Prajapati *et al.* (2017) in blackgram and the results are in same line with studies made by Chavan and Tagad. (2015) in soybean and Abbas *et al.* (2009) in wheat. Increase in mean seedling length, mean seedling dry weight, seedling vigour index-I and seedling dry weight (mg) might be due to increase in root length and shoot length which in turn is attributed to presence of higher amount of stored food material as indicated by Bhaarati *et al.* (2013) in soybean and Chavan and Tagad (2015) in soybean. The results are in same line with studies made by Khandaker (2011) in red amaranth and Hashmi *et al.* (2012) in fennel. Highest TDH activity registered in T<sub>13</sub> might be due to application of antioxidant as foliar spray which resulted in enhanced source to sink relation leads to better accumulation of food reserves like protein. Lowest electrical conductivity documented in T<sub>13</sub>. This might be due to production of quality seeds with high cell wall integrity and low leakage of metabolites from seeds when soaked in water. Similar results were also obtained by Bhingarde *et al.* (2015) in groundnut and Bhaarati *et al.* (2013) in soybean.

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## Green Forage Yield, Nutritional Value and Economics of Dinanath Grass Genotypes as Influenced by Nitrogen Levels

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### ABSTRACT

The field experiment was conducted during *kharif* season of 2021 at Zonal Agricultural Research Station, Vishwesharaiah Canal Farm, Mandya, University of Agricultural Sciences, Bangalore, Karnataka to assess the performance of Dinanath grass genotypes under different nitrogen levels. The experiment was laid out in randomized complete design with factorial concept with 30 treatment combinations and replicated thrice. Among genotypes, JHD-19-4 recorded significantly higher green forage (305.1 q ha<sup>-1</sup>), dry matter (73.2 q ha<sup>-1</sup>), crude protein (4.50 q ha<sup>-1</sup>), total digestible crude protein (3.7 q ha<sup>-1</sup>), crude fibre yield (19.0 q ha<sup>-1</sup>) and nitrogen use efficiency (572.8 kg GFY/kg of nitrogen) over rest of the genotypes. Application of nitrogen 90 Kg ha<sup>-1</sup> recorded significantly higher green forage (296.7 q ha<sup>-1</sup>), dry matter (78.7 q ha<sup>-1</sup>), crude protein (5.6 q ha<sup>-1</sup>), total digestible crude protein (4.8 q ha<sup>-1</sup>), crude fibre yield (19.1 q ha<sup>-1</sup>) and net monetary returns (Rs.34870 ha<sup>-1</sup>). The higher nitrogen use efficiency was noticed with nitrogen @ 30 kg ha<sup>-1</sup> (666.8 kg GFY/kg nitrogen).

**Keywords :** Dinanath grass, Nitrogen levels, Green fodder yield, Dry matter yield, Crude protein yield and total digestible crude protein

**D**INANATH grass (*Pennisetum pedicellatum* Trin.) is annual tufted grass, quick growing, leafy, luscious, thin stem grows well in poor and eroded soil and tolerance to drought conditions (Noitsakis *et al.*, 1994). The grass belongs to family Poaceae and widely distributed in West Africa and India. In India, it is cultivated in Karnataka, Maharashtra, Andhra Pradesh, Bihar, Chhattisgarh, Jharkhand, Odisha and West Bengal (Nayar *et al.*, 2009 and Upadhyaya *et al.*, 2014). Dinanath grass is widely used as green fodder for animal feed, as hay and silage making and also providing good quality forage for maintaining nutritional security in animals health during lean situations. Besides, as forage crop, it is also used as ornamental, soil erosion control and bio-energy crop and improve the physical and chemical properties of the soil (Kumar & Jena, 1996 and Kumar & Ghosh, 2018).

Dinanath grass is also rich in sodium, potassium, phosphorus and calcium. It has potential to be used in alleviating macro and micro-nutrients deficiencies in animals (Mustapha *et al.*, 2018; Suleiman *et al.*, 2020). The quality depends upon the stage of harvest (Asmare *et al.*, 2017 and Tilahun *et al.*, 2017) and nutrient management. Among nutrients nitrogen management plays a pivotal role in enhancing quantity and quality of the fodder crop. The Nitrogen promotes vegetative growth and improves the quality by increasing the crude protein content. Since, it is a constituent of amino acid, the deficiency of this in fodder crops may cause severe disorders in animal health (Midha *et al.*, 2015). Keeping these things in view, it is essential to find out the optimum dose of nitrogen for fetching both quantitative and qualitative fodder. Hence, the present investigation was undertaken to

study the response of Dinanath grass genotypes to varied nitrogen levels for enhancing the green forage yield and quality.

### MATERIAL AND METHODS

The experiment was carried out during *khariif* season of 2021 at Zonal Agricultural Research Station, Vishwesharaiah Canal Farm, Mandya, University of Agricultural Sciences, Bangalore, Karnataka to optimise the nitrogen requirement for higher green forage yield and quality in genotypes of Dinanath grass. The soil of the experimental site is red sandy loam in texture with neutral in reaction (pH-7.36) and low in available nitrogen (234 kg N ha<sup>-1</sup>), medium in available phosphorus (38.7 kg ha<sup>-1</sup>) and potassium (153.4 K<sub>2</sub>O kg ha<sup>-1</sup>). The experiment is consisted of 15 treatment combinations including five genotypes (V<sub>1</sub>- JHD-19-4, V<sub>2</sub>- BAU-DN-110-18-2, V<sub>3</sub>- BAU-DN-109-8, V<sub>4</sub>- BAU-DN-103-18-2, V<sub>5</sub>: Bundel Dinanath-2 (National check) and three nitrogen levels (30, 60 and 90 N Kg ha<sup>-1</sup>) was laid out in factorial randomized block design and

replicated thrice. The crop was sown during the second week of July with a row spacing of 30 cm and 10 cm between plants. The recommended dose of phosphorus (60 Kg ha<sup>-1</sup>) and potassium (40 Kg ha<sup>-1</sup>) was applied at the time of sowing. The nitrogen was applied in the form of urea as per the treatment and applied 50 per cent as basal at the time of sowing and remaining 50 per cent at 30 days after sowing. The cultural practices were followed as per local recommended package of practices for establishment of crop. The crop was harvested at flowering stage and immediately after the harvest, the green fodder yield was recorded respectively as per treatment. The known quantity of fresh sample was taken and kept in thermo statically controlled oven at 60 ± 2 °C temperature and dried till it attained constant weight for the estimation of dry matter content, yield and as well as other quality parameters. The nitrogen use efficiency (NUE) was worked out using following formula and expressed in Kg green fodder per Kg of nitrogen applied. The total digestible crude protein yield (TDCPY) was

TABLE 1  
Growth and yield of Dinanath grass genotypes as influenced by nitrogen levels recorded at harvest

Genotypes	Plant height (cm)	Leaf Stem ratio	Green Forage yield (q/ha)	Dry matter yield (q/ha)	Green Forage yield (q/ha/day)	Dry Matter yield (q/ha/day)
JHD-19-4	96.0	0.27	305.1	73.2	4.4	1.00
BAU-DN-110-18-2	75.4	0.19	217.9	49.9	3.2	0.73
BAU-DN-109-8	79.7	0.21	224.4	54.2	3.1	0.75
BAU-DN-103-18-2	87.0	0.22	254.1	58.4	3.6	0.89
Bundel Dinanath-2	84.1	0.22	257.6	60.6	3.5	0.82
S.Em±	2.23	0.006	8.03	2.38	0.13	0.04
C.D at 5%	6.49	0.018	23.39	6.94	0.38	0.12
<i>Nitrogen Levels (Kg/ha)</i>						
30	68.6	0.20	200.1	39.6	2.8	0.56
60	87.0	0.22	258.9	60.4	3.7	0.86
90	97.8	0.24	296.7	78.7	4.2	1.10
S. Em±	1.73	0.005	6.22	1.85	0.10	0.03
C.D at 5 %	5.03	0.014	18.12	5.38	0.29	0.08
Interaction						
S. Em±	3.86	0.01	13.91	4.13	0.23	0.06
C.D at 5%	NS	NS	NS	NS	NS	NS

calculated using following equation adopted by Iqbal *et al.* (2013). The economics was worked out with prevailing market price and input cost. The statistical analysis of data was carried out for interpretation of the results and draw valuable conclusion.

## RESULTS AND DISCUSSION

### Green Forage Yield (q/ha)

The green forage yield of Dinanath grass genotypes was significantly influenced by nitrogen levels (Table 1). Among genotypes significantly higher green forage yield was noticed with JHD-19-4 (305.1 q ha<sup>-1</sup>) and superior over rest of genotypes. The lower green forage yield was observed with genotype BAU-DN-110-18-2 (217.9 q ha<sup>-1</sup>). Application of nitrogen at 90 Kg ha<sup>-1</sup> recorded significantly higher green forage yield (296.7 q ha<sup>-1</sup>) followed by 60 Kg N ha<sup>-1</sup> (258.9 q ha<sup>-1</sup>). The interaction between genotypes and nitrogen levels was found non-significant. Nitrogen is major plant nutrient, plays a pivotal role in cell division, cell

elongation and differentiation, which leads to better root proliferation and luxuriant growth it is evidenced by higher plant height and leaf stem ratio and resulted higher green forage yield. The similar results were reported by Abraham *et al.* (1980a), Abraham *et al.* (1980b), Reddy *et al.* (1981), Tyagi & Singh (1986), Yadav & Sharma (1986), Bhagat *et al.* (1986), Tripathi & Singh (1991), Iqbal *et al.* (2013), Midha *et al.* (2015), Shekara *et al.* (2022) and Singh *et al.* (1997).

### Dry Matter Yield

The dry matter yield of Dinanath grass genotypes was significantly influenced by nitrogen levels recorded at harvest (Table 1), Among genotypes, JHD-19-4 recorded significantly higher dry matter yield (73.2 q ha<sup>-1</sup>) over other genotypes. Application of Nitrogen 90 Kg ha<sup>-1</sup> recorded higher dry matter yield (78.7 q ha<sup>-1</sup>) followed by 60 N Kg ha<sup>-1</sup> (60.4 q ha<sup>-1</sup>). The interaction between genotypes and nitrogen levels was found to be non-significant. Since, nitrogen is an integral component of chlorophyll and plays a primary role in photosynthesis and helped

TABLE 2  
Quality parameters of Dinanath grass genotypes as influenced by nitrogen levels at harvest

Genotypes	Crude Protein (%)	Crude fibre (%)	Dry Matter (%)
JHD-19-4	6.1	27.7	24.2
BAU-DN-110-18-2	7.0	26.4	22.6
BAU-DN-109-8	6.8	26.8	23.8
BAU-DN-103-18-2	5.3	25.9	22.4
Bundel Dinanath-2	7.2	25.3	23.2
S. Em±	0.14	0.42	0.41
C.D at 5%	0.40	1.21	1.91
<i>Nitrogen Levels (Kg/ha)</i>			
30	5.9	29.1	19.9
60	6.5	25.9	23.4
90	7.2	24.2	26.5
S. Em ±	0.11	0.32	0.32
C.D at 5%	0.31	0.94	0.92
<i>Interaction</i>			
S. Em ±	0.24	0.72	0.71
C.D at 5%	0.70	NS	NS

in accumulation, production and partitioning of photosynthates which resulted higher dry matter content and green forage and led to increased dry matter yield. This is in conformity with the findings of Midha *et al.* (2015), Shekara *et al.* (2020), Singh *et al.* (2021) and Shekara *et al.* (2022).

### Fodder Quality

The genotypes differed significantly with crude protein content, crude protein yield, total digestible crude protein yield and crude fibre yield (Table 2 & 3). Among Dinanath grass genotypes crude protein content was higher with check variety Bundel Dinanath-2 (7.2%). Whereas, the crude protein, total digestible crude protein and crude fibre yield were significantly higher with genotype JHD-19-4 (4.5 q ha<sup>-1</sup>, 3.7 q ha<sup>-1</sup> and 19.0 q ha<sup>-1</sup> respectively). Application of nitrogen at 90 kg ha<sup>-1</sup> significantly recorded higher crude protein content 7.2 (%), crude protein yield (5.6 q ha<sup>-1</sup>), total

digestible crude protein yield (4.8 q ha<sup>-1</sup>) and crude fibre yield (19.1 q ha<sup>-1</sup>). The higher crude fibre content was observed with nitrogen at 30 kg ha<sup>-1</sup> (29.1 %). The interaction between genotypes and nitrogen levels were found significant only with crude protein content and rest of the quality parameters found non-significant. The higher crude protein and total digestible yield was attributed due to the higher crude protein content and dry matter yield with higher level of nitrogen. The results are similar with the findings of Tyagi and Singh (1986), Tripathi and Singh (1991), Rathore and Kumar (1978), Rathore and Kumar (1997b), Asmare *et al.* (2017), Tilahun *et al.* (2017), Mustapha *et al.* (2018), Suleiman *et al.* (2020).

### Nitrogen Use Efficiency

Nitrogen use efficiency of genotypes was significantly influenced by nitrogen levels (Table 3). Among genotypes, JHD-19-4 recorded significantly higher

TABLE 3  
Nutritive value and nitrogen use efficiency of Dinanath grass genotypes as influenced by nitrogen levels at harvest

Genotypes	Crude Protein Yield (q/ha)	Crude fibre Yield (q/ha)	Total Digestible crude protein yield (q/ha)	Nitrogen use efficiency (Kg Green fodder per Kg Nitrogen)
JHD-19-4	4.5	19.0	3.7	572.8
BAU-DN-110-18-2	3.6	13.0	2.8	411.0
BAU-DN-109-8	3.8	14.3	3.0	426.1
BAU-DN-103-18-2	3.4	15.9	2.6	493.4
Bundel Dinanath-2	4.4	15.1	3.6	476.3
S. Em $\pm$	0.18	0.75	0.18	16.0
C.D at 5%	0.53	2.18	0.52	46.6
<i>Nitrogen Levels (Kg/ha)</i>				
30	2.3	11.5	1.6	666.8
60	3.9	15.8	3.1	431.3
90	5.6	19.1	4.8	329.7
S. Em $\pm$	0.14	0.58	0.14	12.4
C.D at 5%	0.41	1.69	0.40	36.1
<i>Interaction</i>				
S. Em $\pm$	0.32	1.30	0.31	27.7
C.D at 5%	NS	NS	NS	NS

TABLE 4  
Economics of Dinanath grass genotypes as influenced by nitrogen levels recorded at harvest

Genotypes	Total Cost of Cultivation (Rs./ha)	Gross Returns (Rs./ha)	Net returns (Rs./ha)	B:C Ratio
JHD-19-4	23731	61083	37352	2.56
BAU-DN-110-18-2	23637	43588	19951	1.83
BAU-DN-109-8	23686	44886	21199	1.89
BAU-DN-103-18-2	23663	50819	27155	2.14
Bundel Dinanath-2 NC	23656	51515	27858	2.17
S. Em ±	34	1607	1602	0.07
C.D at 5%	NS	4679	4664	0.19
Nitrogen Levels (Kg/ha)				
30	22545	40017	17472	1.77
60	24011	51769	27758	2.16
90	24468	59347	34879	2.43
S. Em ±	27	1245	1241	0.05
C.D at 5%	78	3624	3613	0.15
Interaction				
S. Em ±	59	2783	2774	0.12
C.D at 5%	NS	NS	NS	NS

nitrogen use efficiency (572.8 kg green fodder per kg of nitrogen). Application of lower nitrogen levels of 30 kg ha<sup>-1</sup> recorded significantly higher nitrogen use efficiency (666.8 kg green fodder per kg of nitrogen). Whereas, higher level of nitrogen (90 Kg ha<sup>-1</sup>) recorded lower nitrogen use efficiency (329.7 kg green fodder per kg of nitrogen). The nitrogen use efficiency was higher at lower level of nitrogen and decreased with incremental nitrogen levels. This might be due to higher nitrogen levels which might have led to lower utilization of applied nitrogen within short period of growth and also subjected to various forms of nitrogen losses. This is in harmony with the findings of Shekara *et al.* (2008), Devi *et al.* (2014) Joshi *et al.* (2015) and Shekara *et al.* (2022).

### Economic Analysis

Among genotypes JHD-19-4 registered higher net monetary returns (Rs.37,352 ha<sup>-1</sup>) and benefit cost ratio (2.56). Application of nitrogen at 90 Kg ha<sup>-1</sup> recorded higher net monetary returns (Rs.34,879

ha<sup>-1</sup>) and benefit cost ratio of 2.43 (Table 4). This is due to better growth attributers which resulted higher green forage yield with a marginal increased cost of nitrogen at higher nitrogen levels. Similar results were reported by Sharma and Bhunia (2001) and Bama *et al.* (2013) and Shekara *et al.* (2022).

Based on the results it can be inferred that Dinanath grass variety JHD-19-4 with nitrogen level of 90 kg ha<sup>-1</sup> found suitable and economical, which recorded higher green forage, dry matter, crude protein, total digestible crude protein and crude fibre yield. The same variety recorded higher net monetary returns with nitrogen level of 90 kg ha<sup>-1</sup> in southern dry zone of Karnataka under protective irrigated situation.

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## Influence of Irradiation and Packaging on the Shelf Life of White Finger Millet (Ragi) [*Eleusine coracana* (L.)] Flour

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### ABSTRACT

The irradiation of food products as a measure of disinfestations against insects and microorganisms to extend its shelf life is a well established procedure worldwide. However, irradiation alone will not yield good results until and unless packed in suitable packaging material. Hence, the study was planned to irradiate the white finger millet flour at 1.5 kGy (IR) and stored in different packaging materials (LDPE, PP, PET and MPP) for a period of three months by taking flour stored without irradiation in steel box as control. Set of flour packed in different packaging material (LDPE, PP, PET and MPP) without irradiation served as non-irradiated sample (NIR). Every fortnight the samples in three different treatments were drawn for biochemical changes (moisture, alcoholic acidity and FFA), insect infestation and microbial growth. Significantly increase in moisture, alcoholic acidity and FFA content were noticed in control sample followed by non irradiated PP and LDPE packed flour. Insect infestation started after 30 and 60 days of storage in control and NIR-PP and NIR- LDPE covers respectively. Further, study indicated that acceptable level of biochemical changes, no fungal and insect growth was noticed in IR- MPP and IR-PET packed samples. Thus, white finger millet flour stored in MPP and PET pouches with irradiation dose of 1.5 kGy can be safely stored up to three months under room temperature.

**Keywords :** White finger millet, Irradiation, Insect infestation, Free fatty acids

MILLETS, considered as important food staples in human history. They have been in cultivation in East Asia for the last 10,000 years. Africa is the largest producer of millet (20.6 million metric tonnes), followed by Asia (12.4 million metric tonnes) and India (10.5 million metric tonnes). Millets including Pearl millet, Finger millet, Kodo millet, Proso millet, Foxtail millet, Little millet and Barnyard millet are important staples to millions of people world-wide.

Generally, these are rainfed crops grown in areas with low rainfall and thus resume greater importance for sustained agriculture and food security. Almost all the millets are used for human consumption in most of the developing countries but their use has been primarily restricted to animal feed in developed countries. Millets are nutritionally comparable to

major cereals and serve as good source of protein, micronutrients and phytochemicals. Among the millets, finger millet (ragi) is a staple food in many African and South Asian countries. It is also considered as a helpful famine crop, as it is easily stored for lean years (FAO, 2012). The grain is readily digestible, highly nutritious and versatile and can be cooked like rice, ground to make porridge or flour used to make cakes (De Wet, 2006). Finger millet or ragi remains one of the main ingredients of the staple diet in Karnataka. Finger millet (Ragi) is an extremely nutritious cereal and is very beneficial for maintaining good health. Sprouted grains are recommended for infants and elderly people. In Karnataka, finger millet flour, popularly called as 'ragi hittu' could be enjoyed in different forms and preparations such as *ragi roti*,

*ragi dosa*, *ragi porridge*, *ragi upma*, *ragi cakes*, *ragi biscuits*, *ragi malt*, *ragi vermicelli* and *ragi papad* are few popular dishes of *ragi*.

Nutritionally, finger millet is a good source of nutrients especially calcium, phosphorus and fibre. Total carbohydrate content of finger millet has been reported to be in the range of 70 to 79.5 per cent depending upon the type of variety. The carbohydrates include starch as the main constituent being 59.4 to 70.2 per cent (Bhatt *et al.*, 2003). Traditionally brown coloured grains are predominant and preferred by the consumers. Of late, white grains are preferred by the food processing industries because of their high protein, low tannins and increased consumer acceptability (Sharathbabu *et al.*, 2008). White coloured finger millet or *ragi* is mainly used in the form of flour in the preparation of bakery products such as bread, biscuit, sev, muruku vermicelli and many more. The white varieties have higher protein content than the brown varieties of the finger millet. Finger millet contains 44.7 per cent essential amino acids of the total amino acids which is higher than the 33.9 per cent essential amino acids. Since *ragi* does not contain gluten, it is a wonderful grain alternative for people who are gluten-sensitive (Dayakar *et al.*, 2017).

Due to increasing awareness of consumers regarding advantages of consumption of finger millet based staple foods, convenience foods, health mixes, infant foods, the production, availability and access plays a key role towards increasing consumption. The major drawback of finger millet consumption can be attributed to its dark brown or dark red colour has led to decrease its acceptability among children and urbanites. Even the availability of preferred cereals such as rice and wheat at subsidized prices also contributes for lesser usage. However, in the recent past with the support of governments, the cultivated area under this millet is increasing constantly, mean while, finger millet consumption also increased among health conscious consumers of urban areas. In the recent past, with the constant effort of plant breeders, the white coloured finger millet varieties with the same or superior nutritional quality are

available, which serve as a boon to bakery and confectionery industries as well as appearance loving people. Modern home makers finds it difficult to cook many of our traditional recipes due to non availability of white finger millet flour in ready-to-usable form like wheat flour or rice flour. Whole finger millet grain as such having excellent keeping quality, once it is milled into flour, it readily deteriorates due to rancidity and attack of insects and micro organisms. The deterioration in storage due to the infestation by red flour beetle (*Tribolium castaneum*) and other microorganisms lead to losses which in turn has adverse effects on the economy of the nation and health of the people. Once the grains are ground into flour, it will intensify the activity of secondary pests during storage. The infestation by *Tribolium castaneum* could directly result in weight loss (hallow grains) and the beetle indirectly imparts a brownish tinge and pungent smell to infested flour by secretion of benzequinones (Hodges *et al.*, 1996).

It is therefore necessary that such losses after milling or during storage can be reduced through the use of technology so as to provide adequate information that will guarantee food security and food safety to the population. Food irradiation is already recognized as a technically feasible method for reducing postharvest food losses, ensuring the hygienic quality of food and facilitating wider food trade (Jyoti *et al.*, 2009). A food is irradiated to utilize the destructive power of ionization radiation on the microorganisms with minimum changes in food constituents (Zenthen and Sorensen, 2003). The use of irradiation alone as a preservation technique will not solve the problems of post-harvest food losses which are severe but it will definitely play an important role in cutting post harvest losses in many cases. Extensive research work done at the Bhabha Atomic Research Center (BARC) Mumbai had shown that low dose gamma irradiation (0.2-0.3 KGy) is effective in controlling insect infestation in rawa or semolina (Rao *et al.*, 1994) and many other food products. As per the literature cited, wheat and soya flours are normally irradiated at the rate of 1.0 kGy and health mix containing *ragi* is irradiated at the rate of 0.5 kGy. However, FSSAI proposed standards for irradiation of foods under

class 3 (cereals, pulses and their milled products) provided the range is 0.25-1.0 kGy for insect disinfestations and 1.5 to 5.0 kGy for reduction of microbial load. Results of innumerable studies assure that the intake of irradiated food is absolutely safe for the consumers (Farkas, 2006). Regular brown colored finger millet flour or ragi flour in aesthetic designs is already available in the market in good number of packages; however, white finger millet flour in suitable packing material with good shelf life is very essential for bakery, confectionary people or regular consumers to meet their daily needs. Flour packed and stored in right conditions can prevent the loss or gain of moisture, entry of microorganisms, changes in fatty acid profile. Good packaging of any product will serve two purposes which are essentially technical and presentational. Technical aspects in packaging aim to extend the shelf life by providing better protection from all the hazards (physical, chemical and biological) during storage. The temperature variation in flour products could result in either hydrolytic or oxidative rancidity, triggering destabilization of flour quality. Hence, good and shelf stable package under sealed condition could prevent moisture absorption, free radical build up, prolong keeping quality and prevent microbial proliferations.

Hence, Shelf life of any flour is very important from the point of producer as well as the consumer. Studies on storage of millets in different conditions and different packaging materials are available a plenty (Chaturvedi *et al.*, 2013; Bunkar *et al.*, 2014; Thilagavathi *et al.*, 2015; Sindhu *et al.*, 2016 and Bhatt *et al.*, 2017), however, systematic studies on storage of white finger millet flour (white ragi flour) which is the basic raw material for the preparation of conventional as well as bakery products is available in very less numbers. Moreover, studies on combined effect of radiation as well as packaging on quality of white finger millet flour are not available. Hence, the study entitled 'Influence of Irradiation and Packaging on the shelf life of white finger millet flour' was taken up to assess the effect of different packaging and radiation treatment on the shelf life of white finger millet flour under room temperature.

## MATERIAL AND METHODS

The White finger millet (KMR-340) was procured from AICRP (Small millets), ZARS, V.C. Farm, Mandya and were cleaned and milled into flour using domestic flour mill and packed in different packaging material [Low density polyethylene (LDPE)-250gauge, Polypropylene (PP)-250 gauge, Metallised Polyester Polyethylene (MPP)-400 gauge and Polyethylene Terephthalate (PET)-420 gauge as per the experimental design. One set of white finger millet flour samples immediately after milling packed into above packaging material were sent to BARC (Bhabha Atomic Research Centre) Mumbai for irradiation. Another set of flour samples packaged in different packaging material (LDPE, PP, PET and MPP) were kept under refrigeration until the arrival of irradiated samples. The irradiation process was carried out by exposing the packed milled flour samples to gamma radiation at the rate of 1.5 kGy (IR). Another set of millet flours packed in different packaging material (LDPE, PP, PET and MPP) without irradiation treatment (NIR). The white finger millet flour samples stored in stainless steel boxes (normal house hold practice) was served as Control.

**Storage Study :** Control, irradiated (IR) and non radiation (NIR) samples stored in different packaging material under normal room temperature ( $30 \pm 2$  °C) with a relative humidity of  $75 \pm 5$  per cent for a period of three months. The stored millet flour samples were analyzed every fortnight for various flour quality parameters.

**Nutritional Composition :** Nutritional composition of the white finger millet flour (Moisture, protein, fat, ash, crude fiber, calcium, iron, magnesium, phosphorus and potassium) immediately after milling were analyzed according to standard AOAC (2005) procedure.

**Functional Parameters :** Water Absorption Index (WAI), Water Solubility Index (WSI), Water Absorption Capacity (WAC) and Oil Absorption Capacity (OAC) of white finger millet flour was analyzed immediately after milling as per Thilagavathi *et al.* (2015).

**Bio-Chemical Changes :** Bio-chemical changes of stored white finger millet flour such as moisture, alcoholic acidity and free fatty acids were analyzed every fortnight as per standard AOAC (2005) protocol.

The alcoholic acidity of stored white ragi flour was determined by using the formula given below (AOAC, 2005).

$$\text{Alcoholic acidity (as H}_2\text{SO}_4) = 24.25\text{AN} / \text{W}$$

Where,

N = Normality of standard sodium hydroxide solution

W = Weight of the material taken for test

A = Volume in ml of the std sodium hydroxide used in titration

**Insect Infestation :** Visual observation for dead or alive insects (including larvae and adults) was done using sieve method. Data on insect infestation was recorded on the total number of larvae and adults as insect population from each replication by taking 20 grams of flour into 90 cm diameter Petri dish and counting the same using magnifying glass and converting into per cent age (Mali and Satyavir, 2005).

**Microbial Analysis :** Microbial load of the stored flour including Total Bacterial Count (TBC), Fungal Count (FC) and Escherichia coli (E.coli) were assessed every fortnight as per Chaturvedi *et al.* (2013). For microbial analysis, nutrient agar (Bacteria), Potato Dextrose Agar (Fungi) and MacConkey-Sorbitol Agar (E.coli) were procured from Himedia and enumeration was done using serial dilution technique using appropriate dilutions.

**Statistical Analysis :** Data obtained in triplicates was statistically analyzed using three factor ANOVA to assess the significant difference (0.05 %) between the treatments, between the time intervals and between the packaging material on the shelf life of millet flour.

## RESULTS AND DISCUSSION

### Nutritional and Functional Quality of Milled White Finger Millet (Ragi) Flour

The nutritional and functional properties of white finger millet flour immediately after milling

TABLE 1

Nutritional composition of white finger millet flour (per 100 g)

Nutrients	White finger millet (KMR- 340)
Moisture (%)	10.20 ± 0.10
Ash (%)	2.69 ± 0.01
Fat (%)	4.20 ± 0.10
Protein (%)	8.90 ± 0.10
Crude fiber (%)	3.76 ± 0.10
Carbohydrate (%)	70.95 ± 0.55
Energy (K. Cal)	357.2 ± 10.56
Calcium (mg)	343.20 ± 0.80
Phosphorus (mg)	283.73 ± 0.64
Iron (mg)	3.80 ± 0.12
Functional quality of white finger millet flour :	
Bulk density (g/ ml)	1.62 ± 0.30
Water absorption capacity( ml/100 g)	76.78 ± 0.50
Oil absorption capacity (ml / 100 g)	70.68 ± 0.19
Water absorption Index (%)	4.23 ± 0.04
Water solubility Index (%)	7.34 ± 0.61

Values are mean of three replications ± SD

is depicted in Table 1. The white ragi variety (KMR-340) contained protein (8.90%), crude fiber (3.76%), calcium (343.20 mg%), phosphorus (283.73 mg%) and iron (3.80 mg%). The values reported in this work are in line with Gopalan *et al.* (2004) for most of the nutrients for brown finger millet except in protein and crude fiber. The functional properties such as bulk density (1.62 g/ ml), water and oil absorption capacity (76.78, 70.68.30 ml/ 100 g) reported for white finger millet flour in this work are in line with the values reported for selected millet and pulse flour by Thilagavathi *et al.* (2015) and Shobha *et al.* (2012) for maize flour.

### Effect of Storage on the Biochemical Parameters

**Free Fatty Acids (FFA) :** Free Fatty Acid (FFA) is a key feature linked with the quality and commercial value of fat and free fatty acids are indicators of deterioration of fat. Free Fatty Acids (FFA) are produced by the hydrolysis of oils and fats, since FFA's are less stable than neutral oil, they are more

prone to oxidation thus turning to rancid. The FFA in this study increased significantly from 0 to 90 days and the increase was more pronounced in control (steel box) as it was neither irradiated nor packed in specific packaging, followed by non irradiated LDPE and PP (Table 2), however the changes in FFA content at the end of storage period were significantly less in irradiated PET (0.79%) and MPP (0.85%) covers as compared to non-irradiated samples in different packaging types. The changes between the treatments, between the packaging as well as between the storage duration were found to be significant (Table 2). Significantly higher FFA content of white finger millet flour stored in steel boxes followed by non irradiated PP and LDPE covers is probably because of higher moisture absorption of these samples which leads to rapid hydrolytic action of lipases at higher moisture level leading to increased FFA content. Research work carried out by Panjin *et al.*, (2006) and Monika and Mridula (2015) on the effect of storage period on the dragee based sunflower kernel and nutritious bar respectively indicated a significant increase in free fatty acid content and which was within the acceptable

limit for three months of storage. Free fatty acid is an important parameter for storage of bajra flour and FFA was found significant after 16 days of storage in the cotton bag as compared to other packaging material such as Tin and HDPE container (Bhatt *et al.*, 2017). The FFA level should not exceed 1.5 per cent for noticeable rancidity (Shobha *et al.*, 2012). In this study the irradiated MPP and PET packed white finger millet flour have better retention of freshness compared to other packages. Similar result was also noticed in foxtail millet flour FFA content during strong (Shobha *et al.*, 2021)

*Alcoholic Acidity* : Flours when stored for long undergoes various types of deterioration, which in turn gives high value for alcoholic acidity, is an index of deterioration of flour during storage. Alcoholic acidity refers to combined acidity as we get by hydrolysis of fats by lipases into free fatty acids, hydrolysis of proteins into amino acids by proteolytic enzymes as well as acidity due to presence of certain acids, salts *etc.* The fresh white finger millet flour had alcoholic acidity of 0.08 per cent which

TABLE 2  
Effect of storage on Free fatty acid content of white finger millet flour

Storage days	Irradiated sample (IR)					Non irradiated (NIR)				
	B1	B2	B3	B4	B5	B1	B2	B3	B4	B5
0	0.21	0.21	0.21	0.21	0.21	0.21	0.21	0.21	0.21	0.21
15	0.49	0.43	0.15	0.27	0.34	0.36	0.23	0.27	0.43	0.40
30	0.52	0.81	0.17	0.23	0.38	0.48	0.32	0.36	0.81	0.42
45	0.80	1.03	0.24	0.35	0.42	0.61	0.33	0.53	1.03	0.46
60	0.95	1.12	0.33	0.52	0.61	0.71	0.46	0.55	1.12	0.67
75	1.24	1.18	0.48	0.64	1.13	1.23	0.72	0.72	1.18	1.23
90	1.35	0.85	0.79	0.82	1.32	1.33	0.87	1.13	1.36	1.38

Parameter	F-Value	SEm±	CD@5%
Free fatty acid (% oleic acid)	Between treatment	197.89	0.005
	Between packaging	646.45	0.007
	Between days	1670.4	0.009
	Treatment X packaging X days	1.927	0.027

Note : B<sub>1</sub>- Control, B<sub>2</sub>-MPP , B<sub>3</sub>- PET, B<sub>4</sub>- LDPE , B<sub>5</sub>-PP

increased significantly in control (Table 3) followed by PP and LDPE covers irrespective of irradiation. However, the white finger millet flour stored in irradiated MPP and PET covers showed significantly less changes in alcoholic acidity compared to other treatments.

**Moisture Content :** The moisture content plays a vital role in enhancing the shelf life of any product. Generally moisture content decreases or increases during storage depending upon the storage conditions and packaging material. The effect of storage on the moisture content of white finger millet flour is depicted in Table 4.

Significant increase in moisture content of white finger millet flour from initial value of 7.30 to > 15.00 per cent was recorded in Control followed by non irradiated PP covers. While in case of irradiated sample, moisture increase was significantly less in MPP (10.00%), PET (10.10%), LDPE (12.40%) and PP (12.50%). Among different treatments and packages, the irradiated MPP and PET packed flour exhibited significantly minimal changes in moisture

content as compared to others. The flour stored in steel box (control) absorbed highest moisture from the atmosphere, as it was obvious that during sampling, the lid of the box was widely exposed to the atmosphere leading to higher absorption of atmospheric moisture content. In case of PP and LDPE covers, the permeability for moisture transmission was quite high in these packages irrespective of irradiation treatment, however, the moisture content in all the packages was within the value prescribed by the Codex Alimentarius, where in the upper acceptable limit is 15.5 per cent for safe storage (Saad *et al.* 2014). Similar kind of work carried out by Bhatt, *et al.* (2017), indicated that the moisture content of bajra flour was found significant after 16 days of storage and the increasing trend was found in treatment of all the varieties of bajra flour kept in cotton bag at room temperature. Even the results of Veena *et al.* (2012) also reports higher moisture gain in papad samples during storage period of 90 days. The increase of moisture content in pearl millet grain over 12 months storage was reported by Mali and Satyavir (2005) where in initial

TABLE 3  
Effect of storage on Alcoholic acidity of white finger millet flour during storage

Storage days	Irradiated sample (IR)					Non irradiated (NIR)				
	B1	B2	B3	B4	B5	B1	B2	B3	B4	B5
0	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08	0.08
15	0.41	0.13	0.22	0.28	0.30	0.81	0.55	0.64	0.74	0.41
30	1.05	0.22	0.34	0.42	0.53	0.93	0.72	0.82	0.92	1.05
45	1.14	0.36	0.46	0.57	0.62	1.23	0.92	1.04	1.13	1.14
60	1.24	0.43	0.55	0.64	0.74	1.36	1.04	1.24	1.23	1.24
75	1.43	0.53	0.63	0.76	1.22	1.43	1.13	1.33	1.36	1.33
90	1.56	0.74	0.71	0.81	1.34	1.55	1.31	1.41	1.42	1.41

Parameter	F-Value	SEm±	CD@5%
Alcoholic acidity (% H <sub>2</sub> SO <sub>4</sub> )	Between treatment	4059.2	0.004
	Between packaging	697.3	0.006
	Between days	3721.3	0.007
	Treatment X packaging X days	14.21	0.021

Note : B<sub>1</sub>-control B<sub>2</sub>- PET B<sub>3</sub>- MPP B<sub>4</sub>-LDPE, B<sub>5</sub>-PP

TABLE 4  
Effect of storage on Moisture content of white finger millet flour during storage

Storage days	Irradiated sample (IR)					Non irradiated (NIR)				
	B1	B2	B3	B4	B5	B1	B2	B3	B4	B5
0	7.30	7.30	7.30	7.30	7.30	7.30	7.30	7.30	7.30	7.30
15	10.30	6.50	6.70	7.400	9.00	10.30	7.50	7.66	9.90	10.30
30	11.20	7.00	7.10	8.200	9.50	11.20	8.53	9.23	10.30	11.20
45	12.40	7.30	7.60	9.500	9.90	12.40	9.33	10.23	11.70	12.90
60	13.26	8.00	8.10	10.200	10.20	13.26	10.40	11.46	12.20	13.90
75	14.36	8.50	9.00	11.433	11.43	14.36	11.46	12.46	13.30	14.26
90	15.40	10.00	10.10	12.400	12.50	15.40	12.46	13.56	14.46	15.33

Parameter		F-Value	SEm±	CD@5%
Moisture (%)	Between treatment	2362.7	0.026	0.073
	Between packaging	1069.6	0.042	0.116
	Between days	2247.5	0.049	0.164
	Treatment X packaging X days	5.438	0.155	0.137

Note : B<sub>1</sub>- Control, B<sub>2</sub>-MPP, B<sub>3</sub>- PET, B<sub>4</sub>- LDPE, B<sub>5</sub>-PP

moisture content (5.15%) of the grain increased to 13.7 per cent after 12 months of storage at 25 °C.

### Effect of Storage on Insect Infestation

The effect of storage on the insect infestation of white finger millet flour is depicted in figure 1. Significantly more number of insects (larvae and adults) was noticed in Control sample. Insects appeared after 30<sup>th</sup> day of storage in control, while after 60 days in PP and LDPE packed flours irrespective of irradiation. Number of insects (larvae and adults) was increased significantly in steel boxes from 30 days to till the end of storage

period (> 80 numbers). There was no insect infestation in MPP and PET covers irrespective of irradiation treatment (Fig. 1). The increase in insect infestation in steel boxes as compared to other materials was due to retention of higher moisture inside steel boxes that resulted in faster multiplication of the insect. The present study revealed that the increase of insect population in LDPE and PP packaged flour implies that it is not only the moisture content of the outer environment but also the insect population that created more humidity in the air by their metabolic activity which further increased grain moisture and insect population, has also been reported by Mali and Satyavir (2000).

The type of insects noticed in this study were majorly red coloured Red flour beetle (*Tribolium castaneum*) followed by black coloured Rice weevil (*Sitophilus oryzae*) and few creamish larvae of Rice moth (*Corcyra cephalonica*). Bran of millet contains some essential nutrients and has been implicated in supporting higher population of *T. castaneum* in whole finger millet flour. Similarly, study conducted by Mali and Satyavir (2005) on the storage of pearl millet found that grains

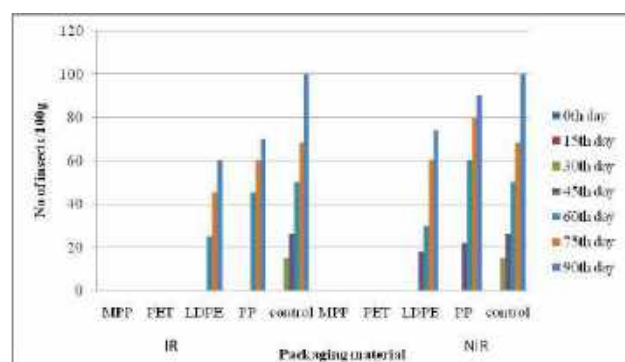


Fig. 1: Insect infestation in white finger millet flour



were majorly infested with the larvae and adults of lesser grain borer (*Rhizopertha dominica*) and red flour beetle (*Tribolium castaneum*). Since *T. castaneum* is a secondary pest which prefers fine flour than grits or semolina for its growth. In this study, as the finger millet is milled into flour has enhanced the development and survival of *T. castaneum* in some packages. The less or no insects in MPP and PET packages in this study was due to the fact that the tightly packed flour reduce the development of eggs laid by *T. castaneum* in these packages irrespective of irradiation treatment

### Effect of Storage on Microbial Quality

The microbial load of white finger millet flour stored in different packaging material is depicted in Fig. 2. More number of bacterial and fungal population was reported in non irradiated PP (14.01, 4.0 cfu/g) and control (10.04, 4.44 cfu/g) samples. The perusal of figure 2 indicated that there was no *E. coli* infestation in any of the samples, indicating that the method followed during flour making and storage was hygienically safe. In case of irradiated MPP and PET

covers less than five numbers of bacteria (1.0 and 1.5 log cfu/ g) were noticed and which did not increase throughout the storage period (Fig. 2).

No fungal colonies were noticed in irradiated MPP and PET packed flour. More number of bacterial and fungal counts in PP package irrespective of irradiation treatment (Fig. 2) might be due to damage caused to PP covers while handling, transportation and storage. Similar results were reported in irradiated processed ragi and barley by Chaturvedi, *et al.* (2013). Even the results of Ramasri *et al.* (2014) concluded that irradiation treatment (0.5kGy up to 3kGy) of health mix reduced the bacterial count and increased the shelf life. Our results are in line with the findings of Singh *et al.* (2006) and Mallesi *et al.* (1996) where in they found that there was no mould growth in irradiated formulation at the dosage of 0.5 kGy. A given radiation dose will kill a certain proportion of the microbial population exposed to it, regardless of the number of microorganisms present.

This property or result of radiation treatment implies that the higher the pretreatment population of bacteria, then higher will be the population after the food has been irradiated. If spoilage has already begun, radiation can do nothing to reverse it. Consequently, as with any other method of food preservation, irradiation is not a substitute for good hygienic practice in food production and processing. Exactly what portion of a given population of microorganisms will be destroyed by irradiation depends on several factors such as temperature at which the radiation treatment is carried out, time of irradiation and commodity which get irradiated.

In this study, the MPP and PET packages served as better packaging material for safe storage of white finger millet flour. Further, irradiation along with good packaging led to control of insect and bacterial population. Similar kind of study conducted by Panjin *et al.* (2006) reported that metalized polyester/polyethylene; labeled metalized PET/ PE containers were most suitable for storage of dragee product. The packaging materials such as metalized polyester/polyethylene; labeled metalized PET/ PE containers

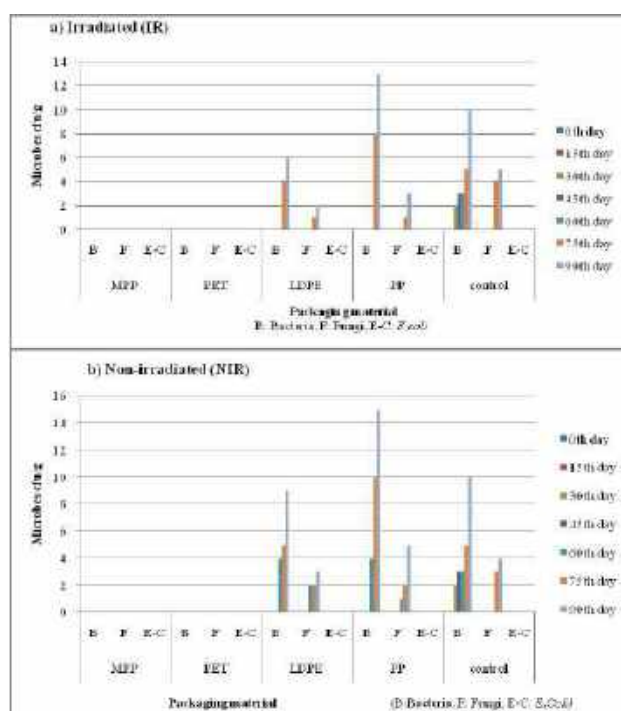


Fig. 2: Microbial quality of white finger millet flour a) Irradiated, b) Non-irradiated

had lowest oxygen permeability (8.0mLm<sup>-2</sup>/dan 'p1bar) which had strong influence in the prevention of hydrolytic and oxidative changes in the final product. Even results of Bhatt *et al.* (2017) demonstrates that Tin and HDPE container are suitable for storage of bajra flour under room temperature for a short period of 16 days but with irradiation the storage period can be extended significantly. Even the shelf life of foxtail millet flour was found to be superior with irradiation (1.5 kGy) when packed in MPP and PET packages (Shobha *et al.*, 2021)

Thus, the study demonstrates that the irradiation alone will not provide lasting disinfestations effect, therefore, it is also important to select the suitable packaging materials that cannot be penetrated by insects or beetles should be used to avoid post irradiation infestation. The use of irradiation alone as a preservation technique will not solve the problems of post-harvest food losses but definitely it will play an important role in cutting losses in many ways when used judiciously along with good packaging. Hence, the present study revealed that the white finger millet flour stored in MPP and PET pouches with irradiation dose of 1.5 kGy found suitable for safe storage up to three months under room temperature.

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## Farmland Values and Sales in Eastern Dry Zone of Karnataka

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### ABSTRACT

The present study aims to analyze the factors influencing farmland value and the reasons for farmland sales in the eastern dry zone of Karnataka. The sample consists of 80 farmers, including 40 respondents from Bagepalli taluk (BTHL) considered under rural area and 40 respondents from Chikkaballapur taluk (CTUP) considered under urban area for the study. It was observed that the process of transformation in the study area has resulted in a doubling of farmland values every five years and particularly in the time interval of 2005 to 2010 land prices have increased more than three times in both rural and urban areas. In the BTHL, the land value has increased from Rs.0.23 lakhs per acre in the year 2000 to Rs.12.57 lakhs in the year 2020. And in the CTUP, the average land value was Rs.2.05 lakhs per acre in the year 2000 and it has increased to Rs.63.91 lakhs in 2020. To clear old debts, perform marriages, construction of the house and higher land values were identified as major influencing factors of farmland sales. To capitalize on current high farmland prices augmented sales of farmlands is observed in both rural and urban areas over the years. It is resulting in the marginalization of farm holdings putting their livelihoods at risk in the long run. Although there was large scale selling-buying of land, not complete giving-up of the land was observed in general. The nature of sale was observed to be voluntary in the BTHL but in the CTUP 12.50 per cent of respondents reported forced sales. The increase in the prices of the farmland over the years may have potential threat to farmlands and the livelihoods of farmers. Hence proper policies should be evolved for the protection of agricultural lands in the study area so that the livelihood of a large number of farmers can be safeguarded in the long run.

*Keywords* : Land values, Farmland sales, Influencing factors, Marginalization of holdings, Livelihood of farmers, Eastern dry zone

URBANIZATION is a global phenomenon that comes with human settlements and accompanying anthropogenic activities and it plays an important role in land use and land cover change and urban sprawl are some of the most noticeable effects of urbanization on land use. Urban influencing factors are playing a critical role in affecting the overall farmland value and high real estate earnings have led to rising farmland prices. To capitalize on current high land prices and resulting capital gains farmers are selling the farmlands. The developments in the area have brought in transition in land use system, land values,

agricultural production systems, farm capital accumulation and diversification in sources of income for livelihood. A similar rapid surge in urban expansion can be observed across the country. For instance, as a result of urban expansion, land use/land cover has changed drastically at the periphery of Jalandhar city and it has led to the transformation of the rural landscape into the urban landscape where an inbuilt up area has increased to 37 per cent (2010) from 8 per cent (1975) at the cost of a reduction in farmland from 52 per cent to 31 per cent (Seema, 2014).

Chikkaballapur city is very close to Bengaluru North (45 km) considered an urban periphery. The developments like the establishment of an International airport, National highways, Hardware Park, Financial city project and other industries in the Bengaluru north have triggered the process of transformation of farmlands by surging prices and this has increased the marginalization of farm holdings in the places near to the city. Similar developments were observed in other countries too. For example, Larry and Burton (2012) reported that 37 per cent of respondents sold farmland to capitalize on current high land prices and resulting capital gains and reported that the farmland values had doubled in just five years and increased five folds during a period of 11 years in South Dakota, USA. These developments attracted the investment by real estate sector and the agricultural lands turned as common floors for construction of flats, villas, cargos and godowns, schools and colleges, hospitals, malls and supermarkets, resorts, hotels and restaurants, courier operators, parking yards, advertisement boards and so on. Xiaowei and Jay (2013) expressed that urban influencing factors were playing a critical role in affecting the overall farmland value and high real estate earnings had led to rising farmland prices in the California.

In the cases of rural areas, despite higher crop prices, Indian farmer's returns are declining because the cost of cultivation, especially wages, is rising faster. Mechanization is expensive and hence, small and marginal farmers who are unable to invest in technology are the worst hit. Pooja and Umesh (2021) opined that low income from agriculture, low employment level in the rural areas and outstanding debt of the households were the major reasons for migration from rural to urban areas. Farmers themselves understand these trends very well. This poor profitability coupled with non-price risks and family as well as social obligations push the farmers to farmland sales. Often when a farmer sells his own land to a speculator/investor, he uses part of the cash to buy land in areas where land is still cheap because of poor crop margins and on the demand side the investors, who want to invest in infrastructure,

factories, housing and even the sons of rural farmers who fled to the city are now willing to buy a few hectares in their villages as a good investment. The buying of land, rural land in particular, by wealthy households has been taking place in several parts of India since the early 2000s (Chakravorty, 2013 and Rajshekar, 2013). Fairbairn (2014) speaks of the role of high net worth individuals in buying up land but does not analyze the implications of such processes. Importantly, as we illustrate, in the context of growing income inequality, investment of savings in rural land by urban elites is an important mechanism through which rural dispossession takes place.

In the above context, the present study aims at analyzing the factors influencing farmland sales, escalation in the farmland values over the period, kind and nature of farmland sales in the study area and the irreversible transformation of farmlands has created a concern about the sustainability of agriculture. Vijayabaskar and Menon (2016) opined that small scale land sales have emerged as important means of dispossession of marginal and small farmers in a context of state neglect of agriculture, particularly irrigation infrastructure. Land markets have therefore worked to dispossess farmland as opposed to helping farmers consolidate viable landholdings. Kavitha *et al.* (2015) expressed their concern to protect and conserve the farmlands by proper policy and guidelines. Because, over the years, the expansion of Bengaluru to the fringes has declined the extent of agricultural land by 16.31 per cent. Similar concerns were expressed by Li Jiang *et al.* (2013), who alerted that the urban expansion is likely to continue and would result in a reduction in production in China due to reduced agricultural land use intensity. Santhakumar (2014) suggested while planning any development activity, the land value and its influencing factors have to be verified for the preparation of plans, project reports and policies to achieve a comprehensive solution.

## METHODOLOGY

Agriculture has seen transitions in terms of land use system, land values, water, labour and marketing

system in the eastern dry zone of Karnataka, because of developments in the area. Hence the study was conducted in the rural-urban continuum of Chikkaballapur district to analyze the influence of the urbanization process and other key factors on farmland values.

A multistage random sampling procedure was employed for the selection of the study area. At the first level Chikkaballapur district was selected and in the next level CTUP (Chikkaballapur Taluk Urban Periphery) and BTHL (Bagepalli Taluk Hinter Lands) were selected. In the next level, the list of farmers who have sold farmland in the year 2019 and 2020 were collected from the respective taluk Sub Registrar's office, Stamps & Registration Department. From the list purposeful sampling was done, selecting 40 respondents from Chikkaballapur taluk and 40 respondents from Bagepalli taluk thus forming a total sample size of 80 farmers. In Bagepalli taluk the data were collected from 22 villages belonging to 4 hoblies and in the Chikkaballapur taluk, the data were collected from 25 villages belonging to 3 hoblies.

The sample farmers were interviewed using a pre-tested schedule and data on socio-economic characters of the respondents, their land holdings, farmland values, reasons for sale and land sale details were collected. Analytical measures like descriptive statistics and percentage changes were used in analyzing the rise in farmland values, number and extent of land sales.

Per cent variation was calculated in reference to base year (beginning year)

Per cent variation = [(Current year value - Base year value) / Base year value] \* 100

### Compound Annual Growth Rate (CAGR)

To assess the growth rate in land values over the reference period the following growth rate formula was used

$$CARG = \frac{(V_{\text{final}})^{1/t} - 1}{V_{\text{begin}}}$$

Where,

CAGR = Compound annual growth rate

$V_{\text{begin}}$  = Beginning value

$V_{\text{final}}$  = Final value

t = Time in years

### t-test or Student's t-test

The t-test was used to assess whether the two data sets rural area and urban area are significantly different from each other. For this, null and alternative hypotheses were formulated. The null hypothesis was constructed as a hypothesis of no difference. The alternative hypothesis was constructed as having significant differences among land holdings in the rural and urban areas of the study region.

The t-test statistic was obtained as depicted below.

$$t = \frac{(\bar{X}_1 - \bar{X}_2)}{\left(\frac{\sigma}{\sqrt{n}}\right)}$$

Where,  $\bar{X}_1$  and  $\bar{X}_2$  were the sample mean from a sample of size n,  $\sigma$  is the standard deviation of the data.

The estimated t value was compared with the critical table value with the appropriate level of significance and degrees of freedom. If the estimated value was greater than the table value, it was inferred that there was a significant difference between the two groups in the study region.

### Rank Based Quotient (RBQ)

To analyze the reasons for the sale of farmland in the study area, a list of reasons for the sale of farmlands was developed during the preliminary survey conducted in the study area. The sample farmers were asked to rank the reasons at the time of interview using a pretested schedule. The quantification of data was done by first ranking the reasons based on the responses obtained from the respondents and then calculating the Rank Based Quotient (RBQ) (Sabarathnam, 1988), using the expression:

$$RBQ = \frac{\sum_{i=1}^n (F_i) (n + 1 - i)}{N * n} \times 100$$

Where,

$F_i$  = Number of farmers reporting a particular reason under  $i^{\text{th}}$  rank

$N$  = Number of respondents (Sample size – 40)

$n$  = Number of reasons identified.

## RESULTS AND DISCUSSION

The extent of land holdings before the land sale in the study area can be observed from Table 1. In case of BTHL, the majority of the respondents were medium farmers (40%) with an average land holding of 3.28 hectares, followed by small farmers with an average land holding size of 1.36 hectares. In the CTUP, majority of the respondents were small farmers (67.50 %) with an average land holding size of 1.49 hectares followed by medium farmers with an average land holding size of 3.52 hectares. The average farm size observed was higher in the BTHL (2.69 ha) compared to the CTUP (1.68 ha) and the result of the t-test infers that there is a significant difference in the size of land holdings between rural and urban areas. Ramalinge Gowda *et al.* (2012) reported similar results in Magadi taluk, Bengaluru district, where in the long-term, the rise in land prices was associated with reduced farm holding size. As the influence of urbanization decreases, the average holding size of farms increases and these changes were statistically significant at a one per cent level.

In any land sale, we observe two prices, one is the registered price indicating the fundamental value fixed by the state government and the other is the sale price or market price *i.e.* the actual price at which the land is transacted. The actual sale price is the true reflector of land values. These values were obtained from farmers through their memory recall by asking them the actual sale price of nearby similar lands which were transacted in that year and the results are presented in Table 2.

In the BTHL, the land value increased from Rs.0.23 lakhs per acre in the year 2000 to Rs.12.57 lakhs in the year 2020. The highest percentage increase was observed during the period 2005 to 2010. The land prices in this period had increased more than three times and it is attributed to the establishment of the international airport in 2008 in Devanahalli and also road developments like NH7. The people who sold lands in the Devanahalli region for the international airport were the part of buyers of agricultural land in the rural areas.

The land values in the CTUP have increased drastically. The average land value was Rs.2.05 lakhs per acre in the year 2000 and it has been increased to Rs.63.91 lakhs in 2020. The highest percentage increase was observed in the time interval of 2005 to 2010. The land prices in this period increased more than three times in the study area. The increase in the price was attributed to, a) Chikballapur district being

TABLE 1  
Classification of sample farmers based on the size of land holdings

Farmer Category	Rural (BTHL)		Urban (CTUP)	
	Sample size n=40	Average land size (ha)	Sample size n=40	Average land size (ha)
Marginal farmer (< 1 ha)	6 (15.00)	0.69	(12.50)	0.75
Small farmer (1-2 ha)	14 (35.00)	1.36	27 (67.50)	1.49
Medium farmer (2-5 ha)	16 (40.00)	3.28	7 (17.50)	3.52
Large farmer (> 5 ha)	4 (10.00)	6.47	1 (2.50)	5.05
Average farm size (ha)		2.69		1.68
t-stat				3.009 **

Note: \*\* Significant at 5 per cent level of significance ; Figures in parentheses represent percentages to total

TABLE 2  
Land values in different periods across Rural (BTHL) and Urban (CTUP) respondents

Year	Rural (BTHL)		Urban (CTUP)	
	Value (Rs.Lakhs/ac)	Percentage Increase	Value (Rs.Lakhs/ac)	Percentage Increase
2000	0.23		2.05	
2005	0.55	139.13	4.82	135.12
2010	2.60	372.72	20.05	315.97
2015	5.25	101.92	34.50	72.06
2020	12.57	139.42	63.91	85.24
Average land holding size(ac)		6.65		4.15
Average land value per farm (2 in lakhs) during 2019 and 2020		83.59		265.22
Average land value per ac (2 in lakhs) during 2019 and 2020		12.57		63.91
CAGR (%)		22.14		18.76

created out of Kolar district in the year 2007. It was carved out of moving Gauribidanur, Gudibande, Bagepalli, Chikkaballapur, Sidlaghatta and Chintamani taluks of the existing Kolar into the new district. b) Chikkaballapur being just 25 km away from Devanhalli where the international airport was established in the year 2008. c) Building the north-south six-lane national highway 7 (NH-7) as well as the east-west highway 69 passing through the district and also d) anticipated future developments in the area. Larry and Burton (2012) reported similar results

stating that the farmland values had doubled in just five years and increased five folds in 11 years in South Dakota, USA.

The average size of farmland sold, kind and nature of the sale is presented in Table 3. The average land sold was high in the BTHL (2.22 ac) compared to the CTUP (1.05 ac) in the study period of 2019 and 2020, as total land holding was higher in rural areas average land sold was also higher in a rural area compared to urban area. Ramalinge Gowda *et al.* (2012) reported

TABLE 3  
Average farmland sold in the years 2019 and 2020, kind and nature  
of sale across Rural (BTHL) and Urban (CTUP) areas

Particulars		Rural (BTHL)	Urban (CTUP)
Sample size		n=40	n=40
Average land sale (ac)		2.22	1.05
Type of sale	Complete sale (No.)	5 (12.50)	2 (5.00)
	Partial sale (No.)	35 (87.50)	38 (95.00)
Nature of sale	Voluntary sale (No.)	40 (100.00)	35 (87.50)
	Forced sale (No.)	0 (0.00)	5 (12.50)

Note : Figures in the parentheses are the percentage of sample size



similar results showing that the average size of land sold in areas with high urban influence areas (0.56 acres) was less than that of farms with low urban influence (6.5 acres). Although there was large scale of selling-buying, no complete giving-up of the land was observed. In the BTHL, only 12.5 per cent of the respondents sold the farmland completely and the remaining were partial sales. In the case of the CTUP, only five per cent of the respondents were identified as complete sellers and the remaining were partial sellers. The nature of sale observed was 100 per cent voluntary in the BTHL and in the CTUP 12.50 per cent of sample respondents indicated that their transaction was under forced sales.

Development of the city could not only be the prime force behind the sale of farmland. There could be other reasons as well which are external and internal to the farmer. This has been presented in Table 4 and 5. In the case of BTHL, the first three major reasons for farmland sales identified were a) To meet financial obligations *i.e.*, mainly to clear old debts (RBQ value 91.65); b) To perform marriages and other ceremonies (RBQ value 88.72) and c) to construct the house (RBQ value 81.29). To purchase agricultural land in a remote area and forced sales were the least influencing factors of farmland sales identified.

In the CTUP, the first three major reasons for farmland sales identified were a) construction of the house (RBQ value 89.98), b) attractively higher land value (RBQ value 85.39) and c) performing marriage and other ceremonies (RBQ value 77.69). Remoteness of the land parcel and forced sales were the least influencing factors of farmland sales identified.

Supply of farmland being inelastic in nature, with the increase in demand the prices have been increased over the years. Both BTHL, as well as CTUP show the tendency of increase in number as well as extent of sales because of attractive rise in prices. Harish and Chinnappa (2017) reported similar results showing that farmers were provoked to sell their farmlands due to high farmland prices in high urban influence areas leading to the marginalization of agricultural holdings which will put their livelihood

TABLE 4  
Reasons for sale of farmland in Rural (BTHL) areas

Particulars	1	2	3	4	5	6	7	8	9	10	11	12	RBQ Value	Rank
To meet the financial obligations	17	11	7	5									91.65	I
Marriage and other ceremonies	7	20	8	3	1	1							88.72	II
Lack of irrigation		1	4	18	16	1							72.51	IV
Construction of house	8	2	11	10	8	1							81.29	III
Higher land value	1		1	2	6	25	4	1					60.82	VI
Educational purpose	3	6	4	1	7	6	6	6	1				66.44	V
To purchase assets like gold, vehicle						5	15	10	1		8	1	40.81	VII
Labour scarcity to take up agriculture				1			6	7	13	11	2		35.19	IX
To purchase site or house	4		5		2		8	3		4	7	7	43.72	VII
To purchase agriculture land in remote area							1	5	7	6	13	8	22.05	XI
Remoteness of land parcel						1		8	13	2	10	6	27.27	X
Forced sale									5	17	18	18	18.53	XII

TABLE 5  
Reasons for sale of farmland in Urban (CTUP) areas

Particulars	1	2	3	4	5	6	7	8	9	10	11	12	RBQ Value	Rank
To meet the financial obligations	5		4	11	15		5						72.70	IV
Marriage and other ceremonies	3	5	10	11	6	5							77.69	III
Lack of irrigation					1	14	13	7	5				49.77	VII
Construction of house	12	16	5	6	1								89.98	I
Higher land value	14	8	8	9		1							85.39	II
Educational purpose	2	3		3		11	4	13	4				55.40	VI
To purchase assets like gold, vehicle					12	2	4	4	5	7			53.10	VIII
Labour scarcity to take up agriculture							4	5	20	5		6	31.23	X
To purchase site or house	4	8	7		5		1	4	2	9			63.93	V
To purchase agriculture land in remote area						5	9	4	4	4	8	6	36.86	IX
Remoteness of land parcel						2		3		3	17	15	18.10	XI
Forced sale										12	15	13	16.45	XII

at high risk in the future. In the near future it may create problems of marginalization of farmlands and conversion of farmland for non-agricultural uses. Construction of house, clearing old debts and to perform marriages and other ceremonies were identified as major influencing factors of farmland sales. Investing farmland sales proceeds in these kinds of activities does not create any future livelihood options for the farmers. In the CTUP, the transacted land is mainly used for non-agriculture uses mainly real estate, conversion into sites, shops, villa plots, factories and a few plots kept vacant and fenced with speculative intention. The urban elites who purchased farmland in the rural areas lease-out to the same farmers and few farmers work on a daily labour basis on their own farms after the dispossession of farmland. Hence proper policies should be evolved for the protection of agricultural lands in the study area and proper awareness should be created among the farmers regarding the problems which will encounter after the land sales so that the livelihood of a large number of farmers can be safeguarded in the long run.

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## Utilization of Bamboo Rice for Product Development

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### ABSTRACT

India is one of the largest bamboo producing country among other tropical countries like China, Egypt, United States, Malaysia and Sri Lanka. Bamboo rice is special rice that is grown out of a dying bamboo shoot and it has become an important and major source of income for tribal living in the forest. The aim of the study was to develop bamboo rice shavige, (shavige is very long, thin strands of noodles made from rice flour and water) evaluation of nutritional composition and sensory acceptability of the product. Nutrient composition of raw flour showed moisture, protein and crude fiber were higher in soaked and dried bamboo rice flour. Whereas, in fat and ash there was no much difference was observed. Carbohydrate and energy were high in white rice flour. Treatment 3 (T3) *i.e.*, 50 per cent white rice flour and 50 per cent soaked and dried bamboo rice flour had the highest score for all the sensory attributes. Whereas nutrient analysis of shavige mix revealed that white rice shavige mix had 10.56g moisture, 4.89g protein, 0.34g fat, 0.20g fiber, 1.24g ash, 81.55g carbohydrate and 355.03Kcal energy whereas, bamboo rice shavige mix had 8.91g moisture, 8.77g protein, 0.90g fat, 0.20g crude fiber, 0.53g ash and 80.89g carbohydrate.

**Keywords :** Bamboo rice, Sensory attributes, Nutrient analysis, Shavige mix , Product development

OVER 90 per cent of the world's rice is produced and consumed in Asia and Pacific regions. Over 2 billion people in Asia derive their energy from rice (Chanu and Shivaleela, 2019). Rice is consumed as polished white rice with the husk, bran and germ fractions removed. However, consumption of brown rice (hulled rice) is increasing in recent years, due to the increased awareness about its health benefits and good nutritional properties due to higher amounts of proteins, ash, dietary fibre and minerals than white rice (Muttagi *et al.*, 2017). India is one of the largest bamboo producing country among other tropical countries like China, Egypt, United States, Malaysia and Sri Lanka. There are over 1,250 woody bamboos in the world in approximately 75 genera. They are native to Africa, the Americas, Asia and Oceania and have been introduced to Europe (Liese and Kohl 2015). Bamboo rice has become an important and

major source of income for tribal living in the forest (Siyanna, 2020). Bamboo rice is special rice that is grown out of a dying bamboo shoot. When the bamboo shoot breathes its last, it flowers into a rare variety of rice seeds, which are known as bamboo rice (Rana, 2017). In bamboo, the fruit is one seeded structure that does not split when ripe (Wong, 2004).

Bamboo seeds are not only used as food by indigenous residents, but also traded as medicines and commodities. However, there is a lack of information about the nutrient profile of bamboo seeds, in contrast to the abundant literature available with nutritional information on cereal crops such as rice, wheat, maize, and so on (Kiruba *et al.*, 2007).

Bamboo rice is also known as Mulayri in Malayalam language and Moongil Arisi in Tamil language by the tribal of southern India. This rice is rich in

carbohydrates, proteins, amino acids, fiber, vitamins and minerals (Singh, 2021). Protein content of bamboo seed is higher than that of rice and wheat. Other than protein the rice also has vitamins including A, B1, B2, B3, B6 and minerals like calcium, iron, phosphorus, and magnesium (Bharathi, 2019).

Bamboo seed is an underutilized species in India, especially bamboo rice or seed species offer enormous potential for contributing to the achievement of the Millennium Development Goal (MDGs), particularly in combating hidden hunger and offering medicinal and income generation options. They are also closely tied to cultural traditions and therefore have an important role in supporting social diversity (Manohari *et al.* 2016).

Rice noodles are the most consumed form of rice product next to cooked rice grain in Asia. Noodles may either be served by frying and mixing with vegetables and meats or served as a soup noodle by boiling in a broth (Ahmed *et al.*, 2016).

So, the study was taken up with the objective of development and sensory evaluation of the shavige prepared by incorporation of bamboo rice flour.

## MATERIAL AND METHODS

### Procurement of Samples

Bamboo rice were procured from local organic market and white rice, jaggery were purchased from the local shop. Bamboo rice was refrigerated until further use.

### Processing of Bamboo Rice

Bamboo rice was cleaned, washed in running water 2-3 times, soaked overnight and dried under shade. Then it was powdered and stored in air tight container for further use. White rice was cleaned, powdered and stored in air tight container for further use.

### Analysis of Proximate Composition of White Rice and Bamboo Rice Flour

Nutrient analysis was carried out for dried white rice and bamboo rice flour in dehydrated form using

Association of Official Agricultural Chemists (AOAC, 2005). Moisture content was determined by drying the sample using oven in triplicates, protein by per cent total nitrogen by the kjeldhal procedure, fat was estimated as crude ether extract using moisture free samples and crude fiber of the sample was estimated by using moisture and fat free samples. Total ash content of sample was obtained by dry ashing the samples completely by heating it oven a flame. Carbohydrate (CHO) calculated by difference method and energy by calculation method.

TABLE I  
Composition of the product mix

Variation	White rice flour (%)	Bamboo rice flour (%)	Jaggery (%)
T1	100	00	40
T2	75	25	40
T3	50	50	40
T4	25	75	40

T1- 100 % White rice flour; T2- 25% bamboo rice flour: 75% white rice flour; T3- 50% bamboo rice flour: 50% white rice flour; T4- 75% bamboo rice flour: 25% white rice flour

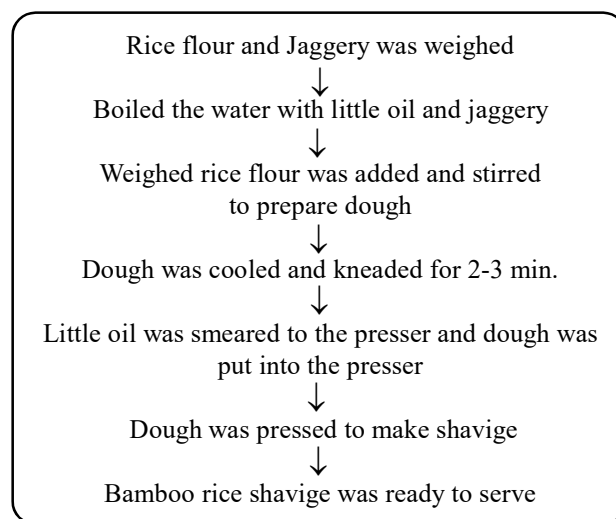


Fig. 1: Flow chart for shavige preparation

### Development of Bamboo Rice Shavige

Shavige was standardized by using white rice flour along with bamboo rice flour at 75: 25, 50: 50 and 25: 75 per cent, respectively and shavige prepared

### Flow Chart for Shavige Mix Preparation

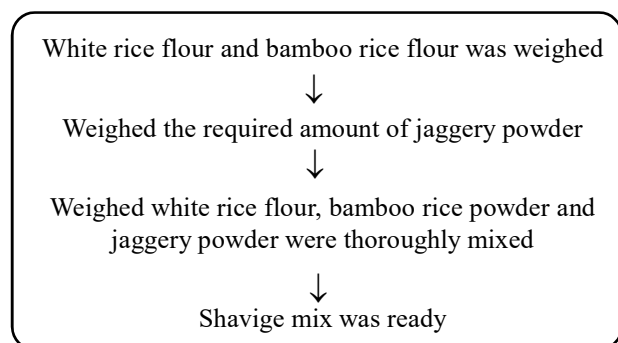


Fig. 2: Flow chart for shavige mix preparation

by using white rice flour considered as a control (Table 1). The method of preparation of shavige is depicted in Fig.1.

So shavige mix was prepared using best accepted *i.e.*, T3 variation by mixing 50 per cent white rice flour and 50 per cent bamboo rice flour with jaggery and method of preparation of shavige mix is depicted in Fig. 2. Further, Nutritional analysis was done.

### Sensory Evaluation of Developed Product

In order to select best acceptable level of bamboo rice powder incorporation. Sensory evaluation was carried out by a panel of 21 semi trained panel member using 9 point hedonic scale (Ranganna, 1986) for appearance, color, texture, flavor, taste and overall acceptability.

### Nutrient Analysis of Developed Product

Developed bamboo rice shavige mix was analyzed for moisture, protein, fat, fiber, ash using Association of Official Agricultural Chemists (AOAC, 2005) method, Carbohydrate was calculated by difference method and energy content was determined by calculation.

### Statistical Analysis

The data reported in the tables are the averages of triplicate observations. The data was analyzed statistically for the mean, standard deviation and ANOVA to test the significance among different levels of bamboo rice flour incorporation at 5 per cent significant level.

## RESULTS AND DISCUSSION

### Nutrient Analysis of White Rice and Soaked & Dried Bamboo Rice Flour

Moisture, protein, fat, fiber, ash, carbohydrate and energy were analyzed for both white rice flour and soaked & dried bamboo rice flour. Results are depicted in Table 2.

The findings indicated that mean moisture, protein, fat, fiber and ash content were significantly higher in soaked & dried bamboo rice flour (11.78g, 7.78g, 0.45g, 5.78g and 0.73g respectively) compared to white rice flour (8.80g, 4.84g, 0.43g, 1.02g and 0.39g

TABLE 2  
Nutrient composition of white rice flour and soaked & dried Bamboo rice flour

Parameters	White rice flour (Mean ± SD)	Soaked Bamboo rice flour (Mean ± SD)	't' test
Moisture (g)	8.80 ± 0.213	11.78 ± 0.252	*
Protein (g)	4.84 ± 0.064	7.78 ± 0.072	*
Fat (g)	0.43 ± 0.001	0.45 ± 0.003	*
Crude Fiber (g)	1.02 ± 0.015	5.78 ± 0.168	*
Ash (g)	0.39 ± 0.005	0.73 ± 0.086	*
Carbohydrate (g)	64.16 ± 0.351	57.83 ± 0.552	*
Energy (Kcal)	279.92 ± 1.167	266.27 ± 1.225	*

\*: Significant at 5% level

TABLE 3  
Sensory evaluation of the prepared product

Treatment	Characteristics (Mean $\pm$ SD)				
	Appearance	Color	Taste	Texture	Overall acceptability
T1	7.45 <sup>a</sup> $\pm$ 0.83	7.40 <sup>a</sup> $\pm$ 1.10	7.65 <sup>a</sup> $\pm$ 1.14	7.10 <sup>a</sup> $\pm$ 0.85	7.40 <sup>a</sup> $\pm$ 0.87
T2	7.45 <sup>a</sup> $\pm$ 0.76	7.10 <sup>a</sup> $\pm$ 0.79	7.10 <sup>a</sup> $\pm$ 0.97	7.15 <sup>a</sup> $\pm$ 0.59	7.20 <sup>a</sup> $\pm$ 0.51
T3	7.50 <sup>a</sup> $\pm$ 0.51	7.40 <sup>a</sup> $\pm$ 0.50	7.70 <sup>a</sup> $\pm$ 0.80	7.20 <sup>a</sup> $\pm$ 0.62	7.45 <sup>a</sup> $\pm$ 0.39
T4	7.00 <sup>a</sup> $\pm$ 0.86	6.85 <sup>a</sup> $\pm$ 0.81	6.80 <sup>b</sup> $\pm$ 0.83	6.80 <sup>a</sup> $\pm$ 0.83	6.86 <sup>a</sup> $\pm$ 0.71
F test	1.473 <sup>NS</sup>	1.521 <sup>NS</sup>	2.829*	0.888 <sup>NS</sup>	2.507 <sup>NS</sup>
SEm $\pm$	0.195	0.215	0.247	0.190	0.168
CD at 5%	-	-	0.685	-	-

\*: Significant at 5% level

NS: Non-significant

Common letter indicates Non- significant

T1- control; T2- 25% bamboo rice flour: 75% white rice flour ; T3- 50% bamboo rice flour: 50% white rice flour ;  
T4- 75% bamboo rice flour: 25% white rice flour

respectively). The computed carbohydrate and energy content of white rice flour was 64.16g/100g and 279.92 Kcal/100g which is significantly higher than soaked and dried bamboo rice flour (57.83g/100g and 266.27 Kcal/100g).

Verma and Srivastav (2017) investigated the proximate composition of aromatic and non-aromatic Indian rice who reported that 100gm of aromatic rice variety Badshah Bhog contains moisture (8.90g), fat (0.61g), protein (7.23g), ash (0.59g), fiber (0.85g), carbohydrate (82.70g) and energy (365.23 Kcal/100g). Hundred grams Non aromatic rice variety contains moisture (11.25g), fat (0.06g), protein (6.87), ash (0.35), fiber (0.64), carbohydrate and energy (353.89 Kcal/100g).

The results of the study is in line with findings of Ahmed *et al.* (2016) reported that cultivars of rice contained quality attributes such as protein, fat and ash range from 6.92 to 8.65, 0.63 to 2.17 and 0.55 to 0.77 per cent, respectively.

### Sensory Evaluation of the Developed Product

Bamboo rice shavige was standardized by incorporating soaked and dried bamboo rice flour with white rice flour at 25 per cent (T2), 50 per cent (T3) and 75 per cent (T4) and control shavige (T1) was

prepared from 100 per cent white rice. The mean sensory score of bamboo rice shavige is presented in table 3.

Bamboo rice shavige score for appearance ranged from 7.00 to 7.50, for color ranged from 6.85 to 7.40, for taste ranged from 6.80 to 7.70, for texture ranged from 6.80 to 7.20 and for overall acceptability ranged from 6.86 to 7.45. Control shavige had the highest scores for all the sensory parameters except texture. Among the variations highest score for appearance, color, taste, texture and overall acceptability (7.50, 7.40, 7.70, 7.20 and 7.45 respectively) were recorded for 50 per cent (T2) bamboo rice incorporated shavige and least score was for 75 per cent bamboo rice incorporated variation *i.e.*, T4 (75% bamboo rice and 25% white rice). The difference in all sensory characteristics *viz.*, appearance, color, texture and overall acceptability among the variations was found to be statistically non-significant at 5 per cent level, except taste which was found to be significant.

Thomas *et al.* (2014) studied the sensory acceptance rate of Bario and Basmati rice noodles. Bario rice had a higher acceptability score of 6.67 compared with Basmati rice (4.8). In term of appearance and overall acceptability, noodles made from Bario rice were ranked higher as compared to Basmati rice.

TABLE 4  
Nutrient composition of shavige mix

Parameter	White rice shavige mix (Mean ± SD)	Bamboo rice shavige mix (Mean ± SD)	't' test
Moisture (g)	10.56 ± 0.120	8.91 ± 0.347	*
Protein (g)	4.89 ± 0.055	8.77 ± 0.190	*
Fat (g)	0.34 ± 0.015	0.90 ± 0.045	*
Fiber (g)	0.20 ± 0.025	1.40 ± 0.041	*
Ash (g)	1.24 ± 0.072	0.53 ± 0.020	*
Carbohydrate (g)	81.55 ± 0.501	80.89 ± 0.231	NS
Energy (Kcal)	355.03 ± 0.724	366 ± 1.130	*

\*: Significant at 5 % level

NS: Non-significant

Ahmed *et al.* (2016) evaluated sensory characteristics for rice noodles. The overall acceptability of rice noodles depends upon appearance, aroma, taste, texture. Mean score for appearance ranged from 4.30 to 7.32, aroma ranged from 5.58 to 6.00, taste ranged from 5.14 to 6.12, texture ranged from 3.66 to 6.92 and overall acceptability ranged from 3.98 to 7.02.

#### Nutrient Analysis of Shavige Mix

Nutritional composition of white rice shavige mix and best accepted bamboo rice shavige mix were analyzed and depicted in Table 4. It was found that moisture, ash and carbohydrate were high in white rice shavige mix (10.56g, 1.24g and 81.55g) when compared with bamboo rice shavige mix (8.91g, 0.53g and 80.89g). Whereas, protein, fat, crude fiber, energy were high in bamboo rice shavige mix (8.77g, 0.90g, 1.40g and 366Kcal) when compared with white rice shavige mix (4.89g, 0.34g, 0.20g and 355.03 Kcal).

Poonsri *et al.* (2019) evaluated nutritional composition of rice noodles. Their findings shows that moisture (72.02%), ash (0.28%), protein (2.97%) and fiber (5.44%) was high in 40 g cassava leaves incorporated noodles and control noodles had moisture (69.50%), ash (0.13%), protein (2.28%) and fiber (0.77%) respectively. Fat (2.34%) and carbohydrate (24.98%) was high in control noodles whereas, 40 g cassava leaves incorporated noodles had 1.65 per cent fat and 19.80 per cent carbohydrate.

The results are on par with the study conducted by Zula *et al.* (2021) in moisture, protein, fat, ash, carbohydrate and energy.

Hence, the study indicated that the shavige mix prepared from soaked and dried bamboo rice flour incorporation at 50 per cent level was found to be the best accepted when compared with other variations including control by the panelists. The final product of shavige nutritional value was increased because of 50 percent incorporation of bamboo rice.

From the study, it can be concluded that moisture, protein and crude fiber were better in soaked and dried bamboo rice flour compared with white rice flour. Shavige prepared with incorporation of bamboo rice at 50 percent level was best accepted with having good amount of protein and energy compared with control. Proximate composition of the product mix revealed that protein and energy were better because of incorporation of bamboo rice flour. Acceptable value added products like shavige mix from bamboo rice can be developed and health benefits of bamboo rice can be exploited.

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## Effect of Pre and Post Emergence Herbicide Application on Growth and Yield of Direct Seeded Finger Millet (*Eleusine coracana* L.)

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### ABSTRACT

An experiment was conducted in Zonal Agricultural research station, V. C. Farm, Mandya during summer 2021 in direct sown finger millet (*Eleusine coracana* L.). The experiment consisted of ten treatments, which was laid with Randomized complete block design replicated thrice. The treatments included two pre-emergence herbicides (T<sub>1</sub>- Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> and T<sub>2</sub>- Bensulfuron methyl + Pretilachlor 6.6 per cent G @ 165 g a.i. ha<sup>-1</sup>), two post emergence herbicides (T<sub>3</sub>- Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i. ha<sup>-1</sup> and T<sub>4</sub>- 2, 4-D Na salt 80 WP @ 1000 g a.i. ha<sup>-1</sup>), four sequential application of pre followed by post emergence herbicides (T<sub>5</sub>- Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> followed by Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i. ha<sup>-1</sup>, T<sub>6</sub>- Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> followed by 2, 4-D Na salt 80 WP @ 1000 g a.i. ha<sup>-1</sup>, T<sub>7</sub>- Bensulfuron methyl + Pretilachlor 6.6 per cent G @ 165 g a.i. ha<sup>-1</sup> followed by Metsulfuron methyl + Chlorimuron ethyl 20WP @ 20 g a.i. ha<sup>-1</sup> and T<sub>8</sub>- Bensulfuron methyl + Pretilachlor 6.6 per cent G @ 165 g a.i. ha<sup>-1</sup> followed by 2, 4-D Na salt 80 WP @ 1000 g a.i. ha<sup>-1</sup>), weed free (T<sub>9</sub>- Two intercultivations at 20 and 40 DAS with one hand weeding at 30 DAS) and weedy check (T<sub>10</sub>- Unweeded control). The treatment with two intercultivations and one hand weeding treatment recorded significantly higher grain and straw yield of finger millet (3520 kg ha<sup>-1</sup> and 4825 kg ha<sup>-1</sup>, respectively). On par grain yield (3476 kg ha<sup>-1</sup>) and straw yield (4722 kg ha<sup>-1</sup>) were obtained by sequential application of Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> (3 DAS) as pre-emergence herbicide followed by post emergence application of Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i. ha<sup>-1</sup> (20 DAS) which was on par with that of Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> (3 DAS) followed by 2, 4-D Na salt 80 WP @ 1000 g a.i. ha<sup>-1</sup> (20 DAS) (3453 kg ha<sup>-1</sup> and 4582 kg ha<sup>-1</sup>). Higher net returns of Rs.48132 ha<sup>-1</sup> and B:C ratio (2.26) was obtained with sequential application of Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> (3 DAS) followed by 2, 4-D Na salt 80WP @ 1000 g a.i. ha<sup>-1</sup> (20 DAS). Based on the current study, it is clear that sequential application of Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> (3 DAS) as pre-emergence herbicide followed by post emergence application of 2, 4-D Na salt 80WP @ 1000 g a.i. ha<sup>-1</sup> (20 DAS) in direct sown finger millet in areas of labour scarcity is an effective weed management strategy to minimize the losses caused by weeds and to enhance the productivity of finger millet.

**Keywords :** Finger millet, Pre emergence, Post emergence herbicide, DAS - Days after sowing

**F**INGER millet, popularly known as ragi is one among the nutri-cereals which forms a staple food for majority of Asian and African population. In India, especially in Southern states it is a part of the daily

routine diet. In India, the area under the cultivation of this crop accounts to 10.04 lakh hectares with a production of 17.55 lakh tonnes and an average productivity of 1747 kg per hectare. Karnataka is a

major contributor of both with respect to area and production which accounts to 63.84 and 66.32 per cent, respectively.

The crop is predominantly cultivated under rainfed situation. Finger millet growers owing to the small and marginal holdings are resource starved and hence the cost and interest incurred in the production of this crop is least. The method of establishment usually practiced by farmers is usually broadcasting and sowing with seed drill. In some parts, where there is irrigation facility the crop is cultivated by adopting transplanting method of establishment. The advantage of transplanting technique is ensured under delayed and assured rainfall situations but crop area under irrigation is very meager.

Finger millet being a slow grower in the initial stages is susceptible for weed infestation. The yield losses as reported by different scientists are 34-61 per cent (Ramachandra Prasad *et al.*, 1991), 35-62 per cent (Manjunath & Muniyappa, 1992 and Lal & Yadav, 1982) under severe crop weed competition. Weeds cause an appreciable reduction in crop density, dry weight and depletion of nutrients from the soil (Pradhan *et al.*, 2010).

The most common methods deployed to control the weeds are by mechanical hand weeding and cultural methods. Although these methods are found to be promising they are more labour intensive and costly. Increasing labour scarcity and wages have led to untimely weeding resulting in the reduction in yield of the crop. There is dire need of exploring an alternative method of weed management which is cheaper and easy to practice. Hence, the farmers are in need of an effective weed management strategy which is easier and cost effective. In this context, the herbicides play an important role in reducing the yield losses due to weeds and also to enhance the productivity levels of the crop. In this background, the current investigation has been carried out to identify the effective herbicide for direct seeded finger millet.

## MATERIAL AND METHODS

A field experiment was conducted in Zonal Agricultural research station, V. C. Farm, Mandya during summer 2021. Geographically, the experimental site is positioned between 11°30' to 13°05' N latitude and 76°05' to 77°45' East longitude in the Cauvery command area of Karnataka at an altitude of 697 metres above the mean sea level. The initial soil samples were analysed by adopting standard procedures. The soil of the experimental site has a textural class of red sandy loam with neutral pH (7.2). The soil was low in available nitrogen (243.2 kg ha<sup>-1</sup>), medium in phosphorus (191.21 kg ha<sup>-1</sup>) and high in available potassium (224.28 kg ha<sup>-1</sup>). The experiment consisted of ten treatments, which was laid out in Randomized complete block design with three replications. The treatments included two pre-emergence herbicides (T<sub>1</sub>- Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> and T<sub>2</sub>- Bensulfuron methyl + Pretilachlor 6.6 per cent G @ 165 g a.i. ha<sup>-1</sup>), two post emergence herbicides (T<sub>3</sub>- Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i. ha<sup>-1</sup> and T<sub>4</sub>- 2, 4-D Na salt 80 WP @ 1000 g a.i. ha<sup>-1</sup>), four sequential application of pre followed by post emergence herbicides (T<sub>5</sub>- Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> followed by Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i. ha<sup>-1</sup>, T<sub>6</sub>- Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> followed by 2, 4-D Na salt 80 WP @ 1000 g a.i. ha<sup>-1</sup>, T<sub>7</sub>- Bensulfuron methyl + Pretilachlor 6.6 per cent G @ 165 g a.i. ha<sup>-1</sup> followed by Metsulfuron methyl + Chlorimuron ethyl 20WP @ 20 g a.i. ha<sup>-1</sup> and T<sub>8</sub>- Bensulfuron methyl + Pretilachlor 6.6 per cent G @ 165 g a.i. ha<sup>-1</sup> followed by 2,4-D Na salt 80 WP @ 1000 g a.i. ha<sup>-1</sup>), weed free (T<sub>9</sub>- two intercultivations at 20 and 40 DAS with one hand weeding at 30 DAS) and weedy check (T<sub>10</sub>- Unweeded control). Finger millet variety KMR 630 which has short duration released from AICRP on Small millets, ZARS, Mandya was used in the experiment with spacing of inter-rows of 30 cm at the seed rate of 12.5 kg ha<sup>-1</sup>. Fertilizers were applied as per the recommendation of 100:50:50 kg N: P<sub>2</sub>O<sub>5</sub>: K<sub>2</sub>O ha<sup>-1</sup>, with 50 per cent of nitrogen applied as basal dose and remaining 50 per cent as top dressing at 30 days after sowing.

The entire phosphorus and potassium were applied as basal dose at the time of sowing. The gross plot size was 5x4 m. The pre-emergence herbicides were sprayed at the rate of 500 liters spray solution ha<sup>-1</sup> on the third day of sowing and sufficient moisture was ensured at the time of spraying. The post emergence herbicide was sprayed at 20 DAS at the rate of 750 liters spray solution ha<sup>-1</sup>.

Periodical observations were recorded with respect to weed count by using quadrant of 0.5m x 0.5m size. The dry weight of weeds was also recorded after oven drying of the samples at 70 °C in hot air oven. At harvest, the data pertaining to growth such as plant height, number of tillers and yield attributes such as number of fingers per ear, finger length, test weight, grain yield and straw yield were recorded. The data was subjected to statistical analysis using standard procedures as described by Gomez and Gomez (1984) by using F test. The weed control efficiency and weed index were worked out using the procedures outlined

by Mani *et al.* (1973) and Gill and Vijaykumar (1969), respectively. The weed control efficiency and weed index were calculated using the formula given below.

$$\text{Weed control efficiency (\%)} = \frac{\text{Dry weight of weeds in weedy check plot} - \text{dry weight of weeds in weeded plots}}{\text{Dry weight of weed in weedy check plot}} \times 100$$

$$\text{Weed index} = \frac{\text{Seed yield from weed free plot} - \text{seed yield from treated plots}}{\text{Seed yield from weed free plot}}$$

## RESULTS AND DISCUSSION

### Effect of Weed Management Practices on Weeds

Weed dry weight and weed control efficiency were significantly influenced by different management practices. Higher weed dry weight indicates severe weed competition and exploitation of nutrients and moisture by weeds in unweeded control which deprives the crop from the same. The highest weed

TABLE I  
Weed dry weight (g m<sup>-2</sup>) and weed control efficiency (%) as influenced by different weed management practices in direct sown finger millet

Treatment details	Weed dry weight (g m <sup>-2</sup> )	WCE (%)
T <sub>1</sub> : Pendimethalin 30 EC @ 500 g a.i. ha <sup>-1</sup>	12.09 (145.3)	72.89
T <sub>2</sub> : Bensulfuronmethyl + pretialchlor 6.6 G@ 165 g a.i. ha <sup>-1</sup>	11.94 (142.0)	73.51
T <sub>3</sub> : 2,4- D Na salt 80WP @ 1000 g a.i.ha <sup>-1</sup>	10.18 (102.7)	80.85
T <sub>4</sub> : Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i.ha <sup>-1</sup>	10.02 (100.0)	81.34
T <sub>5</sub> : Pendimethalin 30 EC @ 500 g a.i./ha fb 2, 4- D Na salt 80WP @ 1000 g a.i.ha <sup>-1</sup>	8.02 (64.0)	88.06
T <sub>6</sub> : Pendimethalin 30 EC @ 500 g a.i./ ha fb Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i.ha <sup>-1</sup>	7.45 (54.7)	89.80
T <sub>7</sub> : Bensulfuronmethyl + pretialchlor 6.6 G @ 165 g a.i./ha fb2, 4-D Na salt 80 WP @ 1000 g a.i.ha <sup>-1</sup>	9.50 (89.3)	83.33
T <sub>8</sub> : Bensulfuronmethyl + pretialchlor 6.6 G @ 165 g a.i./ha fb Metsulfuron methyl + Chlorimuronethyl 20WP (2+2) 20 g a.i.ha <sup>-1</sup>	9.24 (84.7)	84.20
T <sub>9</sub> : Two intercultivations with one hand weeding	7.32 (52.7))	90.17
T <sub>10</sub> : Unweeded control	23.02 (536.0)	0.00
S.Em. (±)	0.69	-
C.D. @ (0.05)	2.04	-

dry weight (134.0 g) was observed in unweeded control treatment whereas the lowest was observed in two intercultivation and one hand weeding treatment (13.2 g). Sequential application of Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> (3 DAS) as pre-emergence herbicide followed by post emergence application of Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i. ha<sup>-1</sup> (20 DAS) or pre-emergence application of Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> (3 DAS) followed by post emergence application of 2, 4- D Na salt 80WP @ 1000 g a.i. ha<sup>-1</sup> (20 DAS) resulted in minimum weed dry weight and also higher weed control efficiency.

### Effect of Weed Management Practices on Growth Attributes

Growth attributes *viz.*, plant height, number of tillers per metre row length and dry matter plant<sup>-1</sup> were significantly influenced by different weed management practices. Significantly higher plant height (94.87 cm), tillers per metre row length (35.7) and dry matter plant<sup>-1</sup> (17.83 g) were observed in two intercultivations and one hand weeding treatment.

All herbicidal treatments resulted in higher growth attributes over unweeded control treatment. Among the herbicide treatments, sequential application of Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> (3 DAS) as pre-emergence herbicide followed by post emergence application of Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i. ha<sup>-1</sup> (20 DAS) or pre-emergence application of Pendimethalin 30 EC @ 500 g a.i. ha<sup>-1</sup> (3 DAS) followed by post emergence application of 2, 4-D Na salt 80WP @ 1000 g a.i. ha<sup>-1</sup> (20 DAS) resulted in higher growth attributes which were on par with that of the treatment receiving two intercultivations and one hand weeding, indicating better weed control with herbicides and also better utilization of resources. Least growth attributes were registered in unweeded control indicating severe crop weed competition for different resources. These results are in accordance with that of Linganagouda *et al.*, 2019 and Pawar *et al.*, 2021.

### Effect of Weed Management Practices on Yield Attributes

The important yield attributing characters *viz.*, number of fingers per ear, finger length (cm), ear head

TABLE 2

Growth attributes as influenced by different weed management practices in direct sown finger millet

Treatment details	Plant height at harvest (cm)	Tillers per metre row length	Plant dry weight at harvest (g plant <sup>-1</sup> )
T <sub>1</sub> : Pendimethalin 30 EC@ 500 g a.i. ha <sup>-1</sup>	77.87	30.0	10.70
T <sub>2</sub> : Bensulfuronmethyl+pretialchlor 6.6 G@ 165 g a.i. ha <sup>-1</sup>	68.53	25.0	9.87
T <sub>3</sub> : 2,4- D Na salt 80WP @ 1000 g a.i. ha <sup>-1</sup>	69.27	25.3	9.90
T <sub>4</sub> : Metsulfuron methyl +Chlorimuronethyl 20WP @ 20 g a.i. ha <sup>-1</sup>	70.53	25.7	10.30
T <sub>5</sub> : Pendimethalin 30 EC@ 500 g a.i. ha <sup>-1</sup> fb 2,4- D Na salt 80WP @ 1000 g a.i. ha <sup>-1</sup>	91.40	33.7	16.67
T <sub>6</sub> : Pendimethalin 30 EC@ 500 g a.i. ha <sup>-1</sup> fb Metsulfuron methyl + Chlorimuronethyl 20WP@ 20 g a.i. ha <sup>-1</sup>	92.00	34.3	17.40
T <sub>7</sub> : Bensulfuronmethyl+pretialchlor 6.6 G @ 165 g a.i. ha <sup>-1</sup> fb 2, 4-D Na salt 80 WP @ 1000 g a.i. ha <sup>-1</sup>	84.53	30.3	13.27
T <sub>8</sub> : Bensulfuronmethyl+pretialchlor 6.6 G@165 g a.i. ha <sup>-1</sup> fb Metsulfuron methyl +Chlorimuronethyl 20WP @ 20 g a.i. ha <sup>-1</sup>	86.07	30.7	14.07
T <sub>9</sub> : Two intercultivations with one hand weeding	94.87	35.7	17.83
T <sub>10</sub> : Unweeded control	57.87	21.7	7.33
S.Em. (±)	2.60	1.6	0.84
C.D.@(0.05)	7.74	4.9	2.49

weight (g plant<sup>-1</sup>) and test weight (g) were significantly influenced by different weed management practices. Significantly higher yield attributing characters were observed in treatment with two intercultivations and one hand weeding (T<sub>9</sub>) over rest of the treatments. However, by sequential application of Pendimethalin 30 EC @ 500 g a.i.ha<sup>-1</sup> (3 DAS) as pre-emergence herbicide followed by post emergence application of Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i.ha<sup>-1</sup> (20 DAS) or Pendimethalin 30 EC @ 500 g a.i.ha<sup>-1</sup> (3 DAS) as pre-emergence application followed by post emergence application of 2, 4-D Na salt 80WP @ 1000 g a.i.ha<sup>-1</sup> (20 DAS) resulted in higher yield attributes which were on par with that of two intercultivations and one hand weeding treatment. The higher values of yield attributes are the result of timely and effective weed control which led to reduced crop weed competition and increased growth attributes which finally resulted in higher yield attributes by better utilization of moisture, nutrients and sunlight by the crop. These

results are in harmony with that of Kamble *et al.*, 2015 in Maize, Kumar and Chawla (2019) in *kharif* maize, Linganagouda *et al.*, 2019 in direct seeded rice and Pawar *et al.*, 2021 in pearl millet. Lower yield attributes were observed in unweeded control treatment which was due to severe crop weed competition which results in reduced growth of crop and ultimately the reduced yield attributes.

### Effect of Weed Management Practices on Yield and Economics

Significantly higher grain yield (3520 kg ha<sup>-1</sup>) and straw yield (4825 kg ha<sup>-1</sup>) were recorded in the treatment with two intercultivations and one hand weeding (T<sub>9</sub>) over rest of the treatments. Among the herbicide treatments, sequential application of Pendimethalin 30 EC @ 500 g a.i.ha<sup>-1</sup> (3 DAS) as pre-emergence herbicide followed by post emergence application of Metsulfuron methyl + Chlorimuronethyl 20WP @ 20 g a.i.ha<sup>-1</sup> (20 DAS) or Pendimethalin 30 EC @ 500 g a.i.ha<sup>-1</sup> (3 DAS)

TABLE 3  
Yield attributes as influenced by different weed management practices in direct sown finger millet

Treatment details	No. of fingers per ear	Finger length (cm)	Ear head weight (g plant <sup>-1</sup> )	Test weight (g)
T <sub>1</sub> : Pendimethalin 30 EC@ 500 g a.i. ha <sup>-1</sup>	6.93	8.13	8.53	2.51
T <sub>2</sub> : Bensulfuronmethyl+pretialchlor 6.6 G@ 165 g a.i. ha <sup>-1</sup>	6.73	7.70	5.40	2.42
T <sub>3</sub> : 2,4- D Na salt 80WP @ 1000 g a.i. ha <sup>-1</sup>	6.73	7.77	6.47	2.42
T <sub>4</sub> : Metsulfuron methyl +Chlorimuronethyl 20WP @ 20 g a.i. ha <sup>-1</sup>	6.87	7.90	6.60	2.50
T <sub>5</sub> : Pendimethalin 30 EC@ 500 g a.i. ha <sup>-1</sup> fb 2,4- D Na salt 80WP @ 1000 g a.i. ha <sup>-1</sup>	8.27	9.53	10.07	2.59
T <sub>6</sub> : Pendimethalin 30 EC@ 500 g a.i. ha <sup>-1</sup> fb Metsulfuron methyl + Chlorimuronethyl 20WP@ 20 g a.i. ha <sup>-1</sup>	8.63	9.60	10.67	2.59
T <sub>7</sub> : Bensulfuronmethyl+pretialchlor 6.6 G @ 165 g a.i. ha <sup>-1</sup> fb 2,4- D Na salt 80 WP @ 1000 g a.i. ha <sup>-1</sup>	7.40	9.10	8.80	2.59
T <sub>8</sub> : Bensulfuronmethyl+pretialchlor 6.6 G@165 g a.i. ha <sup>-1</sup> fb Metsulfuron methyl +Chlorimuronethyl 20WP @ 20 g a.i. ha <sup>-1</sup>	7.47	9.17	8.87	2.59
T <sub>9</sub> : Two intercultivations with one hand weeding	8.73	9.73	10.93	2.60
T <sub>10</sub> : Unweeded control	5.33	6.60	6.67	2.41
S.Em. (±)	0.23	0.12	0.59	0.02
C.D.@(0.05)	0.70	0.34	1.76	0.07

TABLE 4

Yield and economics as influenced by different weed management practices in direct sown finger millet

Treatment details	Grain yield (kg ha <sup>-1</sup> )	Straw yield (kg ha <sup>-1</sup> )	Weed index	Cost of cultivation (Rs. ha <sup>-1</sup> )	Net returns (Rs. ha <sup>-1</sup> )	B:C ratio
T <sub>1</sub> : Pendimethalin 30 EC@ 500 g a.i. ha <sup>-1</sup>	2525	3310	28.3	37668	25380	1.67
T <sub>2</sub> : Bensulfuronmethyl + pretialchlor 6.6 G@ 165 g a.i. ha <sup>-1</sup>	1918	2464	46.9	37726	10084	1.27
T <sub>3</sub> : 2,4- D Na salt 80WP @ 1000 g a.i.ha <sup>-1</sup>	1926	2478	45.3	37620	10402	1.28
T <sub>4</sub> : Metsulfuron methyl + Chlorimuronethy l 20WP @ 20 g a.i.ha <sup>-1</sup>	2073	2750	41.1	39370	12434	1.32
T <sub>5</sub> : Pendimethalin 30 EC@ 500 g a.i./ha fb 2, 4- D Na salt 80WP @ 1000 g a.i.ha <sup>-1</sup>	3453	4582	1.9	38168	48132	2.26
T <sub>6</sub> : Pendimethalin 30 EC@ 500 g a.i./ ha fb Metsulfuron methyl + Chlorimuronethy l 20WP@ 20 g a.i.ha <sup>-1</sup>	3476	4722	1.3	39918	47114	2.18
T <sub>7</sub> : Bensulfuronmethyl + pretialchlor 6.6 G@ 165 g a.i./ha fb2,4-D Na salt 80 WP @ 1000 g a.i.ha <sup>-1</sup>	2979	3890	15.4	38226	36133	1.95
T <sub>8</sub> : Bensulfuronmethyl + pretialchlor 6.6 G@ 165 g a.i./ha fb Metsulfuron methyl + Chlorimuronethyl 20WP (2+2) 20 g a.i.ha <sup>-1</sup>	2986	3943	15.2	39976	34613	1.87
T <sub>9</sub> : Two intercultivations with one hand weeding	3520	4825	0.0	40320	47878	2.19
T <sub>10</sub> : Unweeded control	1460	1876	69.9	35920	474	1.01
S.Em. (±)	151.5	194.8				
C.D.@(0.05)	450.2	578.7				

as pre-emergence herbicide followed by post emergence application of 2, 4-D Na salt 80WP @ 1000 g a.i.ha<sup>-1</sup> (20 DAS) resulted in higher grain and straw yield which was on par with that of two intercultivations and one hand weeding treatment. This is due to the result of early and better weed control which resulted in improvement in all growth and yield attributing characters owing to higher yields. The lowest grain yield (1460 kg ha<sup>-1</sup>) and straw yield (1876 kg ha<sup>-1</sup>) were observed in the unweeded control due to severe weed competition put forth by weeds for space, light, nutrients and moisture throughout the crop growth period which resulted in lower growth and yield attributes and

ultimately lower yield. These outcomes are in concurrence with Pradhan *et al.*, 2012.

Among all the treatment combinations, sequential application of Pendimethalin 30 EC @ 500 g a.i.ha<sup>-1</sup> (3 DAS) as pre-emergence herbicide followed by post emergence application of 2, 4-D Na salt 80WP @ 1000 g a.i.ha<sup>-1</sup> (20 DAS) recorded higher net returns of 48132 Rs.ha<sup>-1</sup> and B:C ratio (2.26) compared to two intercultivations and hand weeding treatment (Rs.47878 ha<sup>-1</sup> and 2.19, respectively). This was mainly due to higher cost of cultivation which was because of higher labour charges. The results are in conformity with that of Linganagowda *et al.*, 2019 and Pawar *et al.*, 2021.

From the present investigation, it can be concluded that sequential application of Pendimethalin 30 EC @ 500 g a.i.ha<sup>-1</sup> (3 DAS) as pre-emergence herbicide followed by post emergence application of 2, 4-D Na salt 80WP @ 1000 g a.i.ha<sup>-1</sup> (20 DAS) in direct sown finger millet in areas of labour scarcity is an effective weed management strategy to minimize the losses caused by weeds and to enhance the productivity of finger millet.

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## Progression of Rice Sheath Blight in Relation to Weather Variables and Exploratory Development of Prediction Equations

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### ABSTRACT

Rice sheath blight caused by *Rhizoctonia solani* Kuhn has become an important constraint in rice cultivation in India and other tropical countries. The present study explores the effects of weather factors (temperature, relative humidity, rainfall and bright sunshine hours) on paddy sheath blight severity. During *kharif* 2020-21 at Zonal Agricultural Research Station, V.C. Farm, Mandya, all tested nine genotypes (Jyothi, Jaya, IR 64, Thanu, MTU 1001, MTU 1010, BR 2655, KRH4 and HR12) showed distinct responses to sheath blight disease. The disease started on the 28<sup>th</sup> November 2020 with a mean severity of 0.31 per cent, gradually increasing to a peak on 11<sup>th</sup> January 2021 (37.41%). The Jyothi (highly susceptible) genotype showed the highest mean disease severity (20.16%), whereas the BR 2655 which is moderately susceptible, showed the lowest mean sheath blight severity (10.90%). The optimal weather conditions for disease development were maximum temperature (25.50-29.62 °C), minimum temperature (16.50-18.50 °C), morning relative humidity (87.94-93.23%), evening relative humidity (53.74-75.54%), rainfall (2.00-20.50 mm) and sunshine (3.20-10.50 hours). Correlation analysis showed that the maximum air temperature was the key factor in governing the disease in the field among all the meteorological factors. A maximum temperature (25.50-29.62 °C) was found favor the development and spread of sheath blight after its establishment in the field. A predictive model was developed with a coefficient of determination ( $R^2$ ) of 0.804-0.848 using statistical language R. A step-wise multiple regression analysis approach was adopted to identify the most appropriate predictive variables to constitute the linear regression model.

**Keywords :** Rice sheath blight, Disease severity, Weather parameters, Correlation, Regression, Prediction equation

**R**ICE (*Oryza sativa* L.) is the most important staple food for more than half of the human population, providing approximately 19 per cent of the daily calories consumed worldwide (<https://esa.un.org>). It is a crop of Asian origin that belongs to the family Poaceae and about 90 per cent of the global rice area, production and consumption are concentrated in Asia. India is the world's second largest rice producer and consumer next to China. In India it is cultivated in an area of 44.44 Mha with a production of 112 Mt and productivity of 4.1 mt per hectare during 2020-21 (Deepak *et al.*, 2021).

Rice crops face several future challenges that will seriously jeopardise their annual production. Among these challenges, fungal diseases threaten rice production which cause decreasing annual yields and increasing cultivation costs. Among these sheath blights is a major production constraint in profusely tillering, fertilizer responsive, high yielding varieties and hybrids under intensive rice production systems. The yield losses ranging from 4-50 per cent have been reported depending on the crop stage at the time of infection, the severity of the disease and environmental conditions (Bhunkal *et al.*, 2015). This

disease is caused by the soil borne fungus *Rhizoctonia solani* Kuhn [teleomorph: *Thanatephorus cucumeris* (Frank) Donk]. In India, it was first reported from Gurdaspur by Paracer and Chahal (1963) and has become a major production constraint in different paddy growing states of India due to the widespread cultivation of high yielding rice varieties with a narrow genetic base and apparent change in the climate.

Introduction of high-yielding varieties, there has been a considerable increase in the dosage of nitrogen application, the number of plants/unit area and the number of irrigations. All these factors result in luxuriant plant growth, thick stand of the crop and constant humid conditions coupled with high temperature during the rainy (*Kharif*) season. Little was known regarding the influence of meteorological factors that influence the continuous build-up of sheath blight in the field. Weather-based prediction models have been used to forecast rice diseases like bacterial leaf blight and blast disease (Hashimoto *et al.*, 1984 and Calvero *et al.*, 1996) but there was very little initiative to model sheath blight incidence using epidemiological parameters, particularly in Indian condition. Hence, an attempt has been made to elucidate the effect of weather parameters on sheath blight severity and to develop a prediction model that could guide the management decisions and will be very helpful in reducing yield loss.

## MATERIAL AND METHODS

### Field Experiment

To explicate the influence of weather parameters on the development of sheath blight disease, the field experiment was conducted during the late *kharif* 2020-21 at V.C. Farm, Mandya (12°34' N latitude, 76°50' E longitude and at an altitude of 695.0 m above mean sea level).

### Plant Material, Nursery Sowing and Transplanting

The planting materials of nine selected genotypes *viz.*, Jyothi, Jaya, IR 64, Thanu, MTU 1001, MTU 1010, BR 2655, KRH4 and HR12 were collected from Zonal Agricultural Research Station (ZARS), Mandya. Nursery was prepared on raised beds with seeds of

each genotype in individual small plots (3 x 3 sq. m). One-month old rice seedlings of individual cultivars were transplanted into the plot of 3m x 3m size. A recommended fertilizer dose of N: P: K at the rate of 58:23:25 kg/acre and routine cultural practices were performed to sustain a vigorous crop stand.

### Disease Severity

After transplanting, the onset time of disease was monitored as the appearance of first symptoms and sheath blight severity was assessed in the field. The severity of the disease was scored using a 0-9 rating scale (IRRI, 2013). Twenty plants per plot were randomly selected in the 'Z' pattern to record the disease severity under natural epiphytotic conditions. The whole plant was assessed at 2 days intervals upto 15 days before harvest. Per cent disease index (PDI) was calculated following the standard formula (Mckinney, 1923).

$$\text{PDI} = \frac{\text{Sum of scores}}{\text{Number of observations assessed} \times \text{highest number in disease rating scale}} \times 100$$

### Correlation and Regression

Correlation and linear regression among the metrological factors and disease severity were determined to study the epidemiology of the rice sheath blight pathosystem. The weather variables *i.e.*, maximum temperature (°C), minimum temperature (°C), morning and evening humidity (%), rainfall (mm) and sunshine hours were obtained from the Meteorological Observatory, V.C. Farm, Mandya for the period of investigation. Meteorological factors significantly affecting the disease severity were identified and established by the correlation and linear regression analysis. The meteorological factors were set as independent variables and disease severity served as the dependent variable (Alase *et al.*, 2021 and Jayashree *et al.*, 2022).

### Statistical Analysis

All the collected data sets were statistically subjected to the correlation of disease severity with the weather factors and linear regression analysis to identify the

responsive variable using the R version 4.1.0 (R Core Team, 2020).

TABLE 1  
Scale for scoring rice sheath blight as per IRRI (2013)

Scale	Relative lesion height
0	No infection
1	Vertical spread of the lesion up to 20% of plant height
3	Vertical spread of the lesion up to 21 - 30% of plant height
5	Vertical spread of the lesion up to 31 - 45% of plant height
7	Vertical spread of the lesion up to 46 - 65% of plant height
9	Vertical spread of the lesion up to 66 - 100% of plant height

RESULTS AND DISCUSSION

Response of Various Rice Genotypes to Sheath Blight Disease

Conducive environmental conditions facilitate early disease initiation, rapid disease development and the highest disease pressure in the highly susceptible genotypes (Jyothi, HR 12, Jaya, MTU 1001, IR 64, and KRH 4). In moderately susceptible genotypes (BR 2655, MTU 1010 and Thanu), delayed disease initiation, slower disease development rates and lower mean disease severities were recorded. Jyothi showed maximum disease severity (45%) followed by Jaya (42.78%), IR 64 (40.56%), MTU 1001 (40.53%), HR12 (39.44%), and KRH 4 (38.33%) indicating the highly susceptible response. Similarly, the minimum disease severity was recorded on BR 2655 (29.44%), MTU 1010 (30%) and Thanu (30.56%), which showed moderately susceptible response to rice sheath blight disease (Table 2 and Fig. 1). The findings were in accordance with the results of Prasad *et al.* (2010) conducted an experiment on evaluation of rice genotypes against *Rhizoctonia solani*, which indicated that the rice varieties such as HR 12 and IR 64 were susceptible to sheath blight.

TABLE 2  
Weather parameters during assessment period and sheath blight disease severity

Date	Tmax (°C)	Tmin (°C)	RHI (%)	RHII (%)	RF (mm)	SSH (Hours)	IR 64	HR-12	KRH - 4	Thanu	BR-2655	MTU 1001	Jyothi	MTU 1010	Jaya	Mean
28/11/2020	25.50	17.25	90.50	74.75	2.00	10.50	0.28	0.28	0	0.00	0.00	0.28	1.39	0.00	0.56	0.31
2/11/2020	27.25	17.88	91.04	65.27	0.00	10.00	1.67	1.39	0.83	0.00	0.00	1.67	2.50	0.00	1.39	1.05
6/12/2020	27.13	17.75	93.09	75.54	0.50	0.00	4.44	4.72	3.06	2.50	1.67	3.89	4.72	1.11	4.17	3.36
10/12/2020	28.38	18.50	93.23	65.39	14.50	9.00	7.22	6.67	6.11	4.44	3.61	6.94	6.94	4.17	6.94	5.89
14/12/2020	27.63	16.50	91.39	53.74	16.00	0.00	10.00	9.72	8.33	5.83	4.72	10.28	10.00	5.28	9.44	8.18
18/12/2020	28.00	17.00	90.64	62.96	10.50	0.00	12.50	13.33	11.11	6.94	5.83	15.28	13.89	7.22	12.50	10.96
22/12/2020	28.13	17.50	91.45	63.14	12.80	0.00	18.33	19.17	13.06	10.28	9.44	19.72	20.83	10.28	21.39	15.83
26/12/2020	29.13	17.50	92.93	55.46	18.00	0.00	25.00	27.22	17.5	15.83	11.94	23.89	26.11	14.44	25.00	20.77
30/12/2020	29.62	17.87	89.58	68.67	20.5	0	29.44	31.11	24.17	19.72	17.22	28.06	32.22	18.06	28.89	25.43
03/1/2021	28.37	16.50	87.94	70.29	11.5	12.4	32.22	32.22	30.28	23.33	21.39	32.24	37.22	22.22	34.44	29.50
07/1/2021	29.25	17.25	90.60	72.96	9.5	3.2	36.67	35.56	34.44	26.67	25.56	36.69	41.11	26.11	38.89	33.52
11/1/2021	28.50	17.625	88.29	69.5	0	0	40.56	39.44	38.33	30.56	29.44	40.53	45.00	30.00	42.78	37.41
						Mean PDI	8.19	18.40	15.60	12.18	10.90	18.29	20.16	11.57	18.87	16.02

Tmax= Maximum temperature, Tmin= Minimum temperature, RHI= Morning relative humidity, RHII= Evening relative humidity, RF= Rainfall and SSH= Sunshine hours

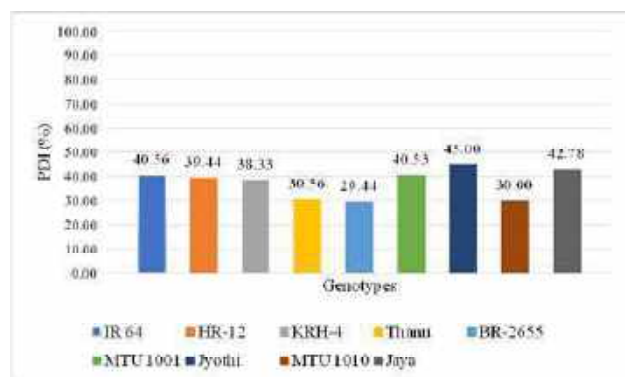


Fig.1 : Sheath blight Severity of different rice genotypes

### Progress of Rice Sheath Blight Disease under Natural Condition

In the present investigation, disease development in relation to weather parameters was studied as described in ‘Material and Methods’. This study depicts the relationship between the weather parameters like maximum and minimum temperature (Tmax and Tmin), morning and evening relative humidity (RHI and RHII), total rainfall (RF) and bright sunshine hour (SSH) with sheath blight disease severity. The frequency of disease intensity (PDI) was scored from 28<sup>th</sup> November *kharif*2020 to 6<sup>th</sup> December *Kharif* 2020 at two days interval for nine genotypes were worked out at four days intervals and represented in Table 2.

Different rice genotypes showed varying responses to rice sheath blight disease under the influence of significant meteorological conditions. During the investigation, disease progress in relation to weather parameters revealed that initially, on 28<sup>th</sup> November *kharif* 2020, disease severity was very low (0.31%), which increased gradually to 37.41 per cent (11-1-2021). The sheath blight disease incidence initiated on 28<sup>th</sup> November *kharif* 2020 is highly susceptible genotypes (Jyothi, HR 12, Jaya, MTU 1001 and IR 64) and from 6<sup>th</sup> December *kharif* 2020 in moderately susceptible genotypes (Thanu, BR 2655 and MTU 1010). In all the nine genotypes, the per cent disease severity (PDS) showed linear progression throughout the cropping season. From Table 2, it is evident that gradual progress in mean disease severity was observed from 28<sup>th</sup> November

*kharif* 2020 onwards. It was coincided with favorable weather conditions *viz.*, maximum temperature (25.25 - 26.50 °C), minimum temperature (17-18 °C), morning relative humidity (90-91%) and evening relative humidity (65-85%). The maximum mean disease severity of rice sheath blight occurred on 11<sup>th</sup> January 2021 with the weather parameters like maximum temperature (25.50-29.62 °C), minimum temperature (16.50-18.50 °C), morning relative humidity (87.94-93.23%) and evening relative humidity (53.74-75.54%).

An increase in disease severity is mainly attributed to weather conditions predominating in an area or particular year. *Rhizoctonia* is a soil borne pathogen that can survive in the soil for years. The amount of infective propagules available in the soil or crop debris tends to be a crucial factor affecting the growth and development of sheath blight disease in rice. The survival of this infective propagule in the soil is influenced by the environmental factors suitable for the pathogen’s growth. Temperature is a major physiological factor affecting crop production (Yitbarek *et al.*, 1988). When the weather condition is humid and temperatures are stressful to the crop (25-30 °C), sheath blight tends to develop which is been recorded as one of the most destructive pathogen. Gill *et al.* (2001) viewed *R. solani* anastomosis group (AG8), concluding that significant damage was caused when the temperature was lower than 6 to 19 °C at the root region, or when the temperature ranges from 16 to 27 °C.

### Correlation of Rice Sheath Blight with Weather Parameters

The sheath blight disease severity of all nine genotypes and overall mean disease severity were separately correlated with weather variables *i.e.*, maximum temperature (°C), minimum temperature (°C), morning and evening humidity (%), rainfall (mm) and sunshine hours during the investigation period. The correlation coefficients in relation to weather parameters of nine genotypes are presented in Table 3.

TABLE 3  
Correlation of weather parameters with sheath blight disease severity recorded on nine genotypes

Weather Parameters	IR 64	HR-12	KRH-4	Thanu	BR2655	MTU 1001	Jyothi	MTU 1010	Jaya	Mean
X <sub>1</sub>	0.767 **	0.786 **	0.711 **	0.723 **	0.683 *	0.758 **	0.742 **	0.710 **	0.743 **	0.742 **
X <sub>2</sub>	-0.185	-0.183	-0.214	-0.182	-0.180	-0.216	-0.203	-0.188	-0.197	-0.196
X <sub>3</sub>	-0.595 *	-0.577 *	-0.655 *	-0.625 *	-0.655 *	-0.609 *	-0.628 *	-0.638 *	-0.604 *	-0.621 *
X <sub>4</sub>	0.068	0.040	0.145	0.133	0.182	0.056	0.105	0.131	0.087	0.101
X <sub>5</sub>	0.261	0.302	0.173	0.190	0.127	0.253	0.229	0.173	0.233	0.222
X <sub>6</sub>	-0.275	-0.302	-0.207	-0.224	-0.195	-0.287	-0.242	-0.214	-0.257	-0.249

\*\* : Correlation is significant at the 0.01 level (2-tailed); \* : Correlation is significant at the 0.05 level (2-tailed). X<sub>1</sub> = Maximum temperature (°C), X<sub>2</sub> = Minimum temperature (°C), X<sub>3</sub> = Morning relative humidity (%), X<sub>4</sub> = Evening relative humidity (%), X<sub>5</sub> = Rainfall (mm) and X<sub>6</sub> = Bright sunshine hours (Hours).

The per cent disease severity correlated positively and was highly significant with maximum temperature in all nine genotypes with correlation coefficient (r) ranging from -0.683 to -0.786 in BR 2655 and HR12, respectively. Whereas, in relation to morning relative humidity, per cent disease severity of all nine genotypes showed a significantly negative correlation with correlation coefficient (r) ranging from -0.577 (HR12) to -0.655 (KRH 4 and BR 2655). In all nine genotypes under study, minimum temperature and bright sunshine hours showed a negative correlation in disease progression whereas, evening relative humidity and rainfall showed positive correlation in relation to disease development (Table 3 and Fig. 2).

The mean per cent disease severity showed a strong positive association of maximum temperature with a correlation coefficient (r) of 0.742 and a negative association of morning relative humidity (-0.621). Whereas, minimum temperature and bright sunshine hours showed a positive correlation and evening relative humidity and rainfall showed negative correlation with mean per cent disease severity however, they remained non-significant in contributing to the disease development (Fig.1). Such results signify that a relatively higher air temperature was more conducive to intensify disease incidence. The results of this study confirmed with findings of Thakur *et al.*, (2017) where the correlation analysis between weather parameters and rice sheath blight disease during *Kharif* 2013, 2015 and 2016 revealed that Tmax (30.50- 32.60 °C) and SSH (4.2-9.6 hrs) had a positive effect in the development of sheath blight disease of rice whereas, minimum temperature, morning and evening relative humidity had negative effect on disease development in all *Kharif* seasons of 2013-2016. Yadav *et al.* (2019) also analyzed the correlation of root rot of fenugreek with weather variables and observed a significant positive correlation with Tmax = 0.668 and negative correlation with Tmin = -0.039, RHm -0.457, RHe -0.261 and RF- 0.333. A field experiment was conducted during the rainy (*kharif*) of 2007 and 2008 on rice sheath blight. Correlation analysis showed

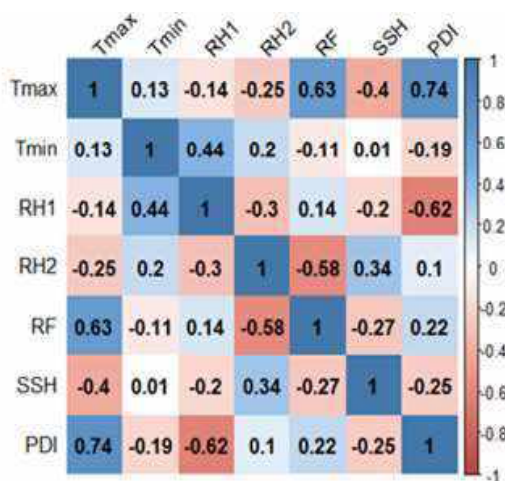


Fig. 2 : Heat map depicting the correlation between mean sheath blight disease severity with weather parameters

that, a maximum temperature around 34 °C and a minimum temperature around 26 °C favourable for the spread of sheath blight after its establishment in the field. Again, high morning relative humidity of more than 90 per cent facilitates the spreading the disease (Biswas *et al.*, 2011).

The relationship of maximum temperature with disease severity on most genotypes was positively correlated (Fig.3a). A increase in temperature resulted in increased disease severity (Table 2). The mean disease severity was increased when the temperature increased from 25.50 °C (28-11-2020)

to 28.50 °C (11-1-2021). In parallel with maximum temperature, the correlation of morning relative humidity with disease severity on most genotypes was negative (Fig. 3c). Though minimum temperature, evening relative humidity and rainfall play a significant role in rice sheath blight epidemics, they were non-significant in contributing to sheath blight severity in the present study (Table 3). A possible reason could be the presence of autocorrelation within these independent variables. Most of the data points lie within 16.50- 18.50 °C (minimum temperature), evening relative humidity (63-75%) and rainfall (10-20 mm) (Table 2). Although these

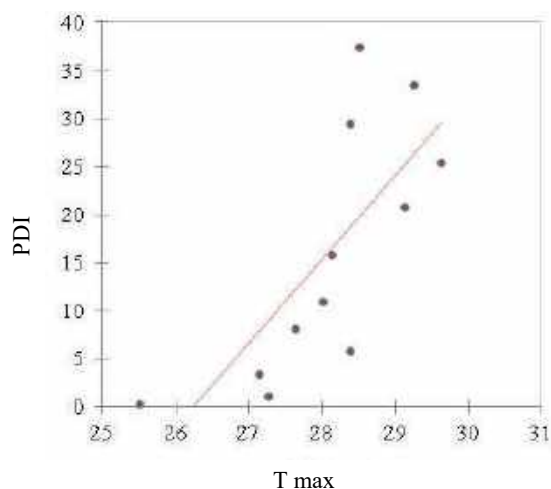


Fig. 3a : Relationship of maximum temperature with mean rice sheath blight severity

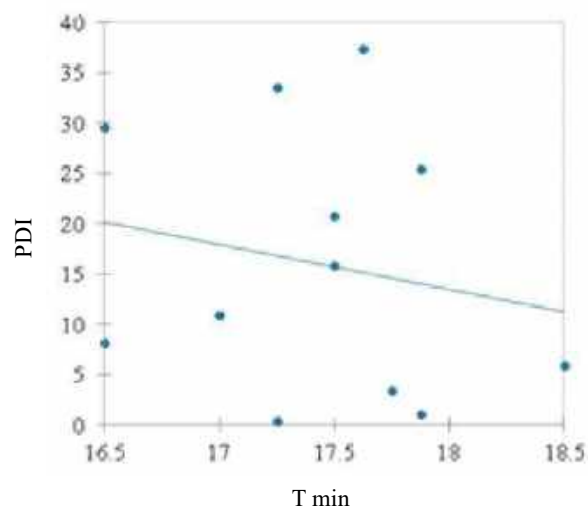


Fig. 3b : Relationship of minimum temperature with mean rice sheath blight severity

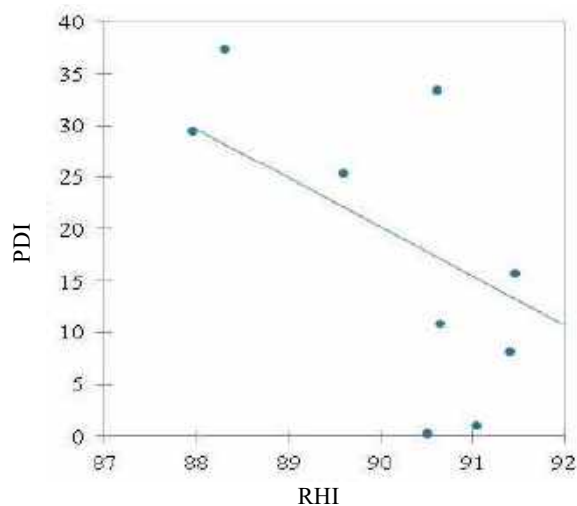


Fig. 3c : Relationship of morning relative humidity with mean rice sheath blight severity

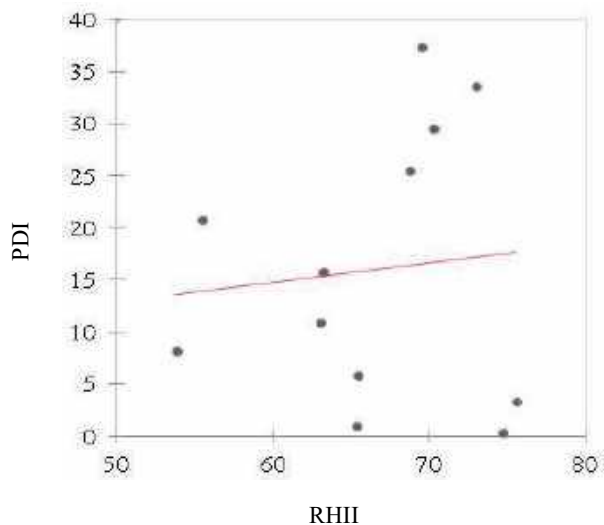


Fig. 3d : Relationship of evening relative humidity with mean rice sheath blight severity

parameters contribute to sheath blight severity at field conditions, no significant difference was observed between these variables between time intervals, making them statistically irresponsive to rice sheath blight severity.

Overall, from Table 3 it is observable that when, weather conditions *viz.*, maximum temperature (25.50-29.62 °C), minimum temperature (16.50 - 18.50 °C), morning relative humidity (87.94-93.23%), evening relative humidity (53.74-75.54%), rainfall (2-20.50 mm) and sunshine (3.20-10.50 hrs) prevails in an area result in rice sheath blight severity.

The mean per cent disease severity showed a strong positive association of maximum temperature with a correlation coefficient ( $r$ ) of 0.742 and a negative association of morning relative humidity (-0.621). Whereas, minimum temperature and bright sunshine hours showed a positive correlation and evening relative humidity and rainfall showed negative correlation with mean per cent disease severity however, they remained non-significant in contributing to the disease development (Fig.+1). Such results signify that a relatively higher air temperature was more conducive to intensify disease incidence. The results of this study confirmed with findings of Thakur *et al.* (2017) where the correlation analysis between weather parameters and rice sheath blight disease during *khariif* 2013, 2015 and 2016 revealed that Tmax (30.50-32.60 °C) and SSH (4.2-9.6 hrs) had a positive effect in the development of sheath blight disease of rice whereas, minimum temperature, morning and evening relative humidity had negative effect on disease development in all *khariif* seasons of 2013-2016. Yadav *et al.* (2019) also analyzed the correlation of root rot of fenu greek with weather variables and observed a significant positive correlation with Tmax = 0.668 and negative correlation with Tmin = -0.039, RHm -0.457, RHe -0.261 and RF -0.333. A field experiment was conducted during the rainy (*Khariif*) of 2007 and 2008 on rice sheath blight. Correlation analysis showed that, a maximum temperature around 34°C and a minimum temperature around 26 °C favourable for the spread of sheath blight after its establishment

in the field. Again, high morning relative humidity of more than 90 per cent facilitates the spreading the disease (Biswas *et al.*, 2011).

The relationship of maximum temperature with disease severity on most genotypes was positively correlated (Fig. 3a). A increase in temperature resulted in increased disease severity (Table 2). The mean disease severity was increased when the temperature increased from 25.50 °C (28-11-2020) to 28.50 °C (11-1-2021). In parallel with maximum temperature, the correlation of morning relative humidity with disease severity on most genotypes was negative (Fig. 3c). Though minimum temperature, evening relative humidity and rainfall play a significant role in rice sheath blight epidemics, they were non-significant in contributing to sheath blight severity in the present study (Table 3). A possible reason could be the presence of auto correlation within these independent variables. Most of the data points lie within 16.50-18.50 °C (minimum temperature), evening relative humidity (63-75%) and rainfall (10-20 mm) (Table 2). Although these parameters contribute to sheath blight severity at field conditions, no significant difference was observed between these variables between time intervals, making them statistically irresponsive to rice sheath blight severity.

Overall, from Table 3 it is observable that when, weather conditions *viz.*, maximum temperature (25.50-29.62 °C), minimum temperature (16.50-18.50 °C), morning relative humidity (87.94-93.23%), evening relative humidity (53.74-75.54%), rainfall (2-20.50 mm) and sunshine (3.20-10.50 hrs) prevails in an area result in rice sheath blight severity.

### Multiple Regression Analysis

The multiple regression analysis was performed for six independent weather variables to identify critical and much contributing weather variable (s) separately towards the dependent variable *i.e.*, sheath blight disease severity, for all the nine genotypes. In multiple regression analysis except for the maximum temperature and morning relative humidity, other parameters were non-significant in contributing to the

TABLE 4  
Stepwise multiple linear regression equations for the prediction of sheath blight severity

Genotypes	Regression equation	R	R <sup>2</sup>
IR 64	$Y = 143.653 + 8.907 X_1 - 4.132 X_2$	0.914	0.835
HR 12	$Y = 120.499 + 9.168 X_1 - 3.955 X_2$	0.918	0.848
KRH 4	$Y = 203.697 + 7.584 X_1 - 4.412 X_2$	0.908	0.824
Thanu	$Y = 140.960 + 6.254 X_1 - 3.349 X_2$	0.897	0.805
BR 2655	$Y = 165.323 + 5.562 X_1 - 3.417 X_2$	0.889	0.790
MTU 1001	$Y = 158.573 + 8.660 X_1 - 4.218 X_2$	0.914	0.836
Jyothi	$Y = 203.793 + 9.485 X_1 - 4.950 X_2$	0.913	0.834
MTU 1010	$Y = 150.045 + 5.972 X_1 - 3.368 X_2$	0.897	0.804
Jaya	$Y = 172.066 + 9.086 X_1 - 4.492 X_2$	0.900	0.810
Mean	$Y = 162.101 + 7.853 X_1 - 4.033 X_2$	0.909	0.826

$X_1$  = Maximum temperature (°C);  $X_2$  = Morning relative humidity (%)

disease severity. This is attributed to the multi co-linearity factor that existed in between the independent variables. Further data was subjected to stepwise regression analysis to find significant contributing variables. Results revealed that maximum temperature and morning relative humidity were the parameters that contributed more to disease severity. Based on this result, prediction equations were formulated for all nine genotypes by employing significant variables *viz.*, maximum temperature and morning relative humidity. The regression coefficient based on stepwise regression analysis for per cent disease severity of sheath blight with respect to significant weather parameters *viz.*, maximum temperature and morning relative humidity has been worked out and presented in Table 4.

The results indicated a multiple linear regression equation, with R-value ranging from 0.897-0.918 in all nine genotypes, indicating a strong association between per cent disease severity with maximum temperature and morning relative humidity. The coefficient of determination value (R<sup>2</sup>) was found to be, 0.804-0.848 indicating that 80.40-84.80 per cent of the variation in sheath blight disease severity was explained by the function of the weather parameters *viz.*, maximum temperature and morning relative humidity. Using these multiple regression models

makes it possible to predict disease in advance and the epidemic nature of the disease could be prevented by timely application of the management measures. The generated results are supported by the findings of Biswas *et al.* (2011), who developed the multiple regression equation in variety PR 115 and PR 116 for sheath blight disease severity (R<sup>2</sup> = 80.48 %). Bhukal *et al.* (2015) also developed a regression equation for the sheath blight disease in rice varieties like HKR 127 and Basmati CSR 30 with a model efficiency of 59 - 98 per cent (R<sup>2</sup>: 0.59-0.98). The root rot of fenugreek was greatly favoured by maximum temperature (28.8 to 30.50 °C) and negative with rainfall. The coefficient of multiple determinations (R<sup>2</sup>) was 61.02 and 75.04 per cent during 2016-17 and 2017-18 (Yadav *et al.*, 2019).

The mean sheath blight PDI values were compared with predicted values. The predicted values were fluctuated around the observed values indicating a good association between them. So, this model can be used to predict sheath blight incidence and thus fungicide application schedule can be arranged accordingly and subject to further evaluation under field conditions (Fig. 4).

The present study revealed that a Tmax range between 25-30 °C played a major role in the progression of sheath blight disease. The dynamic process of plant



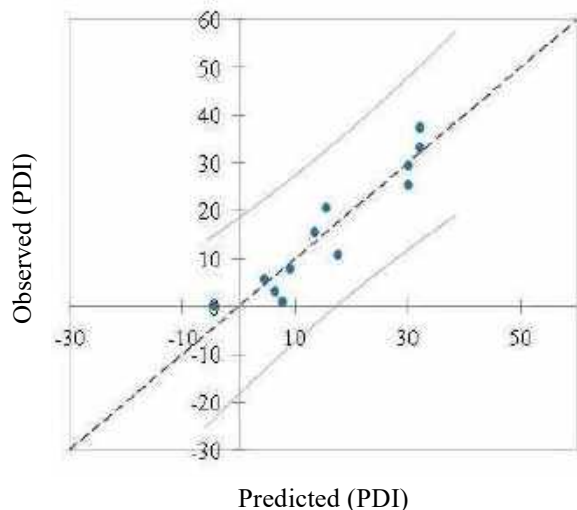


Fig. 4 : Performance of the predictive model  
(Mean  $Y = 162.101 + 7.853 X - 4.033 X^2$ )

disease depends upon the interactions among the host, pathogen and the environment. The variation in any one of the factors influences disease development. Meteorological conditions during rice cultivation periods are subject to change annually and on an hour basis. It is difficult to determine when and how meteorological conditions influence the outbreak of rice sheath blight. Rice sheath blight epidemics differ from field to field even under the same meteorological conditions as the cultivation methods differ. The disease management strategies according to changing climatic conditions with the amalgamation of new strategy will be useful for sustainable food production. Thus, if a sound forewarning system is developed, the epidemic nature of the disease could be prevented by the timely application of the management measures. Such a system will help reduce production cost and promote environmental safety by reducing chemical usage.

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## Growth in Production and Export Performance of Indian Coffee in International Market

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B. N. PRADEEPA BABU :  
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### ABSTRACT

The second most traded commodity after oil in the world is coffee. It is an interesting fact that the coffee drink is preferred by the people of developed countries and crop is grown in developing countries. Therefore, it is the developing countries who are the major producers and exporters of coffee to the world. Brazil is the largest producer and exporter of coffee in the world and India is the seventh largest producer and eighth largest exporter of coffee. The study attempts to analyze the trends, growth and instability in the area, production and productivity of coffee in major coffee producing countries and India. The production and yield of coffee had increased in all the major coffee producing countries from pre- liberalization to post liberalization period. The area had decreased in Brazil and Colombia but in other countries including India the area had increased in both the periods. The stability of the area, production and productivity remains unstable (>20%) over the years in the countries including India however the instability started to decline (15-20%) in the post-liberalization period. The export competitiveness of Indian coffee in the international market has been analyzed using the Trade Intensity Index. The results showed that India had competitive advantage in exporting coffee to Italy, Belgium and Russia which are the major importers of coffee in the world.

**Keywords :** Area, Export, Instability index, Liberalization, Production & productivity of coffee, Trade intensity index

Coffee is brewed drink prepared from roasted coffee beans, which are the seeds of berries from the coffee plant. The plant is native to subtropical Africa and some islands of southern Asia. Coffee is produced in about 70 countries most of them belonging to the developing world from Africa, Latin America and Asia. In contrast, coffee is mostly consumed in developed world in America and Europe, thus making it the world's highest traded commodity after petroleum. The countries that produce the most (75%) coffee are Brazil (63.4 million kg), Vietnam (29 million kg), Colombia (14.3 million kg), Indonesia (12 million kg), Ethiopia (7.3 million kg) and Honduras (6.1 million kg). India is the seventh largest coffee producing country (5.7 million kg) that accounts for three per cent of global coffee output

(International Coffee Organization (ICO), 2019). According to the International Coffee Organization report 2019, a total of 10,176 million kg of coffee were produced worldwide in 2020. Brazil is the largest producer and exporter of coffee in the world. The productivity does not stand the same as Vietnam's productivity is higher compared to Brazil. The productivity of Vietnam in the year 2019 was 2.71 t/ha in the year 2019 followed by Brazil (1.65 t/ha), Colombia (1.04 t/ha), Ethiopia (0.64 t/ha) and Indonesia (0.61 t/ha) (Assefa *et al.*, 2021). In India, Arabica and Robusta are both produced, but Robusta dominates production at 70 per cent, with Arabica making up the remaining 30 per cent. Mohan *et al.* in 2012 estimated that area under coffee showed an increasing trend in India from 1990 to 2010. The area

under coffee during the year 2020 was 422924 ha, production was 3,34,000 million tonnes and productivity of 0.79 tonnes per ha (Coffee Board of India, 2021).

Coffee market had undergone a series of changes since the establishment of International Coffee Organization, 1962. From 1962 to 1988 coffee was traded under a system of quotas. From 1989 the coffee market has been liberalized. Internationally, coffee trade takes place at two major exchanges *viz.*, Inter-Continental Exchange-ICE (previously known as NYBOT - New York Board of Trade) in respect of Arabica and London International Financial Futures & Options Exchange (LIFFE) in respect of Robusta. Bill *et al.*, in 2021 mentioned that liberalization was one of the reasons for increase in world coffee production and supply of better quality coffee for consumption.

The deregulation of coffee market or liberalization of coffee market increased the prices paid to the producers and at the same time increased the price volatility. Since then the coffee production has started to concentrate in fewer origins and new markets have emerged on the demand side. India is the eighth largest exporter of coffee by volume. According to Food and Agriculture Organization (FAO) statistics, 2021 almost one-third of the country's total coffee exports constitute instant coffee. Indian coffee exports display seasonality with exports peaking from March to June. The country exports over 70 per cent of its production, in 2021-22, the total exports recorded 42 per cent rise to US\$ 1.04 billion from 2020-21. In March 2022, exports of coffee were valued at US\$ 114.7 million, a 22 per cent growth from February 2022. However over the years with the liberalization of coffee market the production and export performances of coffee producers has taken major shifts and turns to compete in the international market. The labor availability in coffee estates of India is facing the problem of migration due to price volatility as income generation became unstable (Sagar *et al.*, 2021). With this background the study focuses on production and export aspects of Indian coffee in the international market. To analyze the trends and patterns the study

tries to assess the change in area and production of coffee before and after the coffee market liberalization as well as study the major export destinations of Indian coffee in the international market.

## MATERIAL AND METHODS

The present study focuses on secondary data of area, production and productivity of coffee of major coffee producing countries (Brazil, Vietnam, Columbia, Indonesia and Honduras) and India. The data was collected from FAOSTAT for a period of 1961 to 2020. The data has been divided into two periods (pre-liberalization (1961-1989) and post-liberalization (1990-2020) to know the changes in area and production due to liberalization of coffee market. As well as to calculate the export performance of Indian coffee secondary data from Trade Map was collected for 2002 to 2021 years. The following analyses carried in the study are:

*Trend Lines of Area, Production and Productivity of Coffee* : The trend lines are fitted across the countries from a period of 1961 to 2020 for area, production and productivity. This helps to observe the fluctuations across the periods with a view of coffee market liberalization.

*Compound Annual Growth Rates of Area, Production and Productivity of Coffee Across Five Countries And India* : The compound growth rate is estimated from the following form

$$Y = ab^t e \quad \dots\dots\dots (1)$$

where,

'Y' is the dependent variable (area, production and productivity).

'a' is the intercept term.

'b' is the regression coefficient that measures the relative change in Y for a given absolute change in independent variable t.

't' is the dependent variable.

'e' is the error term.

Eq (1) is converted to linear form by taking log on both sides of the equation and it forms the following

form,

$$\ln Y = \ln a + t \ln b \quad \dots\dots\dots(2)$$

The per cent compound growth rate takes the form

$$\text{CAGR (g)} = [\text{antilog } b - 1] \times 100 \quad \dots\dots\dots (3)$$

*Instability analysis* : The study uses Cuddy-Della Valle Index (CDVI) to measure the instability in harvested area, production and productivity of coffee. This index is preferred over the normal coefficient of variation (CV) as it attempts to de-trend the CV by using co-efficient of determination and showing the exact direction of instability (Cuddy and Valle, 1978).

CDVI is obtained for the CV and the following form of CV is

$$\text{CV} = \frac{\text{standard deviation}}{\text{mean}} \times 100 \dots\dots\dots (4)$$

CDVI is estimated as follows

$$\text{CDVI} = \text{CV} \times \sqrt{1-R^2} \quad \dots\dots\dots (5)$$

where,

CV is the coefficient of variation in percentage

$R^2$  is the coefficient of determination from the regression adjusted for its degrees of freedom. If the index values below 15 per cent then it is categorized as low instability, if the value lies between 15 to 20 per cent then it is categorized as medium instability and more than 20 per cent is categorized as high instability.

*Export performance of Indian coffee* : To analyze the export performance, trade intensity index was calculated to indicate whether a country exports more, as a percentage, to a partner than the world does on an average. The trade intensity index is obtained by the following formula :

$$\text{TII} = 100 \times \frac{(X_{ijk} / X_{ik})}{(X_{wjk} / X_{wk})} \quad \dots\dots\dots (6)$$

where,

' $X_{ijk}$ ' = value of exports of product 'k' from origin country 'i' to destination 'j'

' $X_{ik}$ ' = total exports from 'i' of product 'k'

'w' = indicates the world as origin

Value greater than 100 indicates a relationship more intense than the world average for the partner.

## RESULTS AND DISCUSSION

*Trend Lines of Area, Production and Productivity of Coffee* : The trend lines were fitted for area, production and productivity of coffee in Brazil, Vietnam, Columbia, Indonesia, Honduras and India from the year 1961 to 2020. To depict whether changes exist between pre and post coffee market liberalization a line has been marked at 1989 in the graphs.

Fig. 1, indicates that area under coffee in Brazil has been decreasing from the pre-liberalization period due to frost combined with drought over the decades. Area under Colombia increased in the pre-liberalization due to stable market prices and in the post-liberalization period area had decreased due to price volatility in post-liberalization period. Other countries like Indonesia, Vietnam, Honduras and India have shown an increasing trend in the area to the post-liberalization of coffee market which shows the liberalization had a positive impact in these countries.

Fig. 2, showed that production in the pre-liberalization period was increasing at a lesser rate and in post-independent period the increasing trend was higher in Brazil, Vietnam, Indonesia, Honduras and India. Production increased due to removal of quota restrictions that were imposed on the exports prior to liberalization and planting of high yielding varieties that increased production.

Fig. 3, depicts that productivity remained almost constant in the pre-liberalization period of these countries. But after coffee liberalization in 1989 the productivity increased due to increase in competition to produce high quality coffee seeds and improved

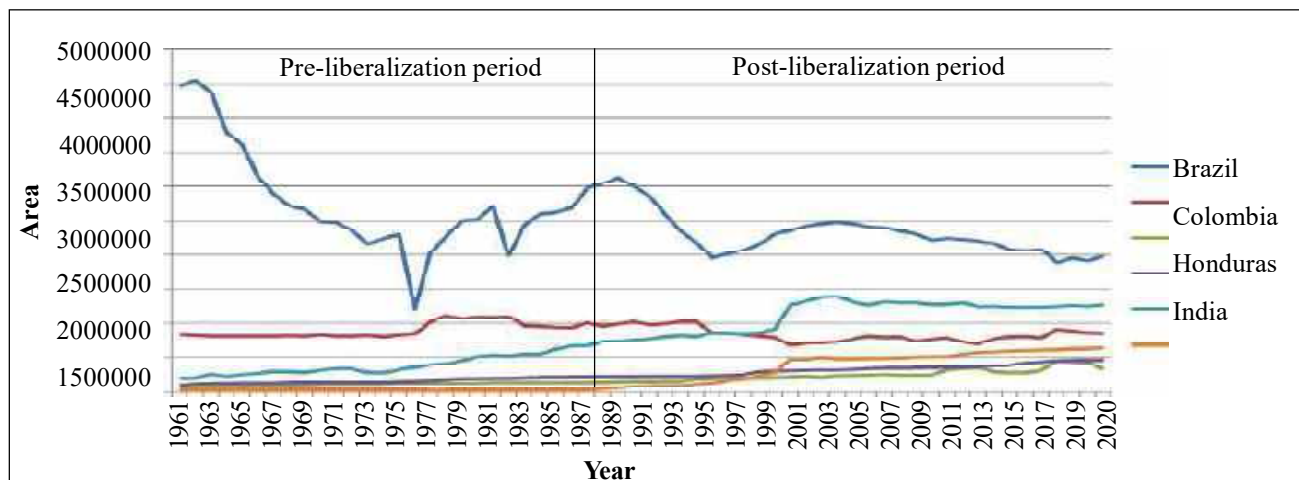


Fig. 1 : Area of green coffee in the five major coffee-producing countries in the world vis-a-vis India

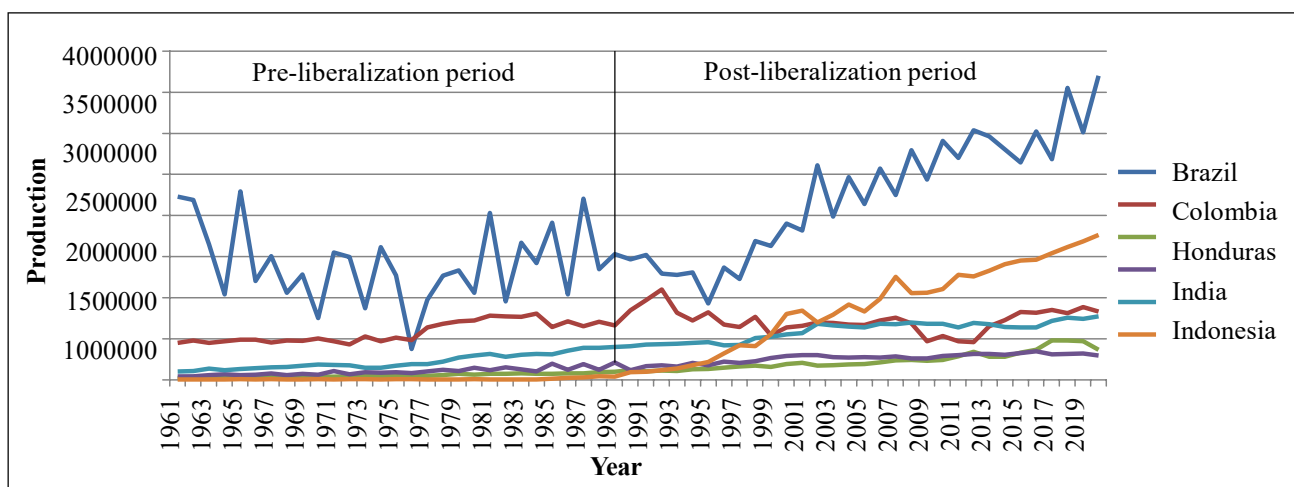


Fig. 2 : Production of green coffee in the five major coffee-producing countries in the world vis-a-vis India

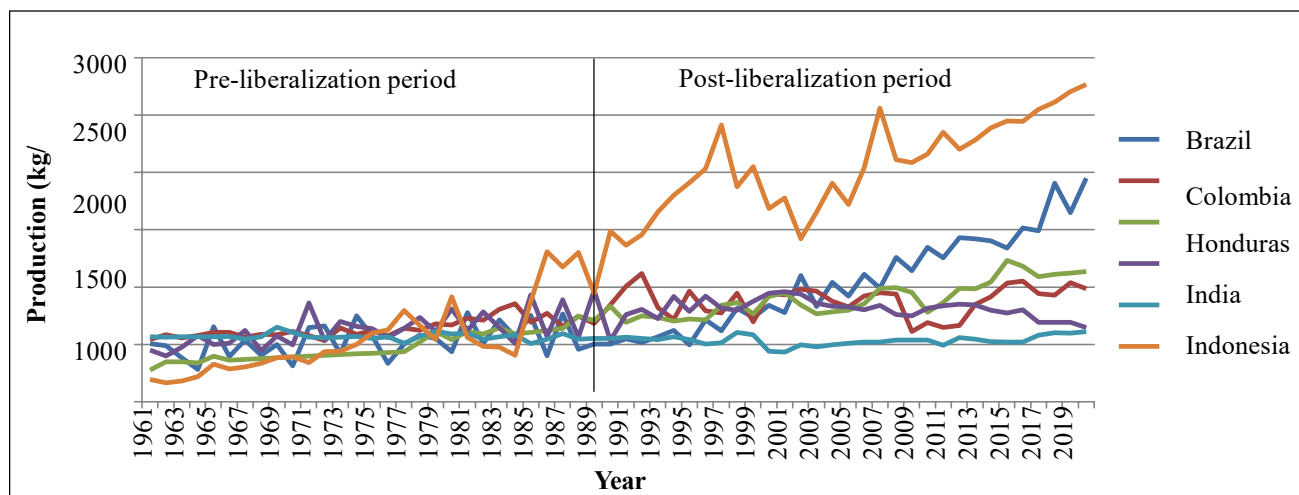


Fig. 3 : Productivity of green coffee in the five major coffee-producing countries in the world vis-a-vis India

cultivation practices over the years. The increase in productivity of Brazil was due to increased area under production. The yield of coffee in Vietnam was highest in the post-liberalization period compared to the previous period (pre-liberalization) as well as other countries because of high-intensity Robusta cultivation, planting improved tree varieties and intensive usage of fertilizers. Overall the fluctuations in productivity between the years in the countries was due to climatic factors that changed the flowering and fruit setting patter over the years as coffee being a highly climate sensitive crop.

*Compound Annual Growth Rates of Area, Production and Productivity of Coffee Across Five Countries and India* : Table 1 shows pre and post liberalization compound growth rates of area, production and productivity of coffee in five major coffee producing countries.

Brazil showed a negative significant growth rate in the area during the pre and post liberalization period. The production and productivity showed positive growth rate in both the periods but the significant growth rate was noticed in the post-liberalization period. In Columbia growth rate of area, production and productivity showed positive significant growth rate in the pre-liberalization period, whereas in the post-liberalization period negative growth rate of area and production. Overall the area under coffee in Columbia has an insignificant positive growth rate with positive significant growth rates in production and productivity. In Honduras, area, production and productivity show positive significant growth in all the periods. Indonesia shows positive significant growth rate in area and production in all the periods. The productivity in the country shows an insignificant positive growth rate. In the pre-liberalization period area under coffee in Vietnam showed negative growth rate and in post-liberalization period area showed positive significant growth rate. The production and productivity of Vietnam shows positive significant growth rate in all the periods. The area, production and productivity in India show a significant positive growth rate in all the periods.

TABLE 1  
Compound growth rate of area, production and productivity of green coffee in the five major coffee-producing countries in the world vis-a-vis India (Per cent)

Countries	Pre-liberalization (1961-1989)			Post-liberalization (1990-2020)			All period (1961-2020)		
	Area	Production	Productivity	Area	Production	Productivity	Area	Production	Productivity
Brazil	-5.71 x 10 <sup>6</sup> **	1.88 x 10 <sup>-6</sup> NS	0.02 NS	-2.22 x 10 <sup>-5</sup> ***	1.14 x 10 <sup>-5</sup> ***	0.02 ***	-6.42 x 10 <sup>-6</sup> ***	5.67 x 10 <sup>-6</sup> ***	0.01 ***
Columbia	5.75 x 10 <sup>-5</sup> ***	5.59 x 10 <sup>-5</sup> ***	0.08 ***	-4.01 x 10 <sup>-5</sup> **	-1.08 x 10 <sup>-5</sup> NS	0.005 NS	8.56 x 10 <sup>-6</sup> NS	1.49 x 10 <sup>-5</sup> **	0.01 **
Honduras	5.02 x 10 <sup>-4</sup> ***	3.79 x 10 <sup>-4</sup> ***	0.06 ***	1.02 x 10 <sup>-4</sup> ***	7.67 x 10 <sup>-5</sup> ***	0.05 ***	5.03 x 10 <sup>-5</sup> ***	4.05 x 10 <sup>-5</sup> ***	0.02 ***
Indonesia	4.87 x 10 <sup>-5</sup> ***	8.71 x 10 <sup>-5</sup> ***	-0.04 NS	3.08 x 10 <sup>-5</sup> ***	7.12 x 10 <sup>-5</sup> ***	0.04 NS	9.17 x 10 <sup>-6</sup> ***	1.99 x 10 <sup>-5</sup> ***	0.01 NS
Vietnam	-8.9 x 10 <sup>-5</sup> NS	5.17 x 10 <sup>-4</sup> ***	0.02 ***	4.18 x 10 <sup>-5</sup> ***	1.71 x 10 <sup>-5</sup> ***	0.02 ***	1.62 x 10 <sup>-5</sup> ***	7.83 x 10 <sup>-6</sup> ***	0.004 ***
India	2.10 x 10 <sup>-4</sup> ***	1.65 x 10 <sup>-4</sup> ***	0.04 ***	1.16 x 10 <sup>-4</sup> ***	1.38 x 10 <sup>-4</sup> ***	-0.02 NS	4.83 x 10 <sup>-5</sup> ***	4.65 x 10 <sup>-5</sup> ***	0.01 **

Note: \*\*, \*\*\* indicates p-value significance at 5% and 1% level of significance and NS indicates non-significance.

TABLE 2  
Instability index of area, production and productivity of green coffee in the five major coffee-producing countries in the world vis-a-vis India (Per cent)

Countries	Pre-liberalization (1961-1989)			Post-liberalization (1990-2020)			All period (1961-2020)		
	Area	Production	Productivity	Area	Production	Productivity	Area	Production	Productivity
Brazil	24.63	33.99	25.31	9.47	12.51	10.50	22.74	35.80	40.92
Columbia	8.37	12.36	8	10.64	19.92	14.98	12.49	22.57	20.72
Honduras	3.38	10.31	8.52	14.20	18.22	9.32	43.56	68.45	31.07
Indonesia	13.02	13.45	5.07	13.22	8.42	8.10	48.97	45.45	7.38
Vietnam	45.33	80.27	32.32	18.39	9.23	11.17	100	107.50	57.95
India	6.35	22.05	19.04	4.45	10.86	11.36	35.12	44.74	20.39

*Instability Analysis* : the instability index for the area, production and productivity of coffee is presented in Table 2. Brazil's instability index for area decreased from pre (24.63%) to post liberalization period (9.47%), production instability decreased from 33.99 per cent to 12.51 per cent and productivity instability also decreased from 25.31 per cent to 10.50 per cent. The decrease in the instability was due to replanting high-yielding and disease resistant varieties in Brazil. In case of Columbia the instability index lies below 15 per cent in area and production except in case of productivity the instability index was more in post-liberalization period (19.92%) than pre-liberalization period (12.36%). The instability of productivity increased due to long-term climate changes and rust disease invasions in the post-liberalization periods of Colombia. In Honduras and Indonesia the instability between the periods did not vary too much. In Vietnam the instability in area, production and productivity decreased from pre liberalization period (45.33%, 80.27% & 32.32%) to post liberalization period (18.39%, 9.32% & 11.17%). This huge difference in instability was due to drastic steps taken by the Vietnam Government like increase area under coffee cultivation with high yielding varieties and conversion of Arabica area to Robusta coffee areas to take advantage of liberalization. India's instability index for area decreased from pre (6.35%) to post-liberalization period (4.45%), production instability decreased from 22.05 per cent to 10.86 per cent and productivity decreased from 19.04 per cent to 10.86 per cent which means that coffee market in India were trying to attain stability.

*Classification of Countries Based in Instability Index Values* : In table 3 the countries were classified into low, medium and high instable countries. In the pre-liberalization period Brazil and Vietnam showed high instability in area, production and productivity, whereas Columbia, Honduras and Indonesia show less instability in area, production and productivity. The coffee harvested area in India were less unstable in the pre-liberalization period with high instability in production and medium instability in productivity. In the post-liberalization period Brazil turned out to be less unstable towards changes in area, production and productivity. Vietnam also stabilized in production and productivity but the area still remains under medium instability. Columbia and Honduras had some fluctuations in stability with regard to production but area and productivity remain under low instability category. India's area, production and productivity reach low instability category in post-liberalization period. By taking the overall instability performance from the period of 1961 to 2020 the major coffee producing countries and India still lie in the high instability zone due to climatic changes as coffee is climate sensitive crop, international negotiations and price volatility over the years. To reach the overall instability it might take few more years as we notice in the post-liberalization period countries are trying to achieve stability in area and production.

*Export Performance of Indian Coffee* : The results in the Table 4 show that trade intensity was highest with Italy followed by Russia, Belgium and Germany. There has been a constant trade intensity of India with



TABLE 3  
Classification of countries based on instability in coffee production

Periods	Particulars	Low (<15%)	Medium (15-20%)	High (>20%)
Pre-liberalization (1961-1989)	Area	Columbia, Honduras, Indonesia, India	-	Brazil, Vietnam
	Production	Columbia, Honduras, Indonesia	-	Brazil, Vietnam, India
	Productivity	Columbia, Honduras, Indonesia	India	Brazil, Vietnam
Post-liberalization (1990-2020)	Area	Brazil, Columbia, Honduras, Indonesia, India	Vietnam	-
	Production	Brazil, Indonesia, Vietnam, India	Columbia, Honduras	-
	Productivity	Brazil, Columbia, Honduras, Indonesia, Vietnam, India	-	-
All period (1961-2020)	Area	Columbia	-	Brazil, Honduras, Indonesia, Vietnam, India
	Production		-	Brazil, Columbia, Honduras, Indonesia, Vietnam, India
	Productivity	Indonesia	-	Brazil, Columbia, Honduras, Vietnam, India

TABLE 4  
Trade intensity index of coffee from India to top coffee importers in the world from 2002-2021  
(Per cent)

Year	United States of America	Germany	Italy	Belgium	France	Japan	Russia
2002	13.58	94.59	311.90	171.80	29.60	33.81	872.30
2003	6.46	79.77	372.60	201.98	36.60	30.75	697.96
2004	15.82	75.25	396.17	147.78	27.69	70.82	700.41
2005	6.73	59.08	460.69	110.19	31.43	40.49	806.58
2006	10.65	84.52	462.76	163.25	32.25	40.92	509.90
2007	12.41	44.95	459.74	148.05	34.96	20.45	372.20
2008	10.00	67.21	452.30	108.76	30.86	17.75	343.63
2009	14.91	45.22	414.24	113.43	21.61	16.55	547.56
2010	18.05	70.71	517.88	133.12	20.20	8.83	388.74
2011	10.45	103.96	419.02	186.68	20.10	9.16	381.58
2012	18.79	67.54	468.87	198.83	20.64	22.51	398.67
2013	25.24	82.76	472.01	179.63	20.04	18.71	214.72
2014	23.36	91.10	446.82	177.36	17.16	17.07	243.50
2015	19.93	79.59	453.94	170.40	20.09	10.95	326.09
2016	26.09	86.89	439.66	222.81	15.98	8.16	328.67
2017	28.25	97.18	391.88	179.80	13.12	6.50	298.47
2018	36.43	76.20	381.15	196.44	12.63	8.54	243.84
2019	30.33	105.06	389.81	190.88	13.17	7.40	295.10
2020	31.40	101.54	386.47	234.18	7.73	9.99	191.91
2021	33.36	81.37	350.90	268.58	5.94	14.85	249.57

respect to Italy, Belgium and Russia whereas with Germany the intensity had been fluctuating over the years. Therefore the major importers of Indian coffee among the major coffee importers in the world are Italy, Belgium and Russia. Italy seems to be the major exporter because of the special characteristic of Robusta coffee flavor. The Robusta coffee exported from Kerala is exposed to the salty sea air during the monsoon season which gives it a specific taste to Italian blends.

The study analyzed the trend, compound growth rates, instability in area, production and productivity of coffee in five major coffee producing countries and India and trade performance among top coffee importers in world. The area under coffee reduced over the years but production and productivity showed increasing trend over the years. Area and production in Columbia showed increasing trend in pre-liberalization period but in the post-liberalization period they show a decreasing trend. Area and production increased in Vietnam, Honduras and Indonesia over the years especially in the post-liberalization period. Area and production of coffee in India had increased over the years but in the post-liberalization period the growth rate was less compared to pre-liberalization period. If the same trend continues in India, then sooner the area and production would reduce therefore government must intervene in the matters to increase production by planting high-yielding and disease resistant varieties, giving incentives to remove old plants and plant the new ones. Special focus must be taken towards adoptive measures for resisting climate changes as coffee is a climate sensitive crop. With respect to export performance of Italy, Belgium and Russia are the major export destinations of Indian coffee. To hold the major share in the international market the export promotion activities with respect to Indian coffee must be focused.

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## Effect of Different Sources of Organic Manures and Jeevamrutha on Growth and Yield of French Bean (*Phaseolus vulgaris* L.)

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### ABSTRACT

A field experiment was conducted at research and demonstration block of Research Institute on Organic Farming, UAS, Bangalore during *kharif*- 2021 and *rabi* 2021-22 to study the combined effect of different sources of organic manures and jeevamrutha on growth and yield of french bean. The experiment was laid out in Factorial RCBD consisting of 16 treatment combinations replicated thrice. Soil of the experimental site was red sandy loam with a pH (6.93), EC (0.27 dS m<sup>-1</sup>) and medium in available nitrogen (291.5 kg ha<sup>-1</sup>), phosphorus (28.2 kg ha<sup>-1</sup>) and potassium (236.4 kg ha<sup>-1</sup>). The experimental results indicated that application of vermicompost on nitrogen (N) equivalent basis resulted in significantly higher plant height (31.79 cm), number of branches per plant (5.96), number of leaves per plant (41.19), green pod yield (150.48 q ha<sup>-1</sup>) and haulm yield (38.06 q ha<sup>-1</sup>) at harvest compared to other organic manure sources, *viz.*, poultry manure and FYM. Among levels of jeevamrutha, *viz.*, application of jeevamrutha at 2000 litre ha<sup>-1</sup> recorded significantly higher green pod yield (139.14 q ha<sup>-1</sup>) and haulm yield (35.44 q ha<sup>-1</sup>) compared to other levels of jeevamrutha, *viz.* 1500, 1000 and 0 litre ha<sup>-1</sup>.

*Keywords* : French bean, Green pod yield, Jeevamrutha, Organic manures, Vermicompost

FRENCH bean (*Phaseolus vulgaris* L.) is an important vegetable crop belonging to family Fabaceae. It is one of the most popular and widely grown vegetable crops in India. It is also known as snap bean, bush bean, kidney bean or string bean. It is consumed by almost every section of society in one or other form *i.e.*, as tender green pod or vegetable or dry beans as dal. French bean tender pod contains 1.7g protein, 0.1g fat, 4.5g carbohydrate and 1.8g fiber per 100g which makes it complete food (Tiwari and Chaubey, 2017). Green pods are an important source of vitamin A which is effective in controlling night blindness in human being (Birajdar, 2006).

Due to irrational and non-judicious use of synthetic chemical fertilizers without applying organic manures in the crop production process over the years has led

to deterioration of multi-nutrient deficiencies particularly various micronutrients *viz.* Zn, B, Mn, Fe, Mo *etc.*, which have made the soils less responsive to application of nutrients. Considering these adverse impacts on crop production along with rapid escalation of fertilizer costs, there is a paradigm shift from inorganic to organic farming. Addition of organic matter as source of nutrients is crucial to sustain soil health in long term basis and thus, organic farming plays a pivotal role in agricultural system in the country. Organic farming mainly focuses on use of on-farm organic resources to sustain soil health. Keeping all these points in consideration, the investigation was carried out at University of Agricultural Sciences, Bangalore to study the influence of organic manures and jeevamrutha on growth and yield of french bean.

## MATERIAL AND METHODS

A field experiment was carried out at research and demonstration block of Research Institute on Organic Farming (RIOF), Gandhi Krishi Vignana Kendra (GKVK), University of Agricultural Sciences, Bangalore which comes under the agroclimatic zone of Eastern dry zone of Karnataka. It is situated at a latitude of 12° 58' North, longitude of 75° 35' East and at an altitude of 930 m above MSL (mean sea level). The experiment was conducted to study the combined effect of different sources of organic manures and jeevamrutha on growth and yield of French bean during *kharif* and *rabi* seasons of 2021-22 under irrigated condition. Experiment was laid out in Randomised Complete Block Design (RCBD) with factorial concept consisting of two factors *viz.*, different organic sources ( $M_1$ : No organic manure,  $M_2$ : FYM,  $M_3$ : Vermicompost and  $M_4$ : Poultry Manure - 100% N equivalent basis) and different levels of jeevamrutha ( $J_1$ : No jeevamrutha,  $J_2$ : 1000 l ha<sup>-1</sup>,  $J_3$ : 1500 l ha<sup>-1</sup> and  $J_4$ : 2000 l ha<sup>-1</sup>) replicated thrice. Treatment combinations include  $T_1$ : Without application of manure and jeevamrutha,  $T_2$ : Application of jeevamrutha at 1000 l ha<sup>-1</sup> without manure,  $T_3$ : Application of jeevamrutha at 1500 l ha<sup>-1</sup> without manure,  $T_4$ : Application of jeevamrutha at 2000 l ha<sup>-1</sup> without manure,  $T_5$ : Application of FYM without jeevamrutha,  $T_6$ : Application of FYM coupled with jeevamrutha at 1000 l ha<sup>-1</sup>,  $T_7$ : Application of FYM coupled with jeevamrutha at 1500 l ha<sup>-1</sup>,  $T_8$ : Application of FYM coupled with jeevamrutha at 2000 l ha<sup>-1</sup>,  $T_9$ : Application of vermicompost without jeevamrutha,  $T_{10}$ : Application of vermicompost coupled with jeevamrutha at 1000 l ha<sup>-1</sup>,  $T_{11}$ : Application of vermicompost coupled with jeevamrutha at 1500 l ha<sup>-1</sup>,  $T_{12}$ : Application of vermicompost coupled with jeevamrutha at 2000 l ha<sup>-1</sup>,  $T_{13}$ : Application of poultry manure without jeevamrutha,  $T_{14}$ : Application of poultry manure coupled with jeevamrutha at 1000 l ha<sup>-1</sup>,  $T_{15}$ : Application of poultry manure coupled with jeevamrutha at 1500 l ha<sup>-1</sup> and  $T_{16}$ : Application of poultry manure coupled with jeevamrutha at 2000 l ha<sup>-1</sup>. Soils of the experimental site was red sandy loam with a pH of 6.93, EC (0.27 dS m<sup>-1</sup>), medium in

available N (291.5 kg ha<sup>-1</sup>), P<sub>2</sub>O<sub>5</sub> (28.2 kg ha<sup>-1</sup>) and K<sub>2</sub>O (236.4 kg ha<sup>-1</sup>). French bean variety Arka Suvidha was sown with a spacing of 30 cm × 15 cm and recommended agronomic practices were followed to raise the crop. Recommended dose of nutrients for french bean was 63:100:75 NPK kg ha<sup>-1</sup> and organic nutrients were supplied on the basis of nitrogen equivalent after analysing the nutrient content. Application of Farm Yard Manure (FYM) at the rate of 25 t ha<sup>-1</sup> as basal application was common for all the treatments. Organic manures were incorporated into the soil, three weeks prior to sowing. Jeevamrutha was applied to the soil by diluting with normal water at 20, 40 and 60 days after sowing (DAS), according to the treatment. Hand weeding as well as earthing up was carried out at 20 DAS to maintain weed free condition and to provide good anchorage to the crop. Other crop protection practices were followed as and when required. Biometric observations on growth and yield parameters were recorded randomly on selected five plants at 20 and 40 days after sowing (DAS) and at harvest in the net plot. Data was subjected to statistical analysis as per the procedure outlined by Gomez and Gomez (1984). To know the effect of individual factors and to compare treatment combinations with control treatments, statistical procedure of factorial randomized complete block was followed.

### Preparation of Jeevamrutha

Jeevamrutha was prepared by mixing 10 kg of cow dung, 10 litre of cow urine, 2 kg of jaggery, 2 kg of pigeon pea flour and hand full of soil collected from farm. All these were put in 200 litres plastic drum and mixed thoroughly and volume was made up to 200 litres by adding water. The mixture was stirred well in clock wise direction thrice a day plastic drum was kept shade covered with wet jute bag. Jeevamrutha was fermented for 10 days and applied to the root zone of French bean plants manually (Devakumar *et al.*, 2008 and Palekar, 2006).

## RESULTS AND DISCUSSION

### Plant Height (cm)

The data of two seasons as well as pooled data pertaining to plant height of French bean as influenced

TABLE 1  
Plant height of French bean at different growth stages as influenced by different organic manures and jeevamrutha during *kharif* 2021

Treatments	Plant height (cm)														
	20 DAS					40 DAS					At harvest				
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean
M <sub>1</sub>	9.17	9.60	10.47	10.63	9.97	23.01	23.20	23.79	24.10	23.53	25.24	25.72	25.96	26.45	25.84
M <sub>2</sub>	10.23	11.07	11.23	11.70	11.06	23.62	25.23	26.05	26.93	25.46	25.68	27.46	28.25	28.95	27.59
M <sub>3</sub>	11.23	13.07	13.57	13.73	12.90	26.00	28.12	29.19	30.80	28.53	28.08	30.36	31.32	32.92	30.67
M <sub>4</sub>	11.07	11.83	12.53	12.77	12.05	25.51	27.14	27.24	28.43	27.08	27.75	28.79	29.45	30.58	29.14
Mean	10.42	11.39	11.95	12.20		24.53	25.92	26.57	27.56		26.69	28.08	28.75	29.73	
Comparison	C.D. (P=0.05)					S.E.m±					C.D. (P=0.05)				
M	0.68					0.40					0.50				
J	0.68					0.40					0.50				
M × J	NS					0.79					1.01				

Note: M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm Yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing

TABLE 2  
Plant height of French bean at different growth stages as influenced by different organic manures and jeevamrutha during *rabi* 2021-22

Treatments	Plant height (cm)														
	20 DAS					40 DAS					At harvest				
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean
M <sub>1</sub>	9.93	10.37	10.67	10.80	10.44	23.92	24.15	24.85	25.16	24.52	27.49	27.97	28.21	28.70	28.09
M <sub>2</sub>	10.93	11.40	11.37	11.63	11.33	25.47	26.56	26.48	27.11	26.41	27.93	29.71	30.50	31.20	29.83
M <sub>3</sub>	11.57	12.57	13.17	14.00	12.83	26.95	29.28	30.68	31.84	29.69	30.33	32.61	33.57	35.17	32.92
M <sub>4</sub>	10.53	11.73	12.83	13.23	12.08	24.54	27.34	29.90	30.29	28.02	30.00	30.71	31.70	32.83	31.31
Mean	10.74	11.52	12.01	12.42		25.22	26.83	27.98	28.60		28.94	30.25	30.99	31.97	
Comparison	C.D. (P=0.05)					S.E.m±					C.D. (P=0.05)				
M	0.71					0.41					0.54				
J	0.71					0.41					0.54				
M × J	NS					0.81					1.09				

Note: M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing

**TABLE 3**  
**Plant height of French bean at different growth stages as influenced by different organic manures and jeevamrutha**  
**(Pooled data of two seasons)**

Treatments	Plant height (cm)														
	20 DAS						40 DAS						At harvest		
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean
M <sub>1</sub>	9.55	9.98	10.57	10.72	10.20	23.47	23.68	24.32	24.63	24.02	26.36	26.84	27.08	27.57	26.97
M <sub>2</sub>	10.58	11.23	11.30	11.67	11.20	24.55	25.90	26.27	27.02	25.93	26.80	28.58	29.37	30.07	28.71
M <sub>3</sub>	11.40	12.82	13.37	13.87	12.86	26.47	28.70	29.94	31.32	29.11	29.20	31.48	32.44	34.04	31.79
M <sub>4</sub>	10.80	11.78	12.68	13.00	12.07	25.03	27.24	28.57	29.36	27.55	28.87	29.75	30.57	31.70	30.23
Mean	10.58	11.45	11.98	12.31		24.88	26.38	27.27	28.08		27.81	29.17	29.87	30.85	
Comparison	C.D. (P=0.05)						C.D. (P=0.05)						C.D. (P=0.05)		
M	S.Em±						S.Em±						S.Em±		
	0.18						0.31						0.52		
J	0.18						0.31						0.52		
M × J	0.37						0.62						1.04		

Note : M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm Yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing

by different organic manure sources and levels of jeevamrutha is presented in tables 1, 2 and 3. In both the seasons, application of vermicompost recorded significantly higher plant height (12.90 and 12.83 cm at 20 DAS, 28.53 and 29.69 cm at 40 DAS and 30.67 and 32.92 cm at harvest, respectively) followed by poultry manure, farm yard manure, as compared to significantly lower plant height in without manure application (9.97 and 10.44 cm at 20 DAS, 23.53 and 24.52 cm at 40 DAS and 25.84 and 28.09 cm at harvest, respectively) was recorded. Among the levels of jeevamrutha, application rate of 2000 litre ha<sup>-1</sup> recorded higher plant height in both *kharif* and *rabi* season (12.20 and 12.42 cm at 20 DAS, 27.56 and 28.60 cm at 40 DAS and 29.73 and 31.97 cm at harvest, respectively) which was at par with application of jeevamrutha at 1500 litre ha<sup>-1</sup>. Without jeevamrutha application treatment recorded lower plant height (10.42 and 10.74 cm at 20 DAS, 24.53 and 25.22 cm at 40 DAS and 26.69 and 28.94 cm at harvest, respectively) (Table 1 and 2). The pooled data indicated that among different sources of organic manures, application of vermicompost recorded significantly higher plant height (12.86, 29.11 and 31.79 cm at 20, 40 DAS and at harvest, respectively) followed by poultry manure, farm yard manure as compared to without manure application (10.20, 24.02 and 26.97 cm at 20 and 40 DAS and at harvest, respectively). Among the levels of jeevamrutha, application rate of 2000 litre ha<sup>-1</sup> recorded higher plant height (12.31, 28.08 and 30.85 cm at 20, 40 DAS and at harvest, respectively) which was at par with application of jeevamrutha at 1500 litre ha<sup>-1</sup>. Lower plant height was observed under no jeevamrutha application (10.58, 24.88 and 27.81 cm at 20, 40 DAS and at harvest, respectively) (Table 3). Plant height at different growth stages did not differ significantly due to the interaction between various organic manures and levels of jeevamrutha application. However, higher plant height was observed with vermicompost in combination with 2000 litre ha<sup>-1</sup> jeevamrutha (13.87, 28.57 and 30.57 cm at 20, 40 DAS and at harvest, respectively) and lower plant height was observed with no manure and no jeevamrutha application (9.55, 23.47 and

26.36 cm at 20, 40 DAS and at harvest, respectively). Significantly higher plant height was reported under vermicompost applied treatment and this may be due to the fact that vermicompost contains humified organic matter characterised by high molecular weight and enzymatically active humic fraction which stimulate seed germination and plant growth. Similar result was reported by Adhikari *et al.* (2016) and Sayfalla *et al.* (2015). Jeevamrutha contains plant growth promoting substances like IAA, GA (Devakumar *et al.*, 2008 and Nileema and Sreenivasa, 2011). These might have stimulated the necessary growth and development in plants, leading to better growth of French bean. Similar results were also found by Siddappa (2015) in field bean.

### Number of Branches

The data of two seasons as well as pooled data pertaining to number of branches per plant of French bean as influenced by different organic manure sources and levels of jeevamrutha is presented in table 4, 5 and 6. During both the seasons, application of vermicompost recorded significantly higher number of branches per plant (1.37 and 1.63 at 20 DAS, 3.83 and 4.20 at 40 DAS and 5.92 and 6.67 at harvest, respectively) followed by poultry manure, farm yard manure, as compared to without manure application (1.07 and 1.07 at 20 DAS, 3.22 and 3.28 at 40 DAS and 4.95 and 5.10 at harvest, respectively). Among the levels of jeevamrutha, application rate of 2000 litre ha<sup>-1</sup> recorded significantly higher number of branches in both *kharif* and *rabi* season (3.80 and 3.88 at 40 DAS and 5.98 and 6.15 at harvest, respectively) except at 20 DAS which was statistically at par with application of jeevamrutha at 1500 litre ha<sup>-1</sup>. Lower number of branches was observed under without jeevamrutha application (1.12 and 1.12 at 20 DAS, 3.30 and 3.37 at 40 DAS and 5.10 and 5.18 at harvest, respectively) (Table 3 and 4). In pooled data of two seasons, among different sources of organic manures, application of vermicompost recorded significantly higher number of branches (1.50, 3.89 and 5.96 at 20, 40 DAS and at harvest, respectively) followed by poultry manure, farm yard manure, as compared to

TABLE 4  
Number of branches per plant of french bean at different growth stages as influenced by different organic manures and jeevamrutha during *kharif* 2021

Treatments	Number of branches per plant																	
	20 DAS						40 DAS						At harvest					
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	S.E.m±	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	S.E.m±	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	S.E.m±
M <sub>1</sub>	1.07	1.00	1.13	1.07	1.07	0.03	3.07	3.07	3.27	3.47	3.22	0.03	4.47	4.87	5.13	5.33	4.95	0.07
M <sub>2</sub>	1.07	1.00	1.13	1.20	1.10	0.03	3.27	3.47	3.67	3.80	3.55	0.03	5.20	5.27	5.67	5.93	5.52	0.07
M <sub>3</sub>	1.20	1.27	1.33	1.67	1.37	0.03	3.47	3.73	4.07	4.07	3.83	0.03	5.47	5.60	6.00	6.60	5.92	0.07
M <sub>4</sub>	1.13	1.27	1.40	1.53	1.33	0.03	3.40	3.53	3.80	3.87	3.65	0.03	5.27	5.67	5.87	6.07	5.72	0.03
Mean	1.12	1.13	1.25	1.37	1.37	0.05	3.30	3.45	3.70	3.80	3.70	0.05	5.10	5.35	5.67	5.98	5.67	0.13
Comparison	C.D. (P=0.05)						C.D. (P=0.05)						C.D. (P=0.05)					
M	0.04						0.03						0.07					
J	0.04						0.03						0.07					
M × J	0.09						0.05						0.13					

Note: M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm Yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing

TABLE 5  
Number of branches per plant of French bean at different growth stages as influenced by different organic manures and jeevamrutha during rabi 2021-22

Treatments	Number of branches per plant																	
	20 DAS						40 DAS						At harvest					
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	S.E.m±	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	S.E.m±	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	S.E.m±
M <sub>1</sub>	1.00	1.00	1.13	1.13	1.07	0.14	3.13	3.13	3.33	3.53	3.28	0.13	4.60	4.73	5.33	5.73	5.10	0.21
M <sub>2</sub>	1.00	1.13	1.47	1.40	1.25	0.14	3.33	3.53	3.80	3.87	3.63	0.13	5.27	5.47	5.87	6.07	5.67	0.21
M <sub>3</sub>	1.27	1.40	1.80	2.07	1.63	NS	3.53	3.87	4.20	4.20	3.95	NS	5.53	5.67	6.13	6.67	6.00	0.14
M <sub>4</sub>	1.20	1.20	1.47	1.60	1.37	NS	3.47	3.60	3.93	3.93	3.73	NS	5.33	5.73	5.93	6.13	5.78	0.14
Mean	1.12	1.18	1.47	1.55	1.37		3.37	3.53	3.82	3.88	3.63		5.18	5.40	5.82	6.15	5.57	
Comparison	C.D. (P=0.05)						C.D. (P=0.05)						C.D. (P=0.05)					
M	S.E.m±						S.E.m±						S.E.m±					
J	0.05						0.04						0.07					
M × J	0.10						0.09						0.14					

Note: M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm Yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing

TABLE 6  
Number of branches per plant of French bean at different growth stages as influenced by different organic manures and jeevamrutha (Pooled data of two seasons)

Treatments	Number of branches per plant																	
	20 DAS						40 DAS						At harvest					
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	S.E.m±	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	S.E.m±	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	S.E.m±
M <sub>1</sub>	1.03	1.00	1.13	1.10	1.07	0.10	3.10	3.10	3.30	3.50	3.25	0.09	4.53	4.80	5.23	5.53	5.03	0.15
M <sub>2</sub>	1.03	1.07	1.30	1.30	1.18	0.10	3.30	3.50	3.73	3.83	3.59	0.09	5.23	5.37	5.77	6.00	5.59	0.15
M <sub>3</sub>	1.23	1.33	1.57	1.87	1.50	NS	3.50	3.80	4.13	4.13	3.89	NS	5.50	5.63	6.07	6.63	5.96	0.10
M <sub>4</sub>	1.17	1.23	1.43	1.57	1.35	NS	3.43	3.57	3.87	3.90	3.69	NS	5.30	5.70	5.90	6.10	5.75	0.10
Mean	1.12	1.16	1.36	1.46	1.28		3.33	3.49	3.76	3.84	3.63		5.14	5.38	5.74	6.07	5.57	
Comparison	C.D. (P=0.05)						C.D. (P=0.05)						C.D. (P=0.05)					
M	S.E.m±						S.E.m±						S.E.m±					
J	0.03						0.03						0.05					
M × J	0.07						0.06						0.10					

Note: M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm Yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing



without manure application. Among the levels of jeevamrutha, application rate of 2000 litre ha<sup>-1</sup> recorded higher number of branches (1.46, 3.84 and 6.07 at 20, 40 DAS and at harvest, respectively) which was statistically at par with application of jeevamrutha at 1500 litre ha<sup>-1</sup> except at harvest where it differed significantly. Significantly lower number of branches were observed under without jeevamrutha application (1.12, 3.37 and 5.18 at 20, 40 DAS and at harvest, respectively) except at 20 DAS where it was at par with jeevamrutha at 1000 litre ha<sup>-1</sup> (Table 6). Number of branches at different growth stages did not differ significantly due to the interaction between various organic manures and levels of jeevamrutha application. Higher numbers of branches were observed with the application of vermicompost which might be due to the fact that vermicompost contains significant quantities of water soluble nutrients which are readily available to the crop during active growth periods (Rajini and Srivastava, 2001; Tomati *et al.*, 1983; Bano *et al.*, 1987 and Bhawalkar, 1991).

**Number of Leaves**

The data of two seasons as well as pooled data pertaining to number of leaves of French bean as influenced by different organic manure sources and levels of jeevamrutha is presented in Table 7, 8 and 9. During both the seasons, application of vermicompost recorded significantly higher number of leaves (9.83 and 10.12 at 20 DAS, 39.50 and 40.82 at 40 DAS and 43.27 and 42.06 at harvest, respectively) followed by poultry manure, farm yard manure, as compared to without manure application. Among the levels of jeevamrutha, application rate of 2000 litre ha<sup>-1</sup> recorded higher number of leaves in both *kharif* and *rabi* season (9.58 and 9.92 at 20 DAS, 37.25 and 37.52 at 40 DAS and 38.00 and 39.78 at harvest, respectively) which was statistically at par with application of jeevamrutha at 1500 litre ha<sup>-1</sup>. Lower number of leaves was observed under without jeevamrutha application (8.47 and 8.73 at 20 DAS, 33.67 and 34.92 at 40 DAS and 33.67 and 35.63 at harvest, respectively) (Table 7 and 8). In pooled data of two seasons, it was observed that among different sources of organic manures, application of

TABLE 7  
Number of leaves per plant of French bean at different growth stages as influenced by different organic manures and jeevamrutha during *kharif* 2021

Treatments	Number of leaves per plant														
	20 DAS						40 DAS						At harvest		
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean
M <sub>1</sub>	8.00	8.00	8.40	8.60	8.25	29.00	30.00	31.00	32.00	30.50	30.40	31.13	32.40	32.53	31.62
M <sub>2</sub>	8.07	8.60	9.03	9.40	8.77	34.00	35.00	36.00	36.00	35.25	31.93	35.53	37.07	37.60	35.53
M <sub>3</sub>	9.20	9.60	10.00	10.53	9.83	36.00	39.00	41.00	42.00	39.50	36.33	40.20	41.47	43.27	40.32
M <sub>4</sub>	8.60	9.20	9.60	9.80	9.30	35.67	36.00	37.00	39.00	36.92	36.00	37.27	38.40	38.60	37.57
Mean	8.47	8.85	9.26	9.58	9.30	33.67	35.00	36.25	37.25	36.25	33.67	36.03	37.33	38.00	36.00
Comparison	C.D. (P=0.05)						C.D. (P=0.05)						C.D. (P=0.05)		
M	S.Em±						S.Em±						S.Em±		
J	0.15						0.44						0.62		
M × J	0.15						0.44						0.62		
	0.30						NS						1.25		
	NS						NS						NS		

Note : M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm Yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing

TABLE 8  
Number of leaves per plant of french bean at different growth stages as influenced by different organic manures and jeevamrutha during rabi 2021-22

Treatments	Number of leaves per plant															
	20 DAS						40 DAS									
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	
M <sub>1</sub>	8.13	8.13	8.60	8.80	8.42	32.00	32.13	33.00	33.13	32.57	32.47	32.93	33.60	34.20	33.30	
M <sub>2</sub>	8.60	8.97	9.07	9.60	9.06	33.27	33.40	34.00	36.13	34.20	33.07	37.53	38.13	39.13	36.97	
M <sub>3</sub>	9.40	9.80	10.20	11.07	10.12	39.13	40.00	41.67	42.47	40.82	39.27	41.33	42.70	44.93	42.06	
M <sub>4</sub>	8.80	9.40	9.87	10.20	9.57	35.27	33.33	37.07	38.33	36.00	37.73	38.13	39.20	40.83	38.98	
Mean	8.73	9.08	9.43	9.92	9.42	34.92	34.72	36.43	37.52	36.43	35.63	37.48	38.41	39.78	37.48	
Comparison	C.D. (P=0.05)						C.D. (P=0.05)						C.D. (P=0.05)			
M	S.Em±						S.Em±						S.Em±			
J	0.16						0.46						0.64			
M × J	0.32						NS						1.28			

Note : M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm Yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing

TABLE 9  
Number of leaves per plant of French bean at different growth stages as influenced by different organic manures and jeevamrutha (Pooled data of two seasons)

Treatments	Number of leaves per plant															
	20 DAS						40 DAS									
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	
M <sub>1</sub>	8.07	8.07	8.50	8.70	8.33	30.50	31.07	32.00	32.57	31.53	31.43	32.03	33.00	33.37	32.46	
M <sub>2</sub>	8.33	8.78	9.05	9.50	8.92	33.63	34.20	35.00	36.07	34.73	32.50	36.53	37.60	38.37	36.25	
M <sub>3</sub>	9.30	9.70	10.10	10.80	9.98	37.57	39.50	41.33	42.23	40.16	37.80	40.77	42.08	44.10	41.19	
M <sub>4</sub>	8.70	9.30	9.73	10.00	9.43	35.47	34.67	37.03	38.67	36.46	36.87	37.70	38.80	39.72	38.27	
Mean	8.60	8.96	9.35	9.75	9.43	34.29	34.86	36.34	37.38	36.46	34.65	36.76	37.87	38.89	37.48	
Comparison	C.D. (P=0.05)						C.D. (P=0.05)						C.D. (P=0.05)			
M	S.Em±						S.Em±						S.Em±			
J	0.14						0.30						0.62			
M × J	0.28						NS						1.24			

Note: M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm Yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing

vermicompost recorded significantly higher number of leaves (9.98, 40.16 and 41.19 at 20, 40 DAS and at harvest, respectively) followed by poultry manure, farm yard manure, as compared to without manure application. Among the levels of jeevamrutha, application rate of 2000 litre ha<sup>-1</sup> recorded higher number of leaves (9.75, 37.38 and 38.89 at 20, 40 DAS and at harvest, respectively) which were statistically at par with application of jeevamrutha at 1500 litre ha<sup>-1</sup>. Lower number of leaves were observed under without jeevamrutha application (8.60, 34.29 and 34.65 at 20, 40 DAS and at harvest, respectively) (Table 9). Number of leaves at different growth stages did not differ significantly due to the interaction between various organic manures and levels of jeevamrutha application. However, higher number of leaves were observed with vermicompost in combination with 2000 litre ha<sup>-1</sup> jeevamrutha (2.07, 4.20 and 6.67 at 20, 40 DAS and at harvest, respectively) and lower number of leaves were observed with no manure and no jeevamrutha application (1.00, 3.13 and 4.60 at 20, 40 DAS and at harvest, respectively). Higher number of leaves were observed with vermicompost application which might be due to the fact that it has hormones like activity and this induces greater root initiation, increased root biomass, enhanced plant growth and development and alters the morphology of plants (Pant *et al.*, 2009 and Singh *et al.*, 2008).

Significant difference in above growth parameters *viz.*, plant height, number of branches per plant and number of leaves per plant was noticed due to the application of vermicompost and this might be due to the fact that vermicompost contains significant amount of water-soluble nutrients which are readily available to the crop during active growth periods. A large group of beneficial microbial population like bacteria, protozoa, nematodes, fungi, actinomycetes are present in vermicompost. It is stabilized by the mutual interaction between earthworms and microorganisms (Rajini and Srivastava 2001). Biologically active metabolites, particularly gibberellins, cytokinins, auxins and group B vitamins are present in vermicompost which can be applied alone or in

combination with organic or inorganic fertilizers, so as to get better yield and quality of diverse crops (Tomati *et al.*, 1983, Bano *et al.*, 1987 and Bhawalkar, 1991). It was also found that the release of NO<sub>3</sub><sup>-</sup> and NH<sub>4</sub><sup>+</sup> form of nitrogen was higher with the application of vermicompost as compared to other organic fertilizer like FYM and poultry manure. Vermicompost is reported to have hormones like activity and this induces greater root initiation, increased root biomass, enhanced plant growth and development and alters the morphology of plants (Pant *et al.*, 2009).

### Green Pod and Haulm Yield (q ha<sup>-1</sup>)

#### Green Pod Yield (q ha<sup>-1</sup>)

The data of two seasons as well as pooled data pertaining to green pod yield of French bean as influenced by different organic manure sources and levels of jeevamrutha is presented in table 10. During both the seasons, application of vermicompost recorded significantly higher green pod yield (141.27 and 159.68 q ha<sup>-1</sup> in *kharif* and *rabi*) followed by poultry manure, farm yard manure as compared to without manure application (93.68 and 103.76 q ha<sup>-1</sup> in *kharif* and *rabi*). Among the levels of jeevamrutha, application at 2000 litre ha<sup>-1</sup> recorded significantly higher green pod yield in both *kharif* and *rabi* seasons (133.53 and 144.76 q ha<sup>-1</sup>) followed by jeevamrutha at 1500 litre ha<sup>-1</sup> followed by jeevamrutha at 1000 litre ha<sup>-1</sup> as compared to without jeevamrutha application (99.07 and 118.27 q ha<sup>-1</sup>). The pooled data of two seasons, it was observed that among different sources of organic manures, application of vermicompost recorded significantly higher green pod yield (150.48 q ha<sup>-1</sup>) followed by poultry manure (136.82 q ha<sup>-1</sup>), farm yard manure (114.69 q ha<sup>-1</sup>), as compared to without manure application (98.72 q ha<sup>-1</sup>). Among the levels of jeevamrutha, application rate of 2000 litre ha<sup>-1</sup> recorded significantly higher green pod yield (139.14 q ha<sup>-1</sup>) followed by jeevamrutha at 1500 litre ha<sup>-1</sup> (131.18 q ha<sup>-1</sup>) followed by jeevamrutha at 1000 litre ha<sup>-1</sup> (121.71 q ha<sup>-1</sup>) as compared to without jeevamrutha application (108.67 q ha<sup>-1</sup>). Green pod yield did not differ significantly

TABLE 10

Green pod yield of French bean at different growth stages as influenced by different organic manures and jeevamrutha during *kharif* 2021, *rabi* 2021-22 and pooled data of two seasons

Treatments	Green pod yield (q ha <sup>-1</sup> )														
	Kharif 2021				Rabi 2021-22				Pooled						
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean
M <sub>1</sub>	84.65	89.65	97.98	102.43	93.68	94.15	100.22	108.58	112.08	103.76	89.40	94.94	103.28	107.26	98.72
M <sub>2</sub>	93.56	103.75	115.56	124.48	109.34	104.31	114.22	126.16	135.48	120.04	98.94	108.99	120.86	129.98	114.69
M <sub>3</sub>	115.40	138.68	150.56	160.45	141.27	143.20	159.29	163.31	172.93	159.68	129.30	148.99	156.94	166.69	150.48
M <sub>4</sub>	102.65	125.75	137.43	146.75	128.15	131.41	142.11	149.88	158.54	145.49	117.03	133.93	143.66	152.65	136.82
Mean	99.07	114.46	125.38	133.53		118.27	128.96	136.98	144.76		108.67	121.71	131.18	139.14	
Comparison	C.D. (P=0.05)				C.D. (P=0.05)				C.D. (P=0.05)						
M	S.Em±				S.Em±				S.Em±						
J	2.47				2.65				2.53						
M × J	2.47				2.65				2.53						
	4.95				5.31				5.06						
	NS				NS				NS						
	7.14				7.66				7.31						
	7.14				7.66				7.31						
	NS				NS				NS						

Note : M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm Yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing

due to the interaction between various organic manures and levels of jeevamrutha application. However, higher green pod yield was observed with vermicompost in combination with 2000 litre ha<sup>-1</sup> jeevamrutha (166.69 q ha<sup>-1</sup>) and lower yield was observed with no manure and no jeevamrutha application (89.40 q ha<sup>-1</sup>).

### Haulm Yield (q ha<sup>-1</sup>)

The data of two seasons as well as pooled data pertaining to haulm yield of french bean as influenced by different organic manure sources and levels of jeevamrutha is presented in table 11. During both the seasons, application of vermicompost recorded significantly higher haulm yield (35.57 and 40.55 q ha<sup>-1</sup>, respectively) followed by poultry manure, farm yard manure, as compared to without manure application (23.42 and 25.94 q ha<sup>-1</sup>, respectively).

Among the levels of jeevamrutha, application rate of 2000 litre ha<sup>-1</sup> recorded significantly higher haulm yield in both *kharif* and *rabi* season (33.47 and 41.05 q ha<sup>-1</sup>, respectively) followed by jeevamrutha at 1500 litre ha<sup>-1</sup> followed by jeevamrutha at 1000 litre ha<sup>-1</sup> as compared to without jeevamrutha application (24.85 and 29.57 q ha<sup>-1</sup>, respectively). In pooled data of two seasons, it was observed that among different sources of organic manures, application of vermicompost recorded significantly higher haulm yield (38.06 q ha<sup>-1</sup>) followed by poultry manure, farm yard manure as compared to without manure application (24.68 q ha<sup>-1</sup>). Among the levels of jeevamrutha, application rate of 2000 litre ha<sup>-1</sup> recorded significantly higher green haulm yield (35.44 q ha<sup>-1</sup>) followed by jeevamrutha at 1500 litre ha<sup>-1</sup> followed by jeevamrutha at 1000 litre ha<sup>-1</sup> as compared to without jeevamrutha application (27.21 q ha<sup>-1</sup>). Green pod yield did not differ significantly due to the interaction between various organic manures and levels of jeevamrutha application. However, higher green pod yield was observed with vermicompost in combination with 2000 litre ha<sup>-1</sup> jeevamrutha (42.42 q ha<sup>-1</sup>) and lower yield was observed with no manure and no jeevamrutha application (22.02 q ha<sup>-1</sup>).

TABLE 11  
Haulm yield of French bean at different growth stages as influenced by different organic manures and jeevamrutha during *khariif* 2021, *rabi* 2021-22 and pooled data of two seasons

Treatments	Haulm yield (q ha <sup>-1</sup> )														
	Khariif 2021					Rabi 2021-22					Pooled				
	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean	J <sub>1</sub>	J <sub>2</sub>	J <sub>3</sub>	J <sub>4</sub>	Mean
M <sub>1</sub>	20.50	22.75	24.83	25.61	23.42	23.54	25.06	27.15	28.02	25.94	22.02	23.90	25.99	26.81	24.68
M <sub>2</sub>	23.72	26.27	29.22	31.45	27.67	26.08	28.56	31.54	35.87	30.51	24.90	27.41	30.38	33.66	29.09
M <sub>3</sub>	29.18	35.00	37.97	40.11	35.57	35.80	39.82	41.83	44.73	40.55	32.49	37.41	39.90	42.42	38.06
M <sub>4</sub>	26.00	31.77	34.69	36.69	32.29	32.85	35.53	38.80	41.05	37.06	29.42	33.65	36.75	38.87	34.67
Mean	24.85	28.95	31.68	33.47	24.85	29.57	32.24	34.83	37.42	32.42	27.21	30.59	33.25	35.44	30.59
Comparison	C.D. (P=0.05)					S.E.m±					C.D. (P=0.05)				
M	1.94					0.85					0.67				
J	1.94					0.85					0.67				
M × J	NS					1.71					1.34				

Note : M<sub>1</sub> : No organic manure, M<sub>2</sub> : Farm Yard Manure, M<sub>3</sub> : Vermicompost, M<sub>4</sub> : Poultry manure, J<sub>1</sub> : No jeevamrutha, J<sub>2</sub> : Jeevamrutha 1000 l ha<sup>-1</sup>, J<sub>3</sub> : Jeevamrutha 1500 l ha<sup>-1</sup>, J<sub>4</sub> : Jeevamrutha 2000 l ha<sup>-1</sup>, DAS- Days After Sowing

Significant higher pod and haulm yield of frenchbean (150.48 and 38.06 q ha<sup>-1</sup>, respectively) was observed with application of vermicompost which might be due to higher and faster release of NO<sub>3</sub><sup>-</sup> and NH<sub>4</sub><sup>+</sup> form of nitrogen in vermicompost as compared to other organic manures like FYM and poultry manure and this might be due to the narrow C : N ratio of vermicompost (Velmurugan and Swarnam 2013). This might have facilitated in release of plant nutrients to labile nutrient pool thereby more availability of nutrients to plants (Eswaran and Mariselvi 2016) which resulted in higher plant growth parameters viz. plant height, number of branches and number of leaves ultimately increasing both pod and haulm yield. Significantly lower pod and haulm yield was observed in case of no manure and no jeevamrutha treatment (98.72 and 24.68 q ha<sup>-1</sup>) and this might be due to insufficient nutrient availability to the crop for its proper growth and development. Similar result was also reported by Ananda and Sharanappa (2017). Significantly higher number of growth components and yield components in jeevamrutha was due to higher amount of nutrient content like nitrogen, phosphorus and potassium (1.96 %, 0.280 % and 0.173 %, respectively) and also contains Mg (46 ppm) and Cu (51 ppm) and maximum microbial population (maximum CFU of bacteria (855), fungi (29), actinomycetes (8), N-fixers (69) and P-solubilizer (80) was observed in jeevamrutha (Devakumar *et al.* 2008 and 2014). This might have enhanced the decomposition process in the soil which might have resulted in relatively quick release of nutrients from compost compared to the treatments where no jeevamrutha was applied. These results are in consonance with findings of Basavaraj Kumbar (2016) in French bean, Basavaraj Kumbar and Devakumar (2016a). Higher application rate of jeevamrutha hastened the decomposition process and increased the availability of mineralized nutrients to the plant which resulted in increased yield of frenchbean crop. This result is in accordance with the findings of Basavaraj Kumbar and Devakumar (2016b).

From this study it can be concluded that application of vermicompost along with 2000 litre ha<sup>-1</sup> jeevamrutha is beneficial in improving growth

and yield of frenchbean by providing better availability of nutrients, improved microbial activity and availability of growth promoting substances.

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## Assessment of Polycross Hybrids of Mulberry for Fruit and Seed Traits

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### ABSTRACT

Mulberry is a fast growing woody perennial tree that maintains high heterozygosity due to the out-breeding reproductive system. The success of breeding programme mainly depends on availability of genetic variability and selection of superior progenies. Polyclonal seed orchard is one of the sources for creating huge desirable genetic variability using natural hybridization with minimum effort for improvement of leaf yield and quality in mulberry. High bush polyclonal seed orchard was established using twenty nine parents (16 female & 13 male) at CSR&TI Berhampore. Morphological assessment of hybrid fruits and seeds were performed for two successive years (2020 & 2021). The results showed that significant differences among polycross hybrids in most of the fruit & seed traits. Fruit weight varied from 0.63g (T-13) to 2.27g (Berhampore-A); fruit length varied from 1.58cm (T-13) to 2.65cm (CSRS-1); fruit width varied from 0.86cm (Matigera-black) to 1.32cm (Berhampore-A); seeds per fruit varied from 25 (C-2038) to 47 (Berhampore-A) and test weight varied from 1.25g (China-white) to 2.20g (C-2045). The highest seed set was recorded in C-2045 (91.95%) followed by Sujanpur-5 (88.70%). Highly significant positive correlation was observed between fruit weight & fruit width (0.87); fruit weight & fruit length (0.84); achenes per fruit & seeds per fruit (0.72). A significant difference was recorded for seed germination among 16 polycross & it varied from 70 per cent (China-white & S-30) to 99 per cent (C-2038). After 60 days, germination was reduced more in Sujanpur-5 (92.5%) and Bogura-4 (90.40%). The information may be useful for the breeders to identify superior seedlings and creating heterozygous hybrid progenies in mulberry.

**Keywords :** Polycross hybrid of mulberry, Polyclonal seed orchard, Seed germination, Seed set, Fruits traits, Seed traits

SERICULTURE is an agro based industry and the final product of this industry is silk. The improved mulberry varieties play an important role in enhancing silk productivity and profitability of sericulture. Due to new silkworm hybrids, more number of silkworm crops per year & demanding high quality silk, superior mulberry varieties have been gaining importance. Hence, creation of desirable variation is indispensable. There are different methods to create genetic variability, bi-parental crossings or traditional breeding is most common where few parents with desired traits are considered.

But, this process is time consuming & laborious. In mass or open pollinated hybrids, pollen source is unknown thus undesirable traits may occur in the OPH varieties as well as general combining ability of a variety cannot be estimated. To overcome such limitation, polycross breeding is being practiced since long time especially for obligate cross pollinators in particular those that can be propagated vegetatively (Frandsen and Frandsen, 1948). A polycross is a mating arrangement for inter pollinating a group of cultivars or clones using natural hybridization in an isolated crossing block (Klein



*et al.*, 1973). Progeny from each entry have a common parent in a polycross design. Thus, half sib families are generated & these are frequently used for evaluating general combining ability. Purpose of polycross breeding is to provide equal opportunities for each entry to be crossed with other entry.

Mulberry is an extremely versatile plant that can fulfill a number of roles in small holding agricultural production. The mulberry (*Morus* sp.) leaf is the sole food source for the mulberry silkworm (*Bombyx mori* L.) and contributes 38.20 per cent towards the success of a cocoon crop (Wani *et al.*, 2018). Mulberry can be propagated through various methods *viz.*, seeds, cuttings, layering, grafting and tissue culture, etc. Mulberry can be easily propagated through cuttings, however has limitations which includes lack of variations resulting in reduced adaptability of daughter plants, restriction of raising only region specific plant varieties, reduced vigour by subsequent generations and lack of tap root system in vegetatively propagated plants with least robustness in them (Leaky, 2014). In recent years, the importance on mulberry fruits/ seeds has increased together with understanding of their nutritional capacities and breeding new potential cultivars for improving fruit yield [Ozgen *et al.*, 2009].

The sexual reproduction of mulberry through seeds is essential to generate and maintain genetic variability. The morpho-physiological traits are prominent, because they can be rapidly determined and have an important predictive value concerning plant adaptation (Sanchez *et al.*, 2015). In this sense, the internal structure of seeds, particularly embryo morphology, is a valuable piece of information for the classification of seed dormancy (Baskin and Baskin, 2007; 2014). The seed size and mass are vitally important traits in the life cycle of a plant, because they have implications in the dispersal, establishment and survival mechanisms of the species. In addition, the hydration degree of seed plays a fundamental role in their longevity and germination performance (Jimenez-Alfaro *et al.*, 2016). However, other seed characteristics could also show the

responses of the species to the environment; for example, the physical defense structures (testa/endocarp) and the nutrient content in the seed reserves-embryo/ endosperm (Daws *et al.*, 2006; Montejo *et al.*, 2015). Therefore, the objective of this study was to characterize the different morpho-physiological traits of the fresh fruits and seeds of sixteen polycross hybrid progenies.

## MATERIAL AND METHODS

The field experiment was conducted during 2018 to 2021 at Central Sericultural Research and Training Institute, Berhampore (West Bengal). Twenty nine parents (16 female and 13 male parents) were selected for establishment of polyclonal seed orchard based on the available pedigree records on flowering synchronization, sex expression, ploidy level, *per se* performance on leaf quality & productivity parameters, genetic diversity and combining ability studies (Table 1). The land for the establishment of seed orchard was selected far away from general mulberry plantation and surrounded by S-1635 plantation which is triploid variety bears no fertile pollen. Saplings were raised from mature cuttings of selected parents. After 10 months, the saplings were transplanted at a spacing of 5'×5' in such a way that each male parent is surrounded by different female parents. In this manner, all males and female parents were repeated many times to ensure sufficient plants to generate sufficient quantity of hybrid seeds. The plants were trained as high bush plantation and pruning was carried out once a year during July-August (Fig.1). Recommended package of practices for mulberry cultivation under irrigated condition was followed to raise healthy plantation (Ray *et al.*, 1973).

### Collection of Hybrid Fruits and Extraction of Seeds :

After natural pollination during flowering season in 2020 & 2021, polyclonal hybrid (PCH) fruits were harvested from individual female parents of seed orchard. Fruits of each mulberry genotypes were soaked in water separately for 48 hours and seeds were extracted by water soaking method (Barbour *et al.*, 2008). Harvested seeds were air dried in a

TABLE 1  
Parents utilized to establish Polyclonal  
Seed Orchard

Parents	Acc No. / Parentage
<i>Female Parents</i>	
Kosen	ME-0066
China White	ME-0042
C-2038	CF1(10) × C-763
C-2045	MHP × CF1(13)
C-2036	MHP × CF1(23)
C-2060	KOP × V1
S-30	MI-0046
Kajli OP	OPH of Kajli
Phillipines	ME-0011
CSRS-1	MI-0084
Berhampore-A	MI-0054
Sujanpur-5	MI-0017
M. Black	MI-0300
Bogura-4	ME-0097
T-13	Elite clone
Kajli	MI-0068
<i>Male Parents</i>	
MS-1	MI - 0054
MS-7	MI - 0001
Mandalaya	ME-0043
Sultanpur-5	MI-0098
Bisanpur-10	MI - 0092
White Badan	MI - 0300
Almora	MI-0141
Molai	ME -0003
Monali	MI-0131
Jodhapur	MI - 0248
<i>M. multicaulis</i>	ME -0006
C-776	<i>M. nigra</i> × <i>M. multicaulis</i>
CT-44	<i>M.indica</i> HP × CF <sub>1</sub> 12

room for two days and stored in desiccators filled with silica gel for maintaining viability for longer time.

*Characterization of Hybrids Fruits and Seeds* : Average performance (Two consecutive years) of fruit length, fruit width, fruit weight, No. of seeds/fruit, No. of achenes/fruit, test weight (weight of 1000 seeds) and seed viability & germination was recorded in PCHs of all the crosses following the standard

procedures (Dwivedi, 1990). Seed viability and germination of hybrids were recorded from one hundred seeds in four replications. Seed viability was measured by Tetrazolium method (Dandin, *et al.*, 1991). Water soaked seeds were washed and cut in to two equal halves then were dipped in 1 per cent 2,3,5-triphenyl tetrazolium chloride solution for 24 hours. Pink colour seeds out of total seeds tested were used to calculate seed viability percentage. Top paper method was used for measuring seed germination (%) using wet 3 layers of blotting paper in petriplates (Rao *et al.*, 2006). Seed viability and germination was observed after 0, 30 and 60 days after seed extraction.

## RESULTS AND DISCUSSION

The genetic improvement of mulberry depends on the availability of genetic variability and selection of suitable genotypes from breeding population. Mulberry is highly heterozygous species and success of breeding programme largely depends on generation of larger amount of desirable variability and selection of superior progenies. The greater amount of variability provides a scope for selecting desirable recombinant for more number of traits. Polycross mating design has been served as a source of enormous desirable genetic variability for improvement of yield and quality in mulberry. Twenty nine parents were utilized to establish a polyclonal seed orchard during 2018-19 (Table 1 & Plate 1). The seed orchard was trained to high bush plantation with recommended cultural practices. The established seed orchard was isolated from other genetic sources ensured production of true hybrids progenies. Polycross hybrid fruits from each parent were collected during March-April months after natural pollination during the year 2020 & 2021. Further, polycross fruits & seeds from 16 parents were characterized and the results revealed presence of enormous genetic variations for characteristics studied.

Morphological characters of PCH fruits & seeds were recorded for two consecutive years (2020 & 2021). Among 16 polycrosses, Berhampore-A cross



Plate 1 : Established polyclonal seed orchard bearing flowers, fruits and seeds

TABLE 2  
Pooled average performance of fruit and seed parameters of Polycross hybrids

Polycross progeny derived from	Fruit length (cm)	Fruit width (cm)	Fruit Weight (g)	Seeds / fruits (No.)	Test Weight (g)	Achenes/ fruit (No.)	Seed set (%)
Kosen	1.59	0.95	0.83	35.80	1.39	47.60	75.21
China White	2.10	1.06	1.26	40.30	1.25	57.60	69.97
C-2038	2.61	1.05	1.43	25.00	1.76	40.00	62.50
C-2045	2.28	1.17	1.52	40.00	2.20	43.50	91.95
C-2036	2.08	1.09	1.78	44.25	1.98	54.00	81.94
C-2060	2.45	0.95	1.67	29.00	1.79	48.60	59.67
S-30	2.35	1.05	1.39	43.20	1.38	57.00	75.79
Kajli OP	2.36	1.25	2.18	41.00	1.32	56.00	73.21
Phillipines	1.69	0.98	0.79	28.00	1.87	35.60	78.65
CSRS-1	2.65	1.11	1.98	39.80	1.56	68.40	58.19
Berhampore-A	2.56	1.32	2.27	47.00	1.43	62.20	75.56
Sujanpur-5	2.19	0.94	0.94	42.40	1.42	47.80	88.70
Matigera Black	1.66	0.86	0.74	37.50	1.55	46.20	81.17
Bogura-4	1.69	0.99	0.78	34.00	1.83	48.60	69.96
T-13	1.58	0.88	0.63	31.25	1.36	40.30	77.54
Kajli	1.93	0.87	0.88	32.20	1.46	46.20	69.70
CD (P 0.05)	0.20	0.07	0.29	3.42	0.15	4.63	5.02



Plate 2 : Morphological variability observed among the seeds of different polycross hybrids

recorded highest fruit weight followed by Kajli OP & CSRS-1. Higher fruit weight and yield among the mulberry genotypes was also reported by Ercisli 2004; Orhan & Ercisli, 2010. Seed size is an important physical indicator of seed quality that affects vegetative growth. The polycross C-2045, C-2036 & Phillipines had higher test weight and significant genetic variation among the varieties studied. The presence of large genetic variation for seed size and higher performance in seedlings derived from larger seed was reported by Ambika *et al.*, 2014. Number of seeds per fruit is important seed yield parameters and it was highest in Berhampore-A followed by C-2036 & S-30. Seed

set percentage varied among polycrosses studied and C-2045, Sujanpur-5 & C-2036 recorded more than 80 per cent seed set (Table 2 & Plate 2).

Higher seed viability was noticed among 16 polycrosses on the day of extraction. Higher viability was maintained in seeds preserved under desiccators even after two months after extraction (Plate 3). The seed germination percentage varied from 70 per cent (China white & S-30) to 99 per cent (C-2038) on the day of extraction among the 16 polycrosses studied. After 60 days, germination percentage of polycross seeds varied between 6 per cent (Sujanpur-5) to 71 per cent (Berhampore-

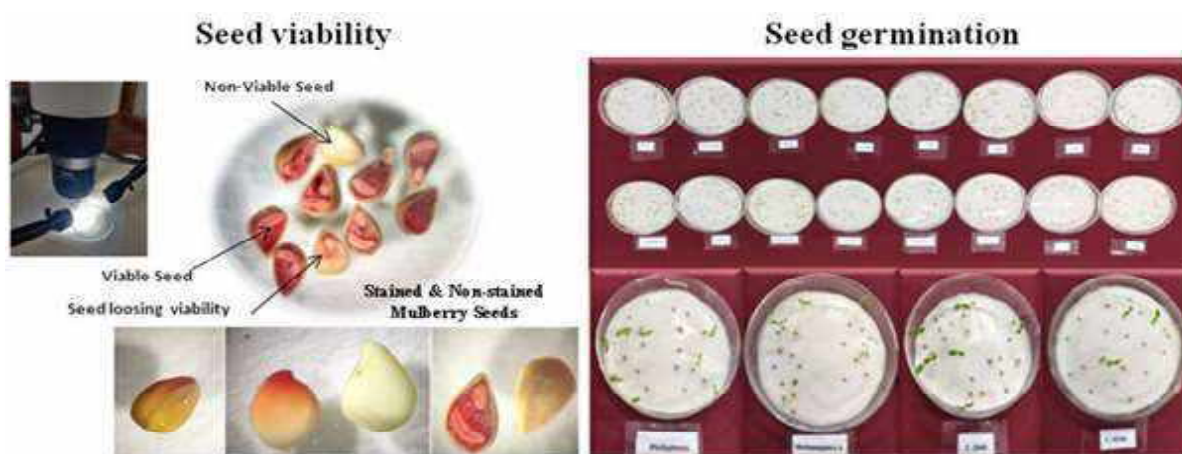


Plate 3 : Viability and germination of seeds of polycross hybrid progenie

TABLE 3  
Seed viability and germination in polycross hybrids

Polycross progeny derived from	Seed Viability (%)			Seed Germination (%)		
	0 days	30 days	60 days	0 days	30 days	60 days
Kosen	100	95	98	85	60	22
China White	99	95	96	70	48	9
C-2038	99	99	97	99	73	16
C-2045	98	94	95	80	51	11
C-2036	99	96	96	80	49	13
C-2060	100	99	100	90	74	40
S-30	99	93	94	70	49	16
Kajili OP	98	96	96	91	61	31
Phillipines	98	94	93	94	80	55
CSRS-1	98	97	98	81	70	18
Berhampore-A	100	97	97	96	80	71
Sujanpur-5	99	98	99	80	50	6
Matigera Black	99	97	97	92	70	34
Bogura-4	99	95	96	83	56	8
T-13	99	98	99	91	53	13
Kajili	97	94	97	89	78	61
CD (P 0.05)	0.44	1.00	0.98	6.75	13.21	10.74
CV (%)	1.91	2.92	2.86	5.53	14.79	28.41

A) indicating decrease in germination with increase in storage period (Table 3). Similar results were reported in mulberry by Gunduz *et al.*, 2019 (20% - 30% in *Morus bombycis*) and Song *et al.*, 2016 (9.3% - 66.5% in different mulberry species).

Reduction in seed germination was calculated after 30 as well as 60 days after extraction. Highest reduction in seed germination was observed in Sujanpur-5 (92.5%) followed by Bogura-4 (90.40) & China white (87.1%) at 60 days. However, at 30

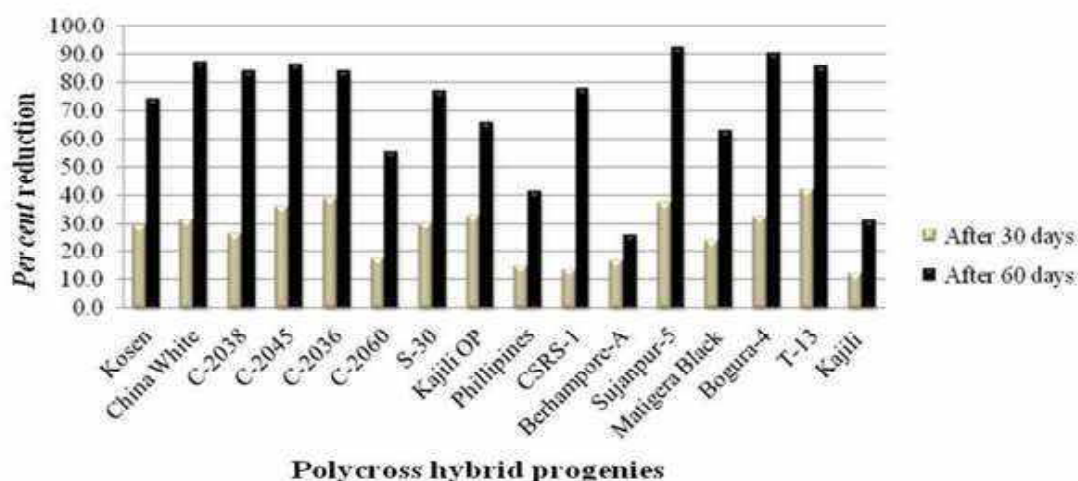


Fig.1 : Per cent reduction of seed germination in polycross hybrid progenies during storage

TABLE 4  
Association of fruit and seed characters of mulberry

Parameters	Fruit length (cm)	Fruit width (cm)	Fruit weight (cm)	Seeds/ fruit (No)	Test weight (g)	Achenes/ fruit (No)	Seed Set (%)
Fruit length (cm)	1						
Fruit width (cm)	0.66 *	1					
Fruit weight (g)	0.84 *	0.87 *	1				
Seeds/ fruit (No)	0.27	0.56 *	0.49	1			
Test weight (g)	0.05	0.09	0.05	-0.23	1		
Achenes/ fruit (No)	0.55 *	0.57 *	0.69 *	0.72 *	-0.37	1	
Seed Set (%)	-0.36	0.01	-0.25	0.42	0.20	-0.31	1

\* Significant at the 0.05 level

days after seed extraction reduction percentage ranged from 12.4 per cent to 41.8 per cent indicating, that mulberry seeds should be sown within a month of harvesting (Fig.1).

In any breeding program of complex characters such as yield for which direct selection is not effective, it become essential to measure the contribution of each of the component variable. The association between two characters that can be directly observed is the correlation of phenotypic value or the phenotypic correlation (Falconer & Mackay, 1996). A significant positive phenotypic association was observed between fruit length and fruit width, fruit weight & achenes per fruit. Similarly, fruit width had significantly positive correlation with fruit weight, seeds per fruit and achenes per fruit. In addition, seed set was found to be in positive association with seeds per fruit & test weight and negative association with fruit length, fruit weight & achenes per fruit (Table 4), which is supported by the findings of earlier workers (Dandin *et al.*, 1987 and Vijayan *et al.*, 1997).

Polycross hybrid progenies were developed using twenty nine genetically diverse parents. Morphological characterization of polycross hybrids revealed huge genetic variability present among sixteen half sib families. Seed viability & germination of different polycross hybrids had a wide range and gradually reduced with the increase in storage period

*i.e.* after two months of seed extraction. Some of the characteristics of fruits & seeds showed positive association among themselves. The information with respect to fruit and seed traits will support the breeders for intra and inter specific hybridization with suitable parents for developing large number of progenies with desired economic characters to select the best one and in turn benefit the sericulture farming community and the industry as a whole in the country.

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## Identification and Characterization of Donor Lines for Drought Tolerance in Finger Millet [*Eleusine coracana* (L.) Gaertn]

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FINGER millet is one of the important staple food crops cultivated mainly as a rainfed crop in Karnataka, India, where it invariably experiences moisture stress and is reported to decrease the grain yield by up to 100 per cent depending on the duration and intensity of the stress. Therefore, identification of germplasm lines for drought tolerance will be useful to select lines for cultivation or to serve as donors in breeding programmes for drought tolerance. Therefore, 181 germplasm lines were evaluated for different physio-morphological and yield contributing traits under well-watered and drought-stress imposed field conditions during reproductive phase. From 181 lines, 10 tolerant and 10 susceptible lines were selected based on drought susceptibility index (DSI). These 20 lines were further evaluated for their stability in grain yield across the locations and cellular level tolerance against high temperature, osmotic stress (PEG stress), biochemical analyses like proline and MDA. For these 20 lines, the drought-tolerant indices like DSI, GMP, MP and HAM were calculated for grain yield and AMMI analysis performed to determine the stability of grain yield across locations. Based on the AMMI stability value (ASV), the genotypes GPU-28, GE-6336, GE-1465 was 12.6 and GE-2421 were found stable. Two contrasting genotypes, GE-6336 (tolerant) and GE-1465 (susceptible), were validated for expression of a stress-responsive gene, EcATFP-6. The expression of gene folds high in tolerant line as compared to the susceptible line. These contrasting lines can be utilized as donors in breeding programmes to develop the varieties for drought tolerance.

## Improvement of Resilience to Abiotic Stress Using Endophytic Fungi in Tomato

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ENDOPHYTES, which include fungi and bacteria are present ubiquitously and reside in the intracellular spaces of the plants symbiotically. Endophytes have been gaining importance for their role in improving stress tolerance in the host plants. In the present study, fungal endophytes (FE) isolated from harsh environmental conditions were employed to enhance drought and salinity stress tolerance in tomato (*Solanum lycopersicum* L.). The fungi belonging to the *Fusarium* genus were found to be predominant as indicated by morphological and molecular analysis. Among ten endophytes evaluated, four endophytes sustained growth under PEG-8000 and NaCl-induced stress conditions. Tomato varieties namely, *Arka Vikas*, *Arka Saurabh*, *Arka Meghali* and *Arka Abha* were screened under PEG-8000 and NaCl-induced stresses and *Arka Saurbh* was found to be stress-sensitive. Colonization of *F. incarnatum* (K-23) and *Fusarium* sp., (P-10) significantly improved seedling growth under PEG stress whereas *F. equiseti* (SF-5) and *F. incarnatum* (K-23) enhanced tolerance for NaCl-induced stress. The best two fungi were further evaluated under rainout shelter conditions for improving drought tolerance in tomato. Colonization of K-23 and P-10 improved photosynthesis and plant growth in tomato under non-stress conditions followed by yield traits and quality parameters under drought stress conditions. The select FEs also improved the growth of tomato under protected conditions. However, there was no significant difference in yield under-protected and field conditions. Hormone profiling of the plants colonized with K-23 and SF-5 indicated a significantly higher content of auxin (IAA) and gibberellic acid ( $GA_3$ ) under stress conditions. Global metabolome analysis indicated a greater number of differentially accumulated metabolites in endophyte-enriched seedlings under stressful conditions. This study demonstrated that habitat-adapted endophytes could be effectively used to improve abiotic stress tolerance in a sensitive variety of tomato.



## On the Molecular and Phenotypic Characterization of an Association Mapping Panel of Finger Millet (*Eleusine coracana* L. Gaertn) Germplasm for Drought Adaptive Traits

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FINGER millet is nutritive crop and grows well in resource limited environments. However, though a hardy species, finger millet is quite sensitive to drought and heat. Improving specific traits is known to impart lasting tolerance to abiotic stresses like drought. Previous research suggests that WUE, water mining and acquired tolerance have great relevance for drought stress. We screened a panel of 350 diverse finger millet germplasm accessions including wild varieties and landraces for important drought adaptive traits in two seasons. Plants were grown in specialised Root-study structures. Significant genetic variability was noticed in these traits across seasons, indicating a strong genetic control. The panel was also investigated for acquired tolerance adopting the temperature induction response (TIR) technique. Specific contrasts for root traits and acquired tolerant traits, drought susceptibility index (DSI) have been identified. The trait donor lines, GE 832, GE 4962 and GE 4596 can be used for future crop improvement programs. Introgression of complex physiological traits requires the help of molecular markers and hence an elaborate GWAS analysis was performed using more than 15 million SNPs obtained from the whole genome re-sequencing of the entire panel. These SNPs were separately identified on the A and B sub-genomes of the cultivated tetraploid finger millet. Additionally, we identified 43 polymorphic SSR markers from five largest super-scaffolds of the draft genome of finger millet. The genetic diversity and population structure analysis detected a total of 671 alleles which ranged from 3 to 33 alleles per locus with PIC values varying between 0.39 and 0.94. The GWAS analysis detected 259 significant marker-trait associations. Also, association mapping by SSR markers through linkage disequilibrium discovered 67 significant marker-trait associations. This analysis is the first of its kind in tetraploid species.

## Identification of Alternative Splice Variants under Drought Stress in Mulberry

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MULBERRY (*Morus* spp.), an important tree crop for sericulture industry is commercially cultivated for foliage to feed silkworms. The tree has many adaptive traits required for abiotic stress tolerance and genes contributing to stress tolerance have been identified. Stress adaptive genes are regulated at transcriptional, post-transcriptional and translational levels. One of the post-transcriptional regulatory mechanisms, alternative splicing (AS), plays a key role in developmental processes, contributing to mRNA and protein diversity. An attempt has been made to study AS events in drought-responsive genes in mulberry under drought stress. Using drought-specific transcriptome from a previous study, AS events were identified in a few transcription factors (*MabHLH144*-like, *MaNFY1*, *MaTIFY10*) and a downstream gene, *MaLTIP*. eNorthern expression analysis of selected genes in *Arabidopsis* indicated stress-responsive nature. *MabHLH144*-like and *MaLTIP* were chosen to study expression pattern. To create drought stress, plants were established in pots under rain-out shelter and stress was imposed by gravimetric approach. Stress effect was assessed by recording physio biochemical parameters 14-days post-drought stress and leaf tissue was collected for analysis of AS events. Expression analysis of *MabHLH144*-like gene by RT-PCR indicates presence of a splice variant (1500bp) under stress, a new variant (variant 3) of 250bp under both control and stress. *MaLTIP* gene indicates presence of transcript variants, variant of size 1500bp was specific to drought. The identity of splice variants was confirmed by sequencing. Determination of AS event and their corresponding variant's biological significance need to be examined, that add to the knowledge of stress tolerance mechanism in mulberry.

## Comparative Analysis of Physiological, Nutritional and Yield Attributing Traits in Finger Millet Varieties Released over the Years

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FINGER millet is considered as an important food and fodder crop with an excellent nutritional quality. The yield improvement of finger millet over the years was achieved through increased harvest index and the grain yield has reached a plateau in the recent years. In this direction, studying the varieties released over the years for yield improvement was carried out. Among the agronomic traits, the mean ear weight and LAI among the physiological traits were found highly associated with yield improvement. Exceptionally, the higher grain yield of MR-6 was due to higher biomass. Genotypes with mean ear weight in the range of 9 to 11 g with medium duration (105-110 days) and medium plant height (100 cm) could be appropriate to increase the grain yield further. However, grain nutrient quality is expected to compensate with increase in grain yield. In the present study, grain protein and zinc content were significantly and negatively associated with the grain yield. Therefore, confirmation of the decreasing quality parameters needs to be ascertained. The present study infers that, considering the nutritional quality, higher mean ear weight (9-11 g) with 3-4 productive tillers / hill could be selected for future breeding programmes to achieve higher productivity of finger millet to meet the regional food and fodder security.

## Molecular and Biochemical Characterisation of Different Indian Mandarin (*Citrus reticulata*) Types

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MOLECULAR and biochemical characterization of different Indian mandarin (*Citrus reticulata*) types viz., Coorg mandarin, Darjeeling mandarin, Kinnow mandarin, Nagpur mandarin and Khasi mandarin was carried out to understand the variability among the mandarins and also to study the variability within Khasi mandarins grown in different states of India comprising of Arunachal Pradesh, Assam, Manipur, Meghalaya, Mizoram, Nagaland and Sikkim at ICAR-Indian Institute of Horticultural Research (IIHR), Hessaraghatta, Bengaluru during the year 2020-21. Genetic diversity assessed by employing ten simple sequence repeat markers showed high polymorphic information content and allelic diversity. Both dendrogram and population structure analysis showed grouping based on mandarin types and their geographical occurrence. Biochemical parameters (total soluble solids, acidity, vitamin C, total carotenoids, sugars, organic acids, vitamins) and profiling of (sugars, organic acids, water soluble vitamins and fat-soluble vitamins) also showed significant differences among the mandarin types indicating wide genetic diversity among the different mandarin types. Among Khasi mandarins, the Khasi mandarin grown in Meghalaya recorded highest value for TSS, sugars and organic acids indicating good fruit quality characters. Further, molecular characterization data also recorded the distinctness in the genetic diversity. Thus, the present investigation highlighted the genetic distinctness and biochemical difference among different mandarin types and also within Khasi mandarins which can be effectively used in selection of parents for future breeding programmes.

## Isolation and Evaluation of Endophytic Fungi from Pokkali Rice Cultivars for their Ability to Impart Salinity Stress Tolerance to their Host Plants

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ENDOPHYTES (fungal or bacterial) are microorganisms that colonize the intercellular spaces of hosts and complete all or one part of their life cycle within the plant system without causing any damage and visual symptoms. These endo-symbionts have been reported to confer biotic and abiotic stress tolerance to their hosts and are readily isolated from hosts and cultured on artificial media. In this study, an attempt was made to examine endophytic fungal diversity of salt adapted rice cultivars from Pokkali and that from a salt sensitive rice variety, IR-64. The study examined the relative ability of endophytes from these contrasting rice cultivars in tolerating salinity stress. Finally, the study also examined the role of endophytes from salt adapted rice cultivars in imparting salinity stress tolerance to their hosts. A total of 62 isolates were obtained from salt adapted and salt sensitive rice cultivars. They were morphologically characterized into 11 operational taxonomic units (OTUs). Molecular characterization using ITS primers, identified them as *Orbilia foliicola* and *Curvularia lunata* from salt adapted Pokkali varieties and *Cochliobolus miyabeanus* from salt sensitive IR-64. Mycelial growth of *Curvularialunata* was more in salt amended condition (150 mM) than in control. Isolates from salt adapted paddy genotypes were able to tolerate higher levels of salinity stress compared to isolates from salt sensitive paddy genotype. Bavistin-treated Pokkali seedlings were unable to withstand salinity stress because of loss of non-target beneficial fungal endophytes indicating that endophytes may have role in improving growth and development under stress in their host plants.

## Trans-generational Salt Stress Tolerance of Endophyte Enriched IR-64 Rice (*Oryza sativa* L.) Plants

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ENDOPHYTIC fungi dwelling in plant tissues are widely recognised to impart tolerance to their host plants against abiotic and biotic stresses. This study investigates if such effect of endophytic fungi on host plants' tolerance to abiotic stress is transmitted across seed generations with specific reference to salinity stress. The salt adapted endophyte *Fusarium* sp. (GenBankAccNo.MH593375) was isolated from the hydro-halophyte *Suaeda filiformis* Dumart growing in Marakkam backwaters, Tamil Nadu. Manasa *et al.* (2020) inoculated it to a salt sensitive paddy genotype, IR-64 and found it rendered salinity tolerance to their new host plant. This study examined the transmission of a salt tolerant endophytic fungi in rice plants (var. IR 64) and the performance of plants when subjected to salinity stress compared to those not treated with the endophyte. The study showed seedlings grown from plants treated with endophyte were comparably greater in length and have enhanced shoot length at 70-days under salt stress. Endophyte enriched plants showed reduced DAB staining, indicating low ROS levels and reduced oxidative stress. Na<sup>+</sup>: K<sup>+</sup> ratios were significantly reduced in endophyte enriched plants probably due to the reduced uptake of Na<sup>+</sup> into shoot tissue. Endophyte enriched plants had a higher tiller number, panicle number and spikelet number per plant compared to the un-enriched plants. By and large, these results indicate that the phenotypic effects of the endophyte, *Fusarium* sp. persist from one generation to the other and thus paves the way for the use of endophytes as an alternative strategy to develop salinity stress tolerant plants.

## Synthesis of Therapeutically Active Secondary Metabolite, 1-DNJ by *in vitro* Callus Culture in Mulberry

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MULBERRY is known for its phytochemical constituents apart from its primary usage as host plant for silkworms in sericulture. 1-DNJ, an iminosugar belonging to secondary metabolite pool, is one such phytochemical which has therapeutic application against type 2 diabetes since it is a potent  $\alpha$ -glucosidase inhibitor. 1-DNJ is unique to mulberry and few plant species such as *Commelina*. In mulberry 1-DNJ is found at very low levels (0.1% per gram dry weight) in root, stem, leaf and shows varietal, spatial and temporal variation, hence a constrain for large scale plant based DNJ production. The current study focused on *in vitro* strategy of DNJ production and enhancement through the use of elicitors. V-1 being a leading cultivar in southern Karnataka, callus induction was standardised from V-1 leaf and stem explants in media supplemented with both 2, 4-D and BAP. Stem explants due to increased phenolic exudation was not efficient in callusing. Different extraction and estimation methods of 1-DNJ from callus derived from leaf explants revealed that 1-DNJ is synthesized in the callus and further, in one of the treatments, its content matched the leaf levels of about 3 mg/g. Efficient *in vitro* alpha-glucosidase enzyme inhibition with callus extracts compared to leaf indicated that 1-DNJ is in a pure/active form in callus. Leaf treated with chemical elicitors is shown to enhance 1-DNJ synthesis. Future prospects include attempting large-scale *in vitro* liquid culture / elicitation procedures and purification methodologies for plant-based DNJ production, which might be a significant secondary agricultural product used in therapeutic area.

## Bioprospecting of Bacterial Endophytes Isolated from Himalayan Cold Desert for Abiotic Stress Tolerance in Plants

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SYMBIOTICALLY conferred abiotic stress tolerance by endophytes can provide fitness to plants. In this study, 58 bacterial endophytes isolated from plants growing at Himalayan cold desert were explored for induction of drought and salt stress tolerance in rice (IR-64) and tomato (Arka Saurabh) using NaCl and Polyethylene glycol PEG-8000 at different concentrations of which, nine bacteria showed growth up to 2.0 M NaCl and eight bacteria up to 20 per cent PEG. As per probit analysis, the  $LC_{50}$  value of NaCl (Salinity) for rice was 150 mM and 117mM for tomato. The  $LC_{50}$  value of PEG for drought was 14.3 per cent for rice and 15.3 per cent for tomato. These bacteria when inoculated to pre-germinated rice and tomato seeds and grown in paper towel treated with 150 Mm and 117 mM NaCl, the isolates NBE 20 and NBE 23 showed increased growth of tomato seedlings and PBE 8 and NBE 7 showed increased growth of rice seedlings. Under drought (20% PEG) the isolates PBE 2, PBE 4 and CBE 11 showed better growth of rice seedlings and PBE 6, CBE 11 and NBE 5 showed better growth of tomato seedlings. These bacteria were identified as *Bacillus cereus*, *Pseudomonas chlororaphis*, *Lysinibacillus macroides*, *Enterobacter hormaechei*, *Stenotrophomonas maltophilia*, *Acinetobacter lwoffii*, *Pseudomonas fluorescenes*, *Enterobacter cloacae* and *Enterobacter asburiae* by 16S rRNA sequences and further evaluated under greenhouse conditions. The endophytes inoculated rice and tomato plants showed increased growth, yield and physiological traits. Among the 9 selected bacteria, the *E. hormaechei* and *E. asburiae* were efficient in mitigating salt stress (4 dS/m) and *S. maltophilia* and *A. lwoffii* were efficient in mitigating drought stress (50% FC) in rice and tomato crops, respectively. This study revealed that the selected endophytes can mitigate salinity and drought stress in rice and tomato.

## Blast Disease Management in Paddy through Microbial Consortia and their Molecular Characterization

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RICE blast is a serious fungal disease of rice (*Oryza sativa*. L) caused by the pathogen *Phricularia oryzae*. It can infect the rice plant at any growth stage and cause total crop failure. The present study was carried out to manage the blast disease by the application of isolated microorganisms. Thirty bacterial and 6 fungal isolates were isolated from the rhizosphere soil of paddy. The isolates were screened to check the antagonistic activity against the blast pathogen, *Pyricularia oryzae*. Three bacterial isolates (RCB-4, RIB-2, RBB-3) and fungal isolat RCF-1 were found to be effective in inhibiting the growth of the pathogen. Molecular characterization of these isolates were carried out and came to know that the three bactrial isolates (RCB-4, RIB-2, RBB-3) were *Bacillus subtilis*, *Pseudomonas fluorescens* and *Bacillus licheniformis*, respectively and the fungal isolate (RCF-1) was found to be *Trichoderma harzianum*. Further these isolates were used for pot culture studies for blast disease management under greenhouse conditions. There were 11 treatment in which T<sub>1</sub> was absolute control, T<sub>2</sub> (pathogen), T<sub>3</sub> to T<sub>6</sub> included application of single microbial isolate and from T<sub>7</sub> to T<sub>11</sub> consortium of isolates were used. Among all the treatments, T<sub>11</sub>, which contained consortium of all the 4 bioinoculants showed better results with less disease incidence (6.11%), more vegetative growth (10 tillers / pot) and good grain yield (27g / pot) compared to control (T<sub>1</sub>) and single inoculants. Hence, microbial inoculants as a consortia was able to suppress the pathogen and give better yield.

## Effect of Microbial Consortium in the Management of Stalk Rot of Maize at Different Levels of Potassium

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THE actinomycete isolated from maize rhizosphere soil was identified as *Streptomyces rochei* CMB47 with 98.60 per cent similarity. The bioinoculants viz., *Bacillus subtilis*, *Pseudomonas fluorescens* and *Trichoderma viride* were obtained from the Department of Agricultural Microbiology, College of Agriculture, V. C. Farm, Mandya and used in this study. *In vitro* screening of four bioinoculants was done by dual culture method against *Fusarium verticillioides*, a maize stalk rot pathogen. The per cent growth inhibition by *B. subtilis* (67.06 %), *P. fluorescens* (49.11 %), *T. viride* (48.08 %) and *S. rochei* CMB47 (59.07 %) was measured. The microbial consortium was developed by using four bioinoculants. A field experiment was conducted at ZARS, V. C. Farm, with thirteen treatments consisting of various potassium levels (0, 75, 100 and 125 per cent of RDK), microbial consortium and *F. verticillioides*. The pathogen was inoculated at 60 DAS and after seven days microbial consortium was applied. Potassium at 125 per cent recommended dose + microbial consortium + *F. verticillioides* (T<sub>10</sub>) and 100 per cent RDK + consortium + *F. verticillioides* (T<sub>7</sub>) recorded lower stalk rot incidence (disease rating scale) of 2.27 and 2.87, respectively. These treatments recorded significantly higher growth, kernel yield (7.76 and 7.73 tons/ha) and stalk yield (11.83 and 11.79 tons/ha). T<sub>10</sub> and T<sub>7</sub> had highest cost-benefit ratio of 2.12 each, showed reduced disease incidence, improved growth and yield. Absolute control has showed lowest growth and yield. The microbial consortium consisting of bioinoculants improve disease control, crop growth and yield.

## Development of Transgenic Mungbean [*Vigna radiata* (L.) Wilczek] Resistant to Mungbean Yellow Mosaic Virus (MYMV) and Identification of Anti-MYMV Candidate Compounds by *in silico* Analysis

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MUNGBEAN is an important short duration rainfed legume crop of India. The crop productivity of mungbean is affected by various pests and diseases, among which viral disease like mungbean yellow mosaic virus (MYMV) cause devastation in all the mungbean growing areas. The binary plant cloning vector pCAMBIA1305.2 carrying the MYMV *Replicase* (*Rep*) gene was constructed and transformed into a competent *Agrobacterium tumefaciens*, strain LBA4404 and was used for co-cultivation of mungbean varieties, KKM-3, IC-39340-1, China mung and LM-1668. The putative transformants were selected on shooting media, multiple shoots were elongated and rooted on rooting media. The transformed *Agrobacterium* harboring *Rep* gene was used for *in planta* transformation of mungbean varieties by seed *Agro*-inoculation method. The GUS assay and genomic PCR analysis of the transformants revealed, positive putative transgenic lines for *Rep* gene specific primers and had a positive correlation with vector specific *hptII* primers. Likewise, *VirG* amplification revealed no *Agrobacterium* contamination in the apoplast of all the putative transformants. The unknown bacterial contaminates found in the *in vitro* cultured mungbean plantlets were isolated, identified and controlled. Initially the contaminants were morphologically, biochemically and molecularly characterized and identified as *Enterobacter* sp. and *Lysinibacillus* sp. Finally, the antibiotic disc diffusion test revealed that both bacteria were sensitive to the antibiotic Amoxicillin. The 3D structure prediction of *Rep* protein was done *ab-initio* and refined by quality assessment and model validation with different molecular web tools. The binding sites of *Rep* protein were predicted and the ligand- receptor interactions were studied by molecular docking analysis. Among the mungbean bioactive compounds Isovitexin had higher binding affinity towards target protein.

## Transformation of Tobacco (*Nicotiana tabacum* L.) with *Nia* Gene of Papaya Ringspot Virus (PRSV) and Bioinformatics Analysis of *Nia* Protein

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PAPAYA (*Carica papaya* L.) is an important tropical and subtropical fruit crop, belonging to family *Caricaceae*. *Papaya ringspot virus* (PRSV) is a wide spread and economically important destructive virus that affects papaya worldwide and causes heavy losses. The virus genome encodes a single large polypeptide which is subsequently cleaved into smaller functional proteins like nuclear inclusion A (*Nia*) protein, *Nib* protein, helper component protein and coat protein. The current study was undertaken to develop PRSV resistant transgenic tobacco *via* pathogen derived resistance approach using the *Nia* gene, as well as to predict the structure of the *Nia* protein. The present research commenced with the extraction of RNA followed by isolation of the viral *Nia* gene and then cloning in to pTZ57R/T vector and after confirmation, plasmid DNA was sequenced and transformed into *N. tabacum* using binary vector pBI121. The transfer of transgene in the putative transgenic plants ( $T_0$ ) was confirmed by marker specific and gene specific primers. Phylogenetic analysis of *Nia* gene sequence of present study suggested that there was considerable mixing and movement of isolates within and among all the geographical regions. The *Nia* protein structure model was predicted using Homology modeling *via* SWISS MODEL server and validated by generating a Ramachandran plot. The generated model could be further utilized for understanding the functional aspects of the *Nia* protein and its relevance in the infection process and also methods to combat the PRSV incidence.

## **Transformation of Cucumber (*Cucumis sativus* L.) with Papaya Ring Spot Virus (PRSV) Coat Protein (CP) Gene and Serological Assay for the Detection of PRSV**

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*CUCUMIS SATIVUS* L. (cucumber) an important crop of the cucurbitaceae family is a water-rich vegetable mainly consumed fresh or in preserved form. Papaya ring spot virus (PRSV) is a major obstacle to large-scale cucumber production in India. Early disease detection and conferring resistance in crop plants against viral disease are two important measures that aid in disease management. In this context, the present study was aimed to develop transgenic *C. sativus* L. resistant to PRSV using the *CP* gene via pathogen-derived resistance approach, bioinformatics analysis of *CP* gene and standardization of serological assays for detection of PRSV. The study began with the isolation of the *CP* gene from the RNA of infected leaf, which was then cloned, sequenced and transferred to *Cucumis sativus* L. via *Agrobacterium*-mediated transformation using binary vector pBI121. The presence of transgene in the putative transgenic plants ( $T_0$ ) was confirmed by histochemical GUS assay and molecular PCR analysis. The sequence of *CP* gene isolated in this study was then compared with other Indian isolates which showed 94-95 per cent similarity at the nucleotide level followed by the phylogenetic tree construction which showed that PRSV isolate from Bengaluru is closely related to Delhi isolate compared to isolates from Haryana, Pune and West Bengal. The lateral flow immunoassay (LFIA) technique was developed and valid results were generated with field collected PRSV infected leaf samples at dilutions ranging from 1:10 to 1:800 and these results were comparable in terms of sensitivity to the gold standard method ELISA performed in this study.

## **Phenotyping of Cowpea (*Vigna unguiculata* L.) Genotypes for Quantitative Traits and Screening for Disease Resistance Using SSR Markers**

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COWPEA is an important pulse crop of tropics and subtropics grown for vegetable, fodder and green manure purposes but productivity of cowpea is limited due to viral and fungal diseases. In the present study 120 genotypes were evaluated for resistance to *Bean common mosaic virus* (BCMV), *Cowpea mosaic virus* (CPMV), and rust disease along with yield traits in augmented design during *kharif* 2020. Statistical analysis was computed on data collected for traits *viz.*, plant height, number of branches, days to 50 per cent flowering, pods per plant, pod length, number of seeds per pods, hundred seed weight and total seed weight of five plants. Phenotypic coefficient of variance and Genotypic coefficient of variance were found to be narrow for almost all the characters. The per cent of disease incidence was varied from 0.0 to 33.33 per cent and the highest disease incidence was recorded in IC-202777 and IC-237422 (33.33 %). Four BCMV linked markers tested for validation, out of which three (M15, M80 and Y96) gave the desired amplification. Single marker analysis (SMA) revealed that out of four BCMV SSR markers, MA15 explains 3.96 per cent of phenotypic variation. Whereas in CPMV, four SSR linked markers tested, out of which only two (VM1 and AG1/AF48383) gave the desired amplification. SMA for CPMV revealed that out of three SSRs, AG1/AF48383 explains 3.80 per cent of phenotypic variation. For the disease rust, all three SSRs, (VuUGM02, VuUGM08 and VuUGM19) gave the desired amplification in the cowpea genotypes tested and were able to distinguish between resistant and susceptible ones.

## **Studies on Screening and Biochemical Characterization of Ricebean (*Vigna umbellata* L.) Accessions for Bean Common Mosaic Virus**

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THE survey conducted during 2019-20 in six districts of Southern Karnataka to assess the incidence of mosaic disease of cowpea, revealed the occurrence of disease in the range of 18.66 to 50 per cent. Highest disease incidence was recorded in Bengaluru urban (50 per cent) and least disease incidence in Chikkabalapura district (18.66 per cent). Infected cowpea was used as a source of bean common mosaic virus. The virus was infected to rice bean accessions through mechanical sap transmission. Presence of virus in the infected plant was confirmed by RT-PCR technique using primers specific for coat protein gene. Based on the extent of severity of symptoms, it was found that, 39 accessions were immune, EC-37 showed resistance reaction, EC-48, EC-34, IC-3 showed moderately resistance reaction, EC - 27, KBR 1 showed susceptible reaction. No accessions were highly susceptible. Rice bean accessions differing in the level of resistance were used further for biochemical studies. The total chlorophyll content, total sugars, phenolics were less in susceptible accessions. Total protein content was more in susceptible. Activity of antioxidant enzymes like peroxidase, catalase and super oxide dismutase were high in infected plants and varied among the infected rice bean accessions.

## **Study on Drought Stress in Paddy (*Oryza sativa* L.) and Assessment of 8-Oxo-Guanine for DNA Damage**

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CLIMATE change poses a major threat to the rice production and the future food security as the demand for the same is also growing. Genetic improvement and suiting for the change in climate is one of the approaches to address the threat. With respect to drought, one of the current problems in rice cultivation is the lack of variant with both high yield and better drought tolerance. As drought being an integral trait, mapping as much component trait to be introgressed, including the traits that are apparently not related, but with sufficient scientific evidence to strengthen the integral trait is envisaged. Therefore a study was taken to understand the *in vitro* drought response of different paddy genotypes and relative quantification of DNA damage marker; 8-oxo-guanine using EI GCMS. The 23-days-old seedlings that underwent low and mild stress recovered faster. Above 15 per cent PEG, the relative tolerance were found to be; Gen-ric-81 > Gen-ric-767 > Gen-ric-308 > Gen-ric-430 > Gen-ric-288 > Gen-ric-620 > Gen-ric-318 > Gen-ric-755 > Gen-ric-823. The relative quantification of 8-oxoguanine level in all stressed genotypes showed a Gaussian response. From the absolute control, with respect to increase in stress a proportional increase in 8-oxo-guanine was found, but at different rate and level. Considering 8-oxoguanine as a marker of OGG, one of the enzymes of BER, no correlation was found between drought tolerance and DNA repair efficiency. Therefore, in view of strengthening a target trait, large-scale independent massive trait mining is to be taken up for future crop improvement. ric-823. The relative quantification of 8-oxoguanine level in all stressed genotypes showed a Gaussian response. From the absolute control, with respect to increase in stress a proportional increase in 8-oxoguanine was found, but at different rate and level. Considering 8-oxo-guanine as a marker of OGG, one of the enzymes of BER, no correlation was found between drought tolerance and DNA repair efficiency. Therefore, in view of strengthening a target trait, large-scale independent massive trait mining is to be taken up for future crop improvement.



## Biochemical Investigations on Post-Cooking Stability of Chickpea (*Cicer arietinum* L.) Seeds of Local Cultivar 'Karikadle'

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FOOD spoilage affect health and contributes in economic loss. Cooked foods are more prone to spoilage because of high water content and presence of small organic molecules. Therefore, many aspects of food preservation is being explored in food industries including the use of nature similar or nature identical systems and molecules. In this sense, traditional knowledge on post cooking stability of *karikadle*, one of the cultivar of black chickpea, was validated along with three reference varieties viz., *kabuli*, *desi-brown* and *desi-green*. Highest post cooking stability (PCS) was observed in in both *karikadle* and *desi-green* types. Biochemical investigations showed the presence of high phenolic content and high antioxidant activity in seed coats of *karikadle* and *desi-green*. A comparative analysis between unit phenolic content and the respective antioxidant activity showed the *karikadle* phenolics with exceptionally high antioxidant activity with several order of magnitude. No relation was found between seed coat structural features of all varieties studied and the respective post cooking stability. A comprehensive antibacterial study showed no significant role of phenolics from seed coat as well as endosperm from all the genotype. However, it was observed that, antibacterial intermediate(s), capable of inhibiting bacterial exogenous hydrolase enzymes and retard bacterial growth was found formed from *karikadle* seed coat phenolics, probably as a result of an early microbial activity. This study points the presence of a '*karikadle* - specific' phenolic compound(s) in the seed coat could be one of the reasons for the observed PCS. Further, studies are required to characterize the same.

## Effect of Heat Stress on Antioxidant Defense Response and DNA Oxidation in different Paddy Genotypes

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FREQUENT extreme high temperature events are one of the consequences of climate change and hence, heat stress has become a major abiotic stress to crop plants. Heat stress tolerance and yield are quantitative traits. Many component traits, including the traits that are closely related to the quantitative trait and other apparently unrelated traits, are involved in the integrated traits. Therefore, basal thermotolerance by vegetative growth, antioxidant defense response (related trait) and ability to protect DNA from oxidative damage (apparently unrelated trait) etc. were studied in eight paddy genotypes. Eight paddy genotypes were assessed for basal thermotolerance and found genotypes GR\_214 and GR\_531 as highly tolerant. In the assessment of antioxidant defense response, highest superoxide radical scavenging potential in GR\_470, GR\_531 and GR\_823; and highest hydrogen peroxide depletion potential in GR\_601 and GR\_823 were observed. To understand the effect of heat stress induced oxidative DNA damage, the marker 8-oxoG was quantified using GCMS. Higher level of 8-oxoG was found in GR\_456, GR\_660 and GR\_285 that were grown at 48°C, 46.5°C and 46.5°C, respectively. On analysis of the basal thermotolerance, antioxidant defense response and 8-oxoG levels, it was found that these traits are independently existing and regulated among eight paddy genotypes studied. Therefore, while exploring for developing heat tolerant crop genotypes, screening genetic resources only based on growth and yield is not sufficient to fulfill the future crop improvement. Hence, exploration, annotation and marking of component traits associated/ apparently associated with targeted integrated trait may help in more efficient crop improvement.

## Biochemical Analysis of Rice Bean Varieties for Tryptophan, Methionine and Antinutritional Factors

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PULSES are the major source of protein and are supplemented along with cereals for protein in the diet. Though pulses are rich in protein they lack sulfur containing amino acids like methionine and tryptophan. Rice bean, a potential leguminous pulse crop contains high quantity of tryptophan and methionine. But it also harbours antinutritional factors such as saponins and polyphenols. In this study protein, tryptophan, methionine and antinutritional factors were quantified in some of the cultivars of rice bean and compared with the green gram variety KKM3. In addition, the key regulatory enzyme involved in the methionine synthesis activity was also analysed at different seed developmental stages. The total crude protein in rice bean genotypes was ranging from 21 to 23 per cent. The essential amino acid content was higher than nonessential amino acids in rice bean seeds. The methionine content in most of the rice bean varieties was found to be more than in green gram. Activity of aspartate kinase was maximum during the mid-stage of seed development. Specific activity of aspartate kinase was higher in rice bean than green gram. Antinutritional factors such as polyphenols, phytic acid, saponins and trypsin inhibitors were found to be lower than in the green gram variety KKM3. Polyphenols in rice bean were found within permissible limits. Considering protein content, methionine content and antinutritional factors, KBR1 and RBL50 were found to be best suited for consumption among the rice bean varieties tested in this study.

## Study of Secondary Metabolites Association with Drought in Selected Paddy Genotypes

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THE current unpredictable climate changes are causing frequent and severe droughts. Drought is a major cause of yield loss in rain-fed rice (*Oryza sativa* L.) grown on over 40 million ha in Asia. The objective of this study was to understand the association of secondary metabolites with drought. In the present study fifteen genotypes of paddy were evaluated for total phenolics content, total flavonoid content, fatty acid methyl esters, phytosterols *i.e.*, stigmasterol and campesterol in seed and husk using UV spectrophotometric, GC-MS and HPLC. The results showed that the total phenolics content from seed and husk ranged from 96.94 to 1111.66  $\mu\text{g/gm}$  and 229.16  $\mu\text{g/gm}$  to 608  $\mu\text{g/gm}$ , total flavonoid content from seed and husk ranges from 0.05  $\mu\text{g/gm}$  to 1.27  $\mu\text{g/gm}$  and 0.05  $\mu\text{g/gm}$  to 1.34  $\mu\text{g/gm}$ . The seed stigmasterol and campesterol ranged from 0.84  $\mu\text{g/gm}$  to 10.96  $\mu\text{g/gm}$  and 0.11  $\mu\text{g/gm}$  to 0.41  $\mu\text{g/gm}$ , respectively. The FAME analysis showed that the linolenic acids (18:3), linoleic acid (18:2), palmitoleic acid (16:1) and stearic acid (18:0) were present in all fifteen genotypes. Genotypes 49-3594 and 28-0397 were selected for studying the association of phytosterol content in drought stress following germination test. Under PEG induced drought stress at 5, 10, 15, 20, 25, 30 per cent, respectively. The results showed that stigmasterol content increased with stress intensity in both genotypes till 15 per cent PEG. The campesterol concentration also increased in 49-3594 and 28-0397 but in 49-3594 showed decline in concentration beyond 10 per cent PEG. Further more, investigation is required to understand phytosterols role in drought.

## Study on Transferability of SSR Markers from Finger Millet (*Eleusine coracana* L.) to Browntop Millet (*Urochloa ramosa* L.)

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BROWNTOP millet (*Urochloa ramosa* L.) can adapt to variety of climates, uses limited amount of water for growth and development and has high minerals and fibre content. In current study the diversity present in the germplasm accessions was evaluated by measuring thirteen morphological traits across twenty seven browntop millet accessions. GCV ranged from low (2.73) for days to maturity to high (80.68) for panicle width, PCV ranged from low (4.01) days to maturity to high (80.73) for panicle width. The difference in GCV and PCV was very narrow (<0.2) for panicle width indicating least influence of surrounding environment which was further supported by high value of broad sense heritability (>95%). Grain yield per plant showed significant positive correlation with plant height, number of panicle per plant, panicle length, peduncle length, flag leaf width and flag leaf length. Principal component analysis revealed that 88.73 per cent of total variation was from grain yield per plant, total panicle weight per plant, panicle width and flag leaf length. 100 SSR markers of finger millet tested on two browntop millet accessions showed 52 per cent transferability. Thirty selected markers were studied on 27 browntop millet accessions where, number of alleles per primers varied from 1 to 5, major allele frequency ranged from 0.44 to 1, genetic diversity ranged from 0 to 0.72 and PIC ranged from 0 to 0.68. Dendrogram based on UGPMA clustering was divided into two major clusters. The study identified best accessions for different traits for further use in the browntop millet breeding programmes.

## Computational Methods to Identify Bioactive Phytocompounds of Meadow Saffron (*Colchicum autumnale* L.) for Rheumatoid Arthritis

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RHEUMATOID ARTHRITIS (RA) is an autoimmune disorder that affects roughly 1 per cent of the population worldwide and is characterized by severe pain due to inflammation of the joints. Treatment with DMARD's and NSAID's cause side effects. This research was conducted with the aim of exploring medicinal plant *Colchicum autumnale* L. as an alternative treatment for RA. The study begins with the identification of RA receptors from OMIM, Pharma GKB, GeneCard disease database. Venn-diagram resulted 15 commonly interacting targets and their structures were downloaded from Uniprot and RCSB PDB. The anti-inflammatory and anti-arthritis compounds were identified from commercial plant extract of meadow saffron, with the help of GCMS analysis. Swiss ADME tool showed 35 compounds were active. The structures of those compounds were drawn in Chems sketch and energy minimized using Avagadro. Molecular docking was done for 15 RA targets and 35 phytocompounds in Maestro v13.1. Among all docked complexes, histamine-PTPN22 received a favourable docking score of -7.13 kcal/mol. These compounds were then run in desmond 3.2 for simulations to determine the stability of the interaction. The RMSD value for the 100ns run varies from 0 to 3 Å, indicating that the interaction was stable with ligand contact residues. Pharmacophore modelling was performed by searching for histamine contact positions in the ZINCpharma database. The docking procedure was performed for those compounds, but none of them showed a better docking interaction than histamine. A computational-based study revealed that histamine has a promising future as potential therapeutic candidates against RA.

## Development of Magic Population and Tagging of Resistance to Major Diseases in Maize (*Zea mays* L.)

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NORTHERN corn leafblight (NCLB) and sorghum downy mildew (SDM) of maize are serious constraints to maize production worldwide. An attempt was made to develop multiparent advanced generation intercross (MAGIC) population segregating for NCLB and SDM from the eight diverse founder parents (PDM-4341, CML451, CM202, CM212, SKV50, CAL1443, CAL1518 and VL109545) which were chosen based on SSR diversity and phenotypic response to multiple diseases. The MAGIC population was developed based on simple funnel crossing scheme to obtain 288 F<sub>3</sub> progenies and they were phenotyped during summer and *kharif*2020 using alpha-lattice design in the artificial disease screening nurseries for NCLB and SDM at ZARS, Mandya. Analysis of variance among F<sub>3</sub> progenies showed significant variation for NCLB and SDM disease incidence. The *per cent* disease severity for NCLB and *per cent* disease incidence for SDM showed high PCV and high heritability. Out of 288 F<sub>3</sub> progenies, 14 F<sub>3</sub> progenies were resistant to NCLB whereas, four progenies were resistant to SDM. Genome wide association study was conducted using 90 SSR markers to detect the genomic regions controlling resistance to NCLB and SDM. PCA analysis suggested an absence of unique groupings within the population. Based on the results of Mixed linear model (MLM) four SSRs were found associated with resistance to NCLB and one SSR marker (bnlg109) for SDM. BLAST analysis revealed that five SSRs associated with NCLB and one SSR marker with SDM were found overlapped within annotated genes (*Callose synthase 7*, *Non-specific lipid-transfer protein*, *Homeodomain leucine zipper family IV protein*, *BTB/POZ domain-containing protein NPY2*, *AP2-EREBP-transcription factor 197* and *F-box/LRR-repeat protein*) with plant defence mechanism.

## Identification and Stability of New Sunflower (*Helianthus annuus* L.) Hybrids for High Oleic Acid and Yield

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THE purpose of this study was to improve sunflower oil quality by altering the fatty acid composition, hence an investigation was carried out to find stable high oleic sunflower hybrids with high seed yield and oil content. The 40 high oleic testers identified from high oleic gene pool were crossed with six CMS lines (CMS 1103A, CMS 234A, CMS 903A, ARM 249A, CMS 59A and CMS 103A) in a line x tester mating design during *kharif*2019 to produce 240 experimental hybrids. The resultant experimental hybrids thus obtained were evaluated during *rabi* 2019 to identify the fertility restoration and sterility maintenance behaviour. Out of the 40 high oleic testers evaluated, 28 testers were identified as common restorers while six testers were identified as common maintainers across six CMS lines. The resultant 168 fertile hybrids (6 lines x 28 testers) were evaluated during summer 2020 in alpha lattice design to assess the combining ability and heterosis for high oleic and high yield. Out of 168 hybrids, 30 promising hybrids exhibiting high oleic acid, high seed yield and oil content were identified based on Smith-Hazel selection indices. The hybrid, ARM 249A×F-20 exhibited highest selection index value of 68.15, this cross combination possessed ultra-high oleic acid content of 91.97 per cent. The 30 promising high oleic hybrids along with four check hybrids were evaluated to assess the stability in three diverse locations across Karnataka during *rabi* / summer 2020-21 (December to March) and *rabi* 2021 (September to December). Among the 30 high oleic hybrids evaluated, one cross combination, CMS 903A×K-11 exhibited stable expression of high oleic acid content (73.5%), along with stable high yield and oil content over both the seasons and three locations. The identification of this hybrid combination served the purpose of this study to derive the most stable high oleic hybrid with high yield.

## Genotype x Environment Interaction in Cowpea [*Vigna unguiculata* (L.) Walp.] for Seed Yield, Zinc and Iron Content

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AN investigation was carried out to assess the stability of cowpea genotypes which are rich in zinc and iron with optimum seed yield. Twenty advanced breeding lines which are derived from two crosses viz., Pant Lobia-2 (PL-2) × NBC-39 and Pant Lobia-5 (PL-5) × EC-402104 along with four checks viz., PL3, PL4, PGCP-6 and KBC-9 following RCBD with two replications were sown at GKVK, Bengaluru, CoA, V.C. Farm, Mandya and ARS, Pavagada during summer 2021. Based on Eberhart and Russel (1996) model, the genotypes G-6, G-8 and G-18 were stable for seed yield plant<sup>-1</sup>. While the genotypes G-1, G-3 and G-7 were identified as promising and stable for zinc content. Genotypes G-7, G-8 and G-13 were found promising and stable for iron content. The AMMI analysis showed significant differences for all traits. The contribution of environment towards stability of the genotypes found significant for all characters except for days to 50 per cent flowering. G × E interaction was found to be significant for five traits. The GGE Biplot for ranking of genotypes the G-18 has both high mean yield and stable across environments for seed yield plant<sup>-1</sup>. While G-7 with high zinc and G-13 with high iron content was found to be stable across environments. The stable genotypes identified based on ASV and SI genotypes G-1, G-5, G-7, G-11, G-13 and G-18 were identified as most stable for seed yield, zinc and iron contents. These genotypes could be forwarded for multilocation trials to assess their adaptability and stable yield.

## Efficacy of Biocontrol Agents and Insecticides on Different Strains of Fall Armyworm, *Spodoptera frugiperda* (J.E.Smith) (Lepidoptera : Noctuidae)

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FALL armyworm (FAW), *Spodoptera frugiperda* is an important insect pest of maize causing significant economic damage. It is composed of morphologically similar subpopulations namely corn ('C') and rice ('R') strains that differ in behavior. Corn strain is predominantly found on maize and large grasses whereas rice strain is found on rice and pasture grasses. In the present study, on the efficacy of biocontrol agents and insecticides indicated that FAW populations were collected from different locations and distinguished into 'R' and 'C' strains based on the polymorphic sites of mt COI and Tpi genes. The identified rice and corn strains were subjected to inter-strain mating and evaluated for their susceptibility to biocontrol agents and insecticides. The cross between 'R' female and 'C' male produced offsprings whereas, opposite cross failed. The adult males of 'C' and 'R' strain showed significant mean antennal response of 1.8458 and 1.17205 mV, respectively towards the pheromone. The biocontrol agents *Bacillus thuringiensis* Berliner HD-1, *Metarrhizium anisopliae* (Metchnikoff) Sorokin (Ma-35) and *Heterorhabditis indica* Poinar (NBII-38) recorded the lowest LC<sub>50</sub> values and caused the similar levels of larval mortality against rice and corn strains. The *Spf*-NPV recorded the LC<sub>50</sub> of 5.0×10<sup>8</sup>, 6.2×10<sup>8</sup> OBs<sup>-ml</sup> for 'R' strain and lab population. *Telenomus remus* Nixon caused 93.20 and 92.85 per cent parasitism and *Trichogramma chilonis* Ishii caused 76.15 and 76.90 per cent parasitism on rice strain and lab population, respectively. The emamectin benzoate was relatively more toxic followed by spinetoram and chlorantraniliprole against FAW.

## Faunistic Studies on Agromyzid Flies Associated with Economically Important Crops of Karnataka

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THE study was carried out at University of Agricultural Sciences, GKVK, Bengaluru. Agromyzid flies were collected from different parts of Karnataka during October, 2020 to November, 2021 by using sweep net, malaise trap and rearing from infested host plants. A maximum of 76.85 per cent of the specimens were collected by rearing, followed by sweep net (19.90%) and malaise trap (3.24%). A total of 432 agromyzids were collected, of which 89 males and 343 were females. Among the collected flies 59.02 and 40.97 per cent belonged to the subfamilies Agromyzinae and Phytomyzinae, respectively. Agromyzids were collected from different infested parts of host plants viz., leaves, pods, seeds and stems. Among these, the highest number of flies were obtained from leaf mines (56.92%) followed by pods (30.72%), stem (6.92%) and seeds (5.42%). Agromyzids like *Melanagromyza obtusa* Malloch, *M. hibisci* Spencer and *M. sojae* Zehntner were basically collected from crops viz., *Cajanus cajan* (L.) *Abelmoschus esculentus* (L.), *Glycine max* (L.), respectively. Serpentine leaf miner, *Liriomyza trifolii* Burgess collected from *Ricinus communis* (L.), *Vicia faba* (L.), *Cucumis sativus* (L.), *Solanum lycopersicum* (L.), *Spinacia oleracea* (L.) and *Vigna unguiculata* (L.). Whereas, *Ophiomyia lantanae* Froggatt and *Calycomyza* sp. affects seeds and leaves of *Lantana camara* (L.), respectively. A total of 10 agromyzid species were identified. Identification keys and description for each species are provided. In the present study, the genera *Cerodontha* and *Calycomyza* were recorded for the first time from Karnataka.

## Studies on Insect Pest Complex of Watermelon *Citrullus lanatus* (Thunb.) and their Management

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THE present investigation on insect pests of watermelon, *Citrullus lanatus* (T.) and their management carried out under field conditions revealed that the major insect pests observed during cropping period were leaf miner, *Liriomyza trifolii* (Burgess); thrips, *Thrips palmi* (Karny); whitefly, *Bemisia tabaci* (Gennadius) and fruit fly, *Zeugodacus cucurbitae* (Coquillett) with mean populations of 3.89±1.88, 9.71±6.25, 3.67±2.77 and 4.91±5.92, respectively. These insect pests prevailed throughout the cropping period, while, red pumpkin beetle, *Raphidopalpa foveicollis* (Lucas) and leaf eating caterpillar, *Diaphania indica* (Saunders) were recorded as minor defoliators with mean populations of 0.29±0.34 and 0.27±0.31, respectively. Management of major insect pests with insecticides showed that cyantraniliprole 10.26 per cent OD was found to be superior over other insecticides against *L. trifolii* and *B. cucurbitae* with 79.78 and 48.68 per cent reduction over control respectively. Thiamethoxam 25% WG was effective against *B. tabaci* with 87.07 per cent reduction over control. While, two sprays of fipronil 5% SC was most effective against *T. palmi* with 76.18 and 76.89 per cent reduction over control during first and second sprays, respectively. Influence of usage of insecticides on natural enemies (spiders, coccinellids) and pollinators (Honey bees) were recorded. The results revealed that Spinosad 45% SC was the safest insecticide to natural enemies and pollinators compared to all the other insecticidal treatments. Crop sprayed with cyantraniliprole 10.26% OD recorded highest fruit yield among different insecticidal treatments with 51.83 t/ha. Whereas, the highest B:C ratio was found in fipronil 5% SC with 3.39.

## **Incidence Pattern, Source of Resistance and Management of Hispa Beetle, *Dicladispa armigera* (Olivier) in Rice**

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THE investigations on incidence pattern, source of resistance and management of hispa beetle, *Dicladispa armigera* (Olivier) in rice were carried out at College of Agriculture, V.C. Farm, Mandya, Karnataka, during 2021-22. The per cent leaf damage and incidence of adult hispa beetles were found higher when transplanting was done on 15<sup>th</sup> of August, followed by 1<sup>st</sup> of September along with a positive correlation with minimum temperature, afternoon relative humidity, rainfall and number of rainy days. While the maximum temperature, morning relative humidity and sunshine hours had a negative association. The field evaluation of 300 rice genotypes against hispa beetle resulted in identifying Madras sanna among landraces and BR-2655 among popular cultivars as resistant genotypes. The studies on the biochemical basis of resistance against rice hispa revealed that the amount of total sugars, reducing sugars, crude proteins and free amino acids were in higher amounts in susceptible genotypes. In contrast, a higher amount of total phenols and tannins were observed in all the resistant genotypes. Among the nutrient factors, nitrogen, phosphorous, magnesium, sulphur and iron were found to be positively correlated with hispa damage. While, potassium, calcium, zinc, copper, manganese and silicon showed a negative association. Among the seven insecticides evaluated carbosulfan 25 EC @ 2 ml/l, fipronil 5 SC @ 2.5 ml/l and acephate 95 SG @ 1.3 g/l were found highly effective against rice hispa beetle. However, acephate 95 SG @ 1.3 g/l was found superior in recording higher net profit and B:C ratio compared to other treatments.

## **Studies on the Biology and Substrate Preference of the Burrowing Cockroach, *Pycnoscelus surinamensis* (Linn.) (Blaberidae : Blattodea)**

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BIOLOGICAL studies were conducted on *Pycnoscelus surinamensis* revealed that the presence of four instars. The mean duration was 30.5±1.55, 31.5±1.55, 34.25±2.63 and 29±1.08 days, for the four instars respectively and takes 134.8±4.94 days to complete the life cycle. The adult longevity was 111.25±3.35 days. Morphometric studies shows that the antennal length, head width, body length, pronotum length and width increased from first to fourth instar and peaked at the adult stage, all the developmental stages well fit into Brooks-Dyar rule. The studies on parental care revealed that, the early instar nymphs were not able to survive in the absence of adult female indicating strong parental care in *P. surinamensis* and second instar nymphs have 1.5 times longer mean developmental time than other instars and recorded 207 days. *Pycnoscelus surinamensis* prefers substrates to live in, Cocopeat and vermiculite tested as best substrates and high mortality rates were found without substrate. The effect of sociality on survival rate of *P. surinamensis* revealed that, the treatment with only one individual has lowest survival *i.e.*, 11 weeks compared to the treatments with four, eight and twelve individuals per container survived up to 16 to 18 weeks. The composition of the nutrients in the blatticompost and vermicompost at 90DAF showed significantly higher nitrogen content in partially decomposed farm waste+cockroach (3.16%), higher phosphorous content in partially decomposed farm waste+earthworm (1.81%), higher potassium content in partially decomposed kitchen waste+cockroach (1.042%). Farm waste and kitchen waste provided to earthworm has more nutrient concentration and found less in comparison with earthworm.

## Validation of Suitable Genetic Loci as Targets for Genetic Population Control of Mango Fruit Fly, *Bactrocera dorsalis* (Hendel) (Diptera : Tephritidae)

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*BACTROCERA DORSALIS* (Hendel) (Diptera : Tephritidae) is a destructive pest causing tremendous economic losses. The present study was aimed at validation of genetic targets for genetic biological control of this notorious pest *via* CRISPR/Cas9 mediated genome editing. For efficient microinjection, factors affecting egg-laying like ovipositional substrate preference and time interval were optimized. The gravid females had preference for banana pulp as oviposition substrate and optimum number of embryos ( $66.50 \pm 1.91$ ) were obtained in 15 minutes. As microinjection causes physical injury to embryos, statistically significant lower hatching (8.40% against 26.18% in control) and survival (6.45% against 21.4% in control) rates were witnessed. Later, two genes, *yellow* and *white* crucial for melanin pigmentation and cuticle sclerotization were cloned and characterized. Off-target minimised guide RNAs for *yellow* (sg692 and sg915) and *white* (sg360) were synthesised *in vitro* and used for restriction assay. Site directed mutagenesis was attempted by microinjecting ribonucleo protein (RNP) complex into G<sub>0</sub> embryos. Consequently, *white* gene was knocked out. Thus, in contrast to blue-green eye in the wild flies, the mutants exhibited white-eyed phenotype with altered head spot patterns. Amino acids alignments also predicted significant differences in conformation (2D and 3D) of protein critical for its functional significance. Thus, our study has validated gene targets for CRISPR/Cas9 based genetic population control of *B. dorsalis*.

## Molecular and Metabolite Profiling of *Ustilaginoidea virens* (Cooke) Isolates, Understanding Host Pathogen Interaction, Epidemiology and Management of False Smut of Rice

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RICE (*Oryza sativa*) is one of the most important cereal crops consumed by more than half of the global population. A significant proportion of rice produced is lost each year due to false smut caused by *Ustilaginoidea virens*. Infection that occurs at the booting stage hinders spikelet development by filling its inside space with mycelia and spores. Phylogenetic analysis of partial ITS region sequence of 81 isolates collected from different rice growing regions of Karnataka revealed three distinct clusters. Cluster-I consisted 79 isolates, while clusters-II (132-BNK-KA-54 from Bannikallu) and III (158-ALV-KA-80 from Alnavar) had one isolate each. Host-pathogen interaction study conducted by inoculating *U. virens* to susceptible cv. GNV 10-89 and resistant cv. IR28, revealed a total of 4172 differentially expressed genes (DEGs) of which 1650 genes were up-regulated and 2522 down-regulated. Epidemiological studies were carried out during the two consecutive *kharif* seasons, 2019 and 2020 with two cultivars GNV 10-89 and KRH 4 and four different dates of sowing. Relative humidity (60.37 - 95.34%), temperature (15.14 - 30.86°C), evaporation (2.29 - 3.00 mm), rainfall (1.50 - 5.50 mm) and rainy days (0-1) played a dominant role in relation to disease development. Fungicides were more effective than the bio-formulations and bio-agents and spraying trifloxystrobin 25% + tebuconazole 50% WG at the booting stage was effective for the management false smut of rice.



## Virome Analyses in Chilli (*Capsicum annuum* L.) by Next Generation Sequencing (NGS)

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NEXT-GENERATION sequencing (NGS) based virome analyses of mRNA has become a liable approach for the detection of total viruses (including known and novel viruses) present in the plants. Chilli is an important vegetable and spice crop with ethnopharmacological importance and susceptibility to co-infection of several viruses. To profile the chilli virome, 19 different virus infected leaf samples from the chilli plants showing leaf curl, mosaic, shoe-string and vein banding symptoms were collected. Total RNA of collected 19 samples was pooled into a single sample and sequenced through NGS. The analysis of chilli mRNAome data revealed the presence of *Chilli leaf curl virus* (ChiLCV) and its associated alphasatellite, betasatellite, *Cucumber mosaic virus* (CMV), *Groundnut bud necrosis orthotospo virus* (GBNV), *Pepper cryptic virus-2* (PCV-2), *Pepper vein yellows virus-2* (PeVYV-2) and *Bellpepper alphaendorna virus* (BPEV). From the virus associated contigs, complete genome for ChiLCV, CMV, PCV-2, PeVYV-2 and BPEV and partial genome for alphasatellite, betasatellite and GBNV was reconstructed. Pairwise identity analysis showed that all the identified viruses showed >90 per cent nucleotide identity with their respective reference isolates available in the NCBI database. Recombination breakpoint analysis detected the intra and inter specific recombinants in ChiLCV, RNA1 of CMV, PeVYV-2 and BPEV. PCV-2, PeVYV-2 and BPEV are the first reports in India on chilli.

## Virome Profiling in Cucumber (*Cucumis sativus* L.) for Identification of Known and Novel Viruses

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THE viral diseases are major constrain in cucumber (*Cucumis sativus* L.) cultivation. Plant virus diagnosis is important for managing viral diseases. The advanced sequencing technology, Next generation sequencing (NGS) based virome profiling becomes common approach for reliable detection of viruses and viroids. Virus infected leaf samples collected during 2020-2021, total RNA extracted from these samples which were pooled together and used as input for mRNA sequencing. Pre-processed mRNAome data was subjected to *de novo* assembly (Trinity), such that reads were assembled into contigs. The standalone MEGABLAST of these individual contigs against reference viral genomes in NCBI, identified virus associated contigs. Five RNA viruses, *Cucumber mosaic virus* (CMV), *Tobacco streak virus* (TSV), *Cucurbit aphid borne yellows virus* (CABYV), *Cucurbit yellows stunting disorder virus* (CYSDV), *Melon yellow spot virus* (MYSV) and a DNA virus, *Tomato leaf curl New Delhi virus* (ToLCNDV) were identified from pooled sample. Mapping of contigs against reference viral genome through Bioedit and Clustal X, reconstructed the complete/near complete genome of identified viruses. The sequence analysis using sequence demarcation tool (SDT) study showed per cent nucleotide identity with other similar isolates of GenBank. Taxonomic position of identified viruses was elucidated based on phylogenetic analysis and ICTV threshold level criteria for strain and species demarcation. Further, recombination analysis using six methods integrated in RDP4 program indicated presence of only intra specific recombination events in RNA2 and RNA3 of CMV and RNA1 of TSV. Interestingly, CYSDV and MYSV infection on field cucumber is the first report of their natural infection from India.

## **Reaction of Popular Cultivars and Landraces of Rice Against Sheath Blight Caused by *Rhizoctonia solani* Kuhn. and its Management**

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SHEATH blight disease of rice, caused by *Rhizoctonia solani* Kuhn is one of the major disease in all rice-growing regions of the world. Considering the significance of this disease, the present study was conducted to screen popular rice varieties and landraces for resistance to sheath blight, biochemical associated with disease resistance, *in vitro* evaluation of fungicides, botanicals and field management of sheath blight disease by new generation fungicides. Out of 100 rice genotypes screened, based on PDI and AUDPC values, the genotypes were categorized into 5 groups *i.e.*, resistant (2), moderately resistant (38), moderately susceptible (33), susceptible (25) and highly susceptible (2). Further, to know the resistance mechanism, 34 selected genotypes were analyzed for biochemical constituents by using standard procedure and protocols and it was found that phenols (-0.79), total soluble sugars (-0.84), reducing sugars (-0.87), crude proteins (-0.84), tannins (-0.91), peroxidase (-0.67), polyphenol oxidase (-0.83) and phenylalanine ammonia lyase (-0.84) was negatively correlated with sheath blight disease severity and these biochemicals were higher in resistance genotypes compared to susceptible genotypes. The maximum inhibition of *R. solani* under *in vitro* was found with Tebuconazole 29.5 EC (100%) and Senna (82.22%) among fungicides and plant extract tested, respectively. The foliar application of Azoxystrobin 18.2 + Difenconazole 11.4 SC at 40 and 60 DAT was most effective in suppressing the disease which showed least disease severity (16.29% and 12.44%) with higher grain yield (5400 kg/ha and 4800 kg/ha) in the field experiment conducted during *kharif* 2021 and *summer* 2022, respectively.

## **Rain Water Harvesting and its Utilization on Growth, Yield and Quality of Dolichos Bean (*Lablab purpureus* L.) Varieties under Open Field and Polyhouse Condition**

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A field trial was conducted at Department of Horticulture, GKVK, University of Agricultural Sciences, Bangalore during *rabi* seasons of 2019-20 and 2020-21 with eight genotypes of dolichos bean (*Lablab purpureus* L.) to study their growth, yield and quality by adopting randomized complete block design with three replications under open field and polyhouse conditions. The study of pole genotypes under field conditions revealed significant differences with the genotype Chintamani local registering the highest plant height (344.50 cm), number of branches per plant (15), days to first flowering (73.17), pod length (6.27 cm), pod width (2.22 cm), ten pod weight (41.08 g) and number of pods per plant (107). Pod yield per plant ranged from 159.33 g to 389.67 g in different genotypes, the highest being observed in Chintamani local (13.89 t/ha) followed by IC-0623013 (11.32 t/ha) and IC-0623056 (10.37 t/ha). In comparison, the HA-4 variety (a spreading type) was earliest in flowering, green pod maturity (60 days) and with yield of 5.81 tonnes per hectare. No significant results were obtained in yield under polyhouse condition, however pooled data of two seasons for green pod yield per hectare ranged from 0.39 to 0.94 tonnes only with the highest being recorded in Chintamani local (0.78 t/ha) among pole types, while HA-4 variety registered green pod yield of 0.94 tonnes per hectare. Among quality parameters, higher protein content was recorded in Chintamani local (26.35 g/100 g) followed by IC-0623013 (25.67 g/100 g) and HA-4 variety registered 17.57 g per 100 g under open field condition. Under open field condition, cost benefit ratio with different genotypes ranged from 1:1.09 to 1:2.39, the highest being observed in Chintamani local (1:2.39). Molecular analysis indicated the existence of high degree variation among genotypes and these lines could be used as novel germplasm source for further crop improvement.

## Studies on Nutrient Formulations for Mini-Tuber Production of Potato through Aeroponic Technique

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AEROPONIC production of mini-tubers by using early generation planting material in controlled condition is a rapid multiplication method to get early generation, virus free quality seed tubers. The solution containing 50 per cent higher dose of all nutrients over and above the modified Hoagland solution recorded higher values for growth and nutrient uptake at 45 days after transplanting (DAT) during pre-tuberization period. Among two growing conditions (net house and greenhouse), higher day and night temperature with low light intensity accelerated the growth parameters in greenhouse, but higher temperature resulted in reduced tuberization. The higher photosynthetic partitioning favored by congenial environment in net house, recorded substantial mini-tuber production. With little variations in environmental conditions, potato seed tuber production was possible in *kharif* in net house (15.06 in *kharif* as against 22.94 mini-tubers per plant in *rabi*). The reduction of light by shade treatment adversely affected the plant growth, tuberization and nutrient uptake. Regulation of root zone temperature to optimum level resulted in reduced physiological stress on plants and recorded higher number of mini tubers per plant (24.39). The decreased dose of nitrogen application after 45 DAT till the end of crop duration resulted in triggered mini-tuber production (23.97). In subtropical condition like in Bengaluru higher number of tubers per plant was observed in Kufri Himalini (21.9) than in Kufri Jyoti (20.27). Lesser number of mini-tubers per plant (18.98) was recorded with the use of commercial water soluble fertilizers as compared to laboratory grade nutrient salts used in modified Hoagland solution (23.19 mini-tubers per plant).

## Effect of Season on Flowering, Oil Yield, Quality and Antibacterial Properties of Ylang-Ylang (*Cananga odorata* Hook. F. and Thomson)

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AN investigation was carried out at the Department of Horticulture, GKVK campus, University of Agricultural Sciences, Bangalore to study the effect of season on flowering, oil yield, quality and antibacterial properties of Ylang-ylang (*Cananga odorata* Hook. F. and Thomson). The experiment was conducted in two seasons (October-December and March-May) with six treatments in each season and three replications comprising 3 different distillation times and two flower stages. The maximum number of leaves per branch and per tree was found in season 1. The maximum number of flowers per branch (98.27) and per tree (1459.89) was observed in season 2. The highest weight of green flower (348.5 g) and yellow flower (249.78 g) per tree was also recorded in season 2. The maximum oil yield (13.91 ml) was found in season 2 (flowers harvested during the months of March-May). In season 1, the highest oil percentage of 1.87 per cent was found when the flowers were distilled at green stage at 12.00 noon. In season 2, the highest oil percentage recorded was 1.27 per cent with distillation of flowers in green stage at 12.00 noon. GC-MS analysis showed that the percentage composition of specific constituents of oil from the present study was comparable with first grade oil of Ylang-ylang as per Madagascar standards. The oil extracted from both the stages of flower showed inhibitory effect against human pathogenic bacteria, *Bacillus cereus*, *Bacillus subtilis* and *Escherichia coli*.

## Molecular Diversity Analysis of Fern Species of Agumbe Ghats of Karnataka using ISSR Markers

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A survey of fern flora of Agumbe Ghats of Karnataka was undertaken to access the diversity of ferns. The present study resulted in the collection of 12 species of pteridophytes belonging to 11 genera under 11 families. The herbaria were prepared for the present study and were deposited in Mahatma Gandhi Botanical Garden of GKVK, Bangalore. The slightly modified CTAB DNA extraction method with the addition of polyvinyl pyrrolidone (PVP), Phenol : chloroform : Isoamyl alcohol (PCI) yielded a good DNA concentration. 9 ISSR primers were selected to study the genetic diversity among 12 fern species. The annealing temperature for all ISSR primers was standardized by gradient PCR and the annealing temperature for all primers ranged from 42 to 56°C. ISSR fragments generated 32 to 66 bands per primer. A total of 389 polymorphic bands produced 100 per cent polymorphism per primer. The similarity coefficient between the species was within the range of 4 to 35 per cent. The dendrogram generated by ISSR markers revealed two major clusters, indicating that fern species have distributed based on frond shape, frond type, type of rhizome, habitat, stipe colour, texture. Based on molecular data the highest genetic similarity of 35 per cent was observed between *Pteris confusa* and *Pteris biaurita*. while *Tectaria coadunata* and *Adiantum philippense* showed the least genetic similarity index of 4 per cent.

## Performance of Different Varieties of Garden Peas (*Pisum sativum* L.) for Growth, Yield and Quality under Open Field and Shade House Conditions

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AN experiment was conducted at Department of Horticulture, University of Agricultural Sciences, GKVK, Bengaluru during *rabi* season of 2018-19 and 2019-20, with fifteen varieties of garden pea (*Pisum sativum* L.) to study their growth, yield and quality by adapting randomized complete block design with three replications under open field and shade house conditions. The study under open field condition revealed significant differences with Arka Karthik registering greater height of plant (86.11 cm), more number of branches per plant (14.88), length of internode (7.41 cm), number of pods per plant (20.29), length of pod (10.30 cm), wider green pod (2.66 cm), weight of pod (9.42 g), yield of pods per plant (185.80 g) and per hectare (217.75 q ha<sup>-1</sup>). Under shade house condition, Arka Apoorva registered significant vegetative growth, yield of pods per plant (92.62 g) and per hectare (157.50 q ha<sup>-1</sup>). With respect to post harvest quality parameters, under open field condition, higher protein content (25.18 %) and firmness (24.55 N) were registered with Arka Karthik and under shade house condition, higher protein content (23.31%) was registered with PSM-4. Yield of pods per plant exhibited significant and positive correlation with many of growth and yield attributes under open field and shade house conditions. Among 15 varieties of Garden Pea, highest Benefit : Cost ratio was registered with Arka Karthik (5.84) under open field condition and with Arka Apoorva (3.31) under shade house condition.

### ***In vitro* Culture of Ginger (*Zingiber officinale* Rosc.) cv. Rio-de-Janeiro**

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GINGER is one of the important spice and widely used all over the world for its unique taste and aroma. Ginger is conventionally propagated by the rhizomes. High seed rate, poor field establishment, slow growth rate and high risk of disease transmission through infected rhizomes resulted in limitations of the area expansion in the country or state. The present investigation was carried out through *in vitro* culture to obtain disease free quality planting material. The rhizome bud and shoot tips were used as explants. The surface sterilised explants were cultured on full strength MS media supplemented with different concentrations and combinations of growth regulators namely BAP, Kinetin and NAA. Better response was obtained when growth regulators were used in different combinations than individual. The combination of BAP 3.0 mgL<sup>-1</sup> and NAA 1.0 mgL<sup>-1</sup> produced maximum number of shoots (6.6) in shoot tip, which could be used for producing a greater number of planting material. Whereas, for rhizome bud used as explants BAP and NAA at 2.0 mgL<sup>-1</sup> and 0.5 mgL<sup>-1</sup>, respectively showed better response for shoot length (7.08 cm). The explants placed in combination of BAP and NAA (BAP 3.0 mgL<sup>-1</sup>+ 0.5 mgL<sup>-1</sup>, BAP 3.0 mgL<sup>-1</sup> + 1.0 mgL<sup>-1</sup>) have showed combined effect of both shooting and rooting. The regenerated shoots were transferred to the media with rooting hormones namely IBA and IAA at different concentrations and IBA at 1.0 mgL<sup>-1</sup> showed maximum root length (3.8 cm) and better response on root growth.

### **Evaluation of Cowpea (*Vigna unguiculata* L.) Genotypes for Growth, Yield and Protein Content for Vegetable Purpose**

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COWPEA is a leguminous crop, which is rich source of protein and presently it is cultivated as pulse crop rather than vegetable crop. Hence, the present study entitled 'Evaluation of cowpea (*Vigna unguiculata* L.) genotypes for growth, yield and protein content for vegetable purpose' was carried out at College of Agriculture, Hassan, University of Agricultural Sciences, Bangalore during *rabi* season of 2021-22. Experimental material consisted fortyfive genotypes with three checks and were evaluated following augmented design to generate and interpret the information of parameters of variability, heritability and genetic advance as per cent of mean to understand the concept of diversity in the genotypes. Mean performance showed that, among genotypes evaluated high yielding genotypes were NBC-51 (6.91 t/ha), IC-402159 (6.8 t/ha) and PKB-6 (6.74 t/ha). Apart from yield, genotypes which performed superior for protein content were PKB-6 (25.18 %) followed by IC-206240 (24.45%), NBC-19 (23.33%) and PKB-4 (23.29%) indicating that these genotypes can further be incorporated for trait specific breeding. High PCV and GCV were observed for parameters like leaf area (48.19%; 46.45%), seed germination (25.89%; 23.38%) and seed weight (23.11%; 22.88%). High heritability was observed for pod length, pod yield per plant, seed weight and seeds per pod. High genetic advance as per cent of mean were observed for leaf area, seed weight, seed germination and pod yield per plant. These results can be exploited for further crop improvement programme of cowpea for vegetable purpose.

## Standardization of Spacing and Fertilizer Dose for Statice (*Limonium sinuatum* Mill.) under Shade Net for Bangalore Condition

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STATICE is one of the important flower crop used as filler material in floral arrangements and as a dry flower in dry flower art. The panicles of statice are in high demand in the market, its cultivation is not popularized much and statice has no standard package of Agro-techniques. The field experiment with 12 treatments with 3 replications was laid out in factorial completely randomized design (FCRD) under 50 per cent shade net to know the effect of varied levels of fertiliser dose and spacing on growth, yield and quality of statice. Among all the treatments, 75 per cent fertilizer dose with spacing of 30 cm x 30 cm has recorded highest plant height (42.67 cm), whereas more number of leaves per plant (35.20), more plant spread (1255.76 cm<sup>2</sup>), least number of days taken for panicle initiation (94.33), 50 per cent bloom (127.67), 75 per cent bloom (133.33), longest panicles (103.67 cm), highest number of panicles per plant (12.33), maximum vase life (24.33), vase solution uptake (115.00 ml) and highest B:C ratio (7.01) were recorded in 125 per cent fertilizer dose with spacing of 30 cm x 30 cm. Maximum yield per square meter and maximum yield per hectare were recorded highest in 125 per cent fertilizer dose with spacing of 30 cm x 15 cm (205.26 and 20,79,100 panicles respectively). Similarly least number of days taken for shade drying of panicles (4.33) was recorded in 75 per cent fertiliser dose with spacing of 30 cm x 15 cm.

## Studies on Induced Mutations in *Zamioculcas zamiifolia* Engl. for Novel Plant Architecture

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*ZAMIOCULCAS ZAMIIFOLIA* Engl. (ZZ) is an ornamental foliage plant, recently introduced to the world of interior plant scape. The present study on inducing variability in ZZ plants using mutagens was carried out during the year 2021-2022 in shade net at the 'D' Block, University of Agricultural Sciences, GKVK, Bengaluru. Mature leaflets with rhizome initiated were irradiated with 10Gy, 12Gy, 14Gy, 16Gy, 18Gy and 20 grays of gamma irradiation in the first experiment and were treated with 0.06, 0.08, 0.10, 0.12, 0.14 and 0.16 per cent of EMS in the second experiment. In gamma irradiated treatments, LD<sub>50</sub> was observed at T<sub>6</sub> (18Gy). Early rhizome formation at 16.50 days after rhizome initiation (DAI), early root initiation at 4.80 days DAI, highest number of roots (4.90) at 120DAI, early new shoot bud initiation at 62.50DAI, maximum rhizome diameter of 1.62 cm at 90 DAI was recorded in untreated control mature leaflets. In EMS treatments, LD<sub>50</sub> was observed at 0.12% EMS (T<sub>5</sub>). Maximum days (149.50DAI) for new shoot bud initiation was noticed in T<sub>7</sub> (0.16%). In T<sub>4</sub> (0.10%), two new shoot bud initiation was noticed at 141.00 DAI. Formation of cotyledonary like leaves at 93.60DAI, maximum leaflet number (3.10), leaflet area (1.45cm<sup>2</sup>) and plant height (3.35cm) was recorded in control mature leaflet at 120DAI. Variations could not be recorded in any of the gamma irradiated treatments and EMS treatments with respect to leaf colour, leaflet variegation and leaflet shape due to detrimental effect of mutagens which was lethal and resulted in mortality of all the treated mature leaflets.

## Effect of Plant Spacing and Nutrient Levels on Growth and Yield of Red Cabbage (*Brassica oleracea* var. *Capitata* f. *rubra*)

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RED cabbage is one of the important exotic vegetable. To know the effect of plant spacing and nutrient levels on growth yield of red cabbage a field experiment with 9 treatments and 3 replications was laid out in factorial randomized block design (FRCBD), which was conducted at Department of Horticulture, University of Agricultural Sciences, GKVK, Bengaluru during the year 2021-22. Significantly highest number of leaves (28.93), maximum plant height (36.71 cm), maximum plant spread (68.23 cm), maximum head circumference (42.41 cm), maximum fresh weight of head (1421.80 g), highest head volume (1360.19 cc), maximum head diameter (13.54 cm) and maximum head height (13.93 cm) were recorded in plant spacing of 45 cm x 60 cm with 125% RDF. Minimum days taken for head initiation (45.60) and compact head (0.38) was observed in plant spacing of 45 cm x 30 cm with 75% RDF. Highest yield per hectare (74.43 t) and highest cost-benefit ratio (2.18) were recorded in plant spacing of 45 cm x 30 cm with 125% RDF. Maximum available nitrogen (252.71 kg/ha), phosphorus (75.74 kg/ha) and potassium (140.81 kg/ha) in soil and maximum uptake of nitrogen (375.93 kg/ha), phosphorus (35.94 kg/ha) and potassium (185.11 kg/ha) by the plants was recorded in plant spacing of 45 cm x 60 cm with 125% RDF.

## Study on Effect of Different Organic Manures along with Microbial Consortia on Growth, Yield and Quality of Kalmegh (*Andrographis paniculata* NEES)

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KALMEGH has wide range of medicinal and pharmacological applications. The active ingredient andrographolide is most important bioprotectant. An experiment entitled 'Study on effect of different organic manures along with microbial consortia on growth, yield and quality of kalmegh (*Andrographis paniculate* Nees)' was conducted in Department of Horticulture, CoA, University of Agricultural Sciences, GKVK, Bengaluru during 2021-2022. The experiment was laid out in randomised complete block design with thirteen treatments replicated thrice. Significantly higher plant height (34.30 cm) was observed with the application of 100% RDF through vermicompost. Maximum plant spread (2209.6 cm<sup>2</sup>) and higher OC (0.53%) status of soil after harvest was recorded in the treatment consisting of 50% RDF through FYM + 50% RDF through neem cake + microbial consortia. Application of sheep manure in combination with neem cake resulted in higher number of branches per plant (14), higher number of leaves per plant (152.6), highest fresh weight (56.60g), highest dry weight (21.49 g), highest root to shoot ratio (0.59), highest fresh herbage yield (6.60 kg/ plot and 116 q/ ha), highest B:C ratio (1.66). Higher nitrogen (175.6 kg h<sup>-1</sup>) status of soil was recorded with the application of 100 % RDF through poultry manure. Higher andrographolide content of 2.05 per cent was noticed in the treatment with the use of 50% RDF through vermicompost + 50% RDF through neem cake + microbial consortia, highest count of soil microbial load was recorded in treatment consisting of 50% RDF through poultry manure + 50% RDF through neem cake + microbial consortia.

## **Economic Impact of Urbanization on Irrigation Water Productivity, Migration and Livelihood Security of Households in Rural-Urban Interface of North Bengaluru**

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URBANIZATION and migration in India are influenced by major differences in the patterns of social and economic development. Since urbanization has been identified as a key global concern in the coming decades, research on the rural–urban interface has expanded. The present study is a systematic approach to investigate how urbanisation, migration and irrigation water productivity affect livelihoods of farm households in Bengaluru. The villages around the Bengaluru city were selected randomly and categorized into three gradients like urban, transition and rural gradients. The sample frame consisted of 260 farm households representing 60 from urban and 100 each from the transition and rural gradients. The result showed that highest rate of migration was in rural gradient with 52 per cent of households migrating followed by urban (46.67 %) and transition (35 %) gradient. The rural-urban migration pattern was the most important and dominating one among the four migration streams found in the study. In rural gradient, the migration was higher (41.76 %) because of prevalence of more wages at the destination than in origin followed by availability of better jobs (31.87 %). The result of the probit model showed that, among significant factors, education, household size, non-farm income and outstanding debt of the family had positive effect while, farm income alone had negative effect on the probability of household migration. Calculations on irrigation water productivity of major crops showed that the irrigation water productivity of tomato was higher compared to ragi, carrot and mulberry. The findings of the fractional probit model demonstrated that urban households had a positive and substantial effect on the livelihood security. The livelihood security index showed that urban households (0.61) had greater livelihood security than transition (0.55) and rural (0.52) households.

## **Economic Impact of Agricultural Research by University of Agricultural Sciences, Bangalore in Sugarcane and Paddy in Southern Dry Zone of Karnataka**

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THE study on economic impact of UAS-B research in sugarcane and paddy crops was conducted in Southern Dry Zone of Karnataka. The UAS-B released VCF-0517 variety of sugarcane and KRH-4 paddy hybrid were selected for in-depth analysis. The data were collected from 200 respondents and analyzed using economic surplus, TFP, partial budgeting, economic surplus model and production function analysis. The results of the study revealed that farmers realized 23.53 per cent higher returns from VCF-0517 sugarcane variety (Rs.472500) than check variety CO-86032 (Rs.382500). Similarly, farmers growing KRH-4 paddy have gained 15 per cent higher net returns (Rs.13,408) over check variety Meenakshi. Farmers growing UAS-B varieties were found to be technically, allocatively and economically more efficient in resource use than their counterparts growing check varieties. The economic surplus due to VCF-0517 sugarcane variety in the study area was estimated at Rs.5477.02 crores (2006 to 2020) with relatively higher producer surplus (57.24 %). The economic surplus due to KRH-4 paddy variety was Rs.65.45 crores (2003 to 2020) of which the consumer's surplus was higher (90.70 %). The TFP for sugarcane has increased over the years from 0.93 during 2001 to 2.03 during 2018 with mean TFP score of 1.608 and that for paddy, it has increased from 1.023 during 2001 to 1.648 during 2018 with mean TFP score of 1.209. The coefficients for research investment (0.745) and number of processing mills (0.530) were significantly influencing sugarcane productivity. The investment of one rupee in sugarcane research generated income of Rs.18.48 indicating substantial rate of returns to research investment. Hence, more funds can be allocated for research in these crops and farmers can be encouraged to adopt more of UAS-B varieties to achieve better resource use and augment their farm income.



## An Economic Analysis of GI Tagged Mysore Betel Vine Cultivation in Karnataka

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THE present study, based on primary data from 60 growers comprising of 30 farmers each growing Mysuru betel leaf and Ambadi betel leaf, 20 market intermediaries and 30 consumers was conducted in Mysuru district during 2020-21. Results revealed that awareness about Mysuru betel leaf having GI Tag was less among Mysuru betel leaf growers (43.33%), Ambadi betel leaf growers (16.66%) and consumers (20%). Consumers preferred Mysuru betel leaf over Ambadi leaf because of taste and spicy nature and not because of GI tag. The cost of cultivation of Mysuru betel vine (Rs.3,66,896/ac) was more than Ambadi betel vine (Rs.3,51,339/ac). Mysuru betel leaf growers realised 23.92 per cent higher net returns due to higher price fetched by Mysuru betel leaves (Rs.3,071/pendi) than Ambadi betel leaves (Rs.2,683/pendi). The resource use is revealed that for both the varieties, the coefficients for planting material, human labour and FYM were significant, while coefficients for fertilizers (-0.102) and plant protection chemicals (-0.002) were negative but were not significant in Ambadi betel vine cultivation. Among different marketing channels, Channel-III (Farmer-Commission agent cum trader-Wholesaler cum retailer-Consumer) was found to be the most efficient channel with higher producer's share in consumers rupee for both Mysuru (57.12%) and Ambadi leaf (55.44%) than the other channels. Labour scarcity and price volatility were the major constraints reported by betel leaf growers. Further, the study highlights the need to educate the growers about GI tag and the market authorities to check malpractices of intermediaries in the market.

## Impact of Covid-19 Pandemic on Farm Income of Vegetable Producers in Terai Region of Nepal

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THE present study analysed the impact of COVID-19 pandemic on farm income of vegetable producers in Terai region of Nepal. The study was undertaken in Chitwan district during November, 2019. Data was collected from 90 vegetable growers and analysed using descriptive statistics, costs and returns analysis, production function and decomposition analysis. From the decomposition analysis, it was found that gross returns from the selected vegetables (tomato, cucumber and bitter gourd) production was reduced by 85.65 per cent during COVID period which led to the reduction in household income by 59 per cent. Net returns from tomato cultivation were reduced by NPR 2.06 lakhs (-93.77%) which is attributed to the reduction in its yield by 10.68 per cent, market price by 46.77 per cent and inflated cost of cultivation by 10.98 per cent. Similarly, net returns from cucumber and bitter gourd cultivation were lowered by NPR 1.97 lakhs (-130.55%) and NPR 1.16 lakhs (-99.56%) owing to the reduction in market price by 61.76 per cent and 39.32 per cent. Major problems faced by the respondent farmers during COVID time were pest and disease control (91.11%), marketing of the produce (88.89%) and timely unavailability of quality seeds (82.22%). Pandemic situation was quite distressful for both farmers' and the Government. Though, in the later stage, the Government relaxed certain restrictions on the operations of agri-business the imposition of lockdown has already damaged whole agricultural industry. This study stresses the need for building IT and logistical infrastructure to facilitate smooth transaction in agricultural industry.

## Value Chain Analysis for Robusta Coffee in Kodagu District of Karnataka

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THE study was conducted on value chain analysis for robusta coffee in Kodagu district of Karnataka. Data was collected from 30 coffee growers, five traders, five curing works, five multi-national companies, five local merchant exporters and 120 coffee consumers and analysed using cost and returns analysis, value chain mapping, descriptive analysis, marketing channel, price spread and producer share in consumer rupee. From cost and return analysis of growers, it was found that labour cost accounts for more than 65 per cent of the total cost and rest was material costs. The value chain mapping analysis indicated that the main actors in the coffee value chain are growers, traders, curers, multinational companies (MNCs) and exporters. Governance structure between the grower and downstream actors was market form of governance with limited coordination in terms of quality and specifications in the coffee value chain. In robusta cherry coffee, price spread and producer share in consumer rupee were Rs.85.10 per kg and 44.41 per cent, respectively, while in case of robusta parchment price spread was Rs.62.95 per kg and producer share in consumer rupee was 65.96 per cent. It was further found that, 80 per cent of the respondents were aware of arabica and robusta coffee. Considering the low share of producer in the consumer rupee and high price spread, study stresses the need for implementing integrated digital platform like block chain-based market place for all the stakeholders involved in the coffee value chain.

## Impact of Farm Mechanization on Farm Income – An Economic Analysis in Mandya District

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THE study used data from 60 farmers and 4 custom hiring centres (CHCs) from Mandya district. Data were analysed using descriptive statistics, growth rate analysis, DEA and partial budgeting. Results on growth rate analysis indicated positive and significant growth in consumption of power operated farm machineries and implements, mechanical and total power availability in Mandya and Karnataka state while hand operated and animal operated machineries decreased. Higher proportion of farming operations were mechanized in paddy (49%) compared to ragi (26%). The comparison of net returns between farm categories revealed that farms with higher level of mechanization (HMFs) realised additional returns of Rs.7955 (Paddy) and Rs.2122 (Ragi) over farms with low level of mechanization (LMFs). Paddy cultivating HMFs were found to be more efficient both technically (97.30 %) and economically (80.10 %) than farmers with LMFs (93.60 and 58.50 %). Similarly, in ragi crop also, the farmers with HMFs were technically (97.20 %) and economically (85.40 %) more efficient than farmers with LMFs (91.80 and 78.40 %). The performance analysis of CHCs revealed positive impact on farm mechanization. However, small size of holding, undulated land and high initial investment requirement were the major problems coming in the way of practicing farm mechanization on large scale. CHCs were faced with the problem of lack of skilled labour in managing farm machineries and under-utilisation of available machineries due to poor awareness about CHCs among farmers.

## **Economics of Hybrid Tomato Seed Production in Karnataka**

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THE study was taken-up to examine costs and returns, technical efficiency, marketing channels and constraints faced by farmers and seed company in tomato hybrid seed production. Data were collected from hybrid tomato seed growers and commercial tomato growers 30 each, 5 seed production companies, 5 distributors, 20 retailers and 30 in Karnataka during 2020-21. Findings of the study revealed that the per acre cost of cultivation of hybrid tomato seed production was Rs.3,77,123 and gave a net profit of Rs.3,78,799. The seed growers margin was Rs.2 for every rupee of investment in seed production. Regarding technical efficiency, 66.66 per cent of tomato farms performed at optimum scale. Two major marketing channels were observed in tomato seed marketing *viz.*, Channel I (Seed company- Distributors- Retailers) and Channel II (Seed company- Retailers) for hybrid tomato seeds. The seed producers share in tomato growing farmers rupee in both channels was 12.74 per cent. Relatively higher proportion (40%) of the commercial tomato growers preferred hybrid seeds over other segments. Higher disease and pest attack was the major production constraint, while lack of technical knowledge of seed extraction was the major processing constraint and grading of seeds was major marketing constraint for growers. Mixing of low grade with high grade seeds was the major problem reported by the firms. Further, study highlights the need to educate the growers about maintaining quality in seed production and strengthening of development of disease resistant hybrids as the research and development priority for seed industry.

## **A Study on the Impact of Meeting Urban Domestic Water Requirements with Rural Groundwater Resources on its Sustainability and Farm Income in the Eastern Dry Zone of Karnataka**

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THE study was conducted in Bengaluru Rural district of Karnataka. A total sample of 80 farmers consisting of 40 farmers not selling groundwater (GNF) and 40 farmers selling groundwater to meet urban domestic needs (GSF) were selected. The number of GSF and water sales increased between 2016 and 2019 in the entire district. The average water extracted per farm was higher among GSF (162 acre inches) than among GNF (129 acre inches). The GNF generated 62 per cent of their total income from agriculture while 66 per cent of the income of GSF was from selling water in 2019. The annual irrigation cost per acre was higher among GSF (Rs.16,660) than among GNF (Rs.11,759). The cost per acre inch of groundwater used for farming by GNF was Rs.529 and was Rs.762 for farming and Rs.847 for water selling by GSF. The economic efficiency of groundwater usage in farming by GNF was Rs.1,691 per acre inch, which was less than that by GSF (Rs.1,862 per acre inch). The GSF, even though were economically efficient in using groundwater for both farming and selling, over-used the resource by over-exploiting the water resource for selling which affected the sustainability of groundwater. Over-extraction of the groundwater in excess of its natural restoration, led to depletion of the water table in the area causing bore-well failures thereby, reducing economic life of the bore-wells. Therefore, to reduce the stress on groundwater resources, water saving technologies need to be developed and popularized in all the dwelling places in both urban and rural fronts.

## **Impact of Encroachment of Irrigation Tanks due to Urbanization on Farming in Eastern Dry Zone of Karnataka -An Economic Analysis**

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THE study was undertaken in Bengaluru rural and Ramanagara districts. Primary data was collected from 90 farmers under three scenarios viz., Encroached tank area (ETA) comprising Thymagondlu Hirekere tank of Nelamangala taluk in Thymagondlu hobli having urban influence, Revived tank area (RTA) considering Chikkamaranahalli tank of Nelamangala taluk where encroachers were evicted and Encroachment free tank area (EFA) namely Pura Bhargavathi and Honnapura tanks of Magadi taluk where tank irrigation is practiced. GIS and remote sensing were used to find the change in water body area and land use pattern in ETA and RTA. Natural resource costing was used to analyze irrigation cost. The results of GIS data for the years 2011 and 2020 showed that majority of the water body area (98.34%) around Chikkamaranahalli tank region had not changed, while in Thymagondlu Hirekere tank region, more than half of the tank area (65.34%) had changed to other land use classes. Higher capital investment on borewells resulted in higher annual cost of irrigation (Rs.82,771) and cost per acre inch of groundwater in ETA (Rs.831). Frequent borewell failures and increased depth of borewells resulted in increased negative externality in ETA (Rs.20,465) compared to RTA (Rs.13,849) and EFA (Rs.7,301). Income obtained per acre was observed to be Rs.1,14,885, Rs.1,29,532, Rs.1,58,372 in ETA, RTA and EFA, respectively. Therefore, efforts need to be made by the concerned authorities to prevent encroachment of tanks and defunct tanks need to be rehabilitated to increase the groundwater table.

## **Impact of Climate Change on Land Use and Livelihood in Chikkaballapura District - An Economic Analysis**

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AGRICULTURE is dependent on persisting weather conditions. Impacts of climate change need to be understood to work out strategies to mitigate effects of climate change. The study was undertaken in 2020-2021 to know the influence of climate change on land use and livelihood in Chikkaballapura district. Study used both primary (2019-2020) and the secondary data obtained from 2007-2017 for cropping pattern, 1980-2017 rainfall and temperature from 2001-2020 and land use from 2001-2017. Through purposive sampling 60 farmers from Shidlagatta taluk and 60 farmers from Bagepalli taluk were selected for primary data collection. Analysis showed that rainfall distribution pattern is decreasing over the years with more fluctuation and temperature is increasing over the year which indicates that climate change is having negative impact over the years. In both the taluks. Month wise SPI results showed more variation from June to October. (Cropping season) and STA results showed more variation from March to May. In Chikkaballapura district, rain fed crop yields are unstable (CV >30 %) in both ragi (30.96 %) and ground nut (34.14 %) indicating that climate change (rainfall) is having a significant detrimental impact on the yields. The cost of cultivation of ragi per acre in Shidlagatta is Rs.21624 and the negative net returns obtained were Rs. 2219. The cost of cultivation of groundnut per acre in Bagepalli was Rs.28181 and the net returns obtained was Rs. 12197. Farmers opined that providing early warning through weather forecasting is important to overcome negative climate change impact. It is found that both the sample farmers use traditional knowledge in agricultural and livestock management to mitigate the impact of climate change.

## An Economic Impact Assessment of Supplying Treated Sewage Water to Irrigation Tanks in Chikkaballapura District under Hebbala Nagavara Valley Project

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THIS study endeavours to investigate the economic impacts of Hebbala Nagavara (HN) valley project on agriculturists of Chikkaballapura district. The required data was collected from 45 farmers each from Chikkaballapura (HNVP) and Shidlaghatta (NHNVP) taluk. It was found that cropping intensity (169.92%) and crop diversity (0.89) were higher in HNVP area. Due to this project, the number of operational wells in the HNVP area rose from 58 to 73 whereas, water yield got increased by 21.32 per cent. Cost per acre inch of groundwater in the HNVP area was lesser by 9.48 per cent as compared to NHNVP area (Rs.993.52). The negative externality per well in HNVP area was (Rs.32,134.30) 57.99 per cent less than NHNVP area (Rs.50,768.02). The HNVP farmers realized a yield of 170.20, 234.38 and 11.83 quintals/acre and a net income of Rs.3,64,274.89, Rs.1,90,722.08 and Rs.6,319.38 per acre of rose, tomato and ragi, respectively, which is higher than NHNVP area. HNVP farmers used 11.26 and 13.01 per cent more water in rose and tomato cultivation, respectively, than NHNVP farmers, resulting in poorer AWUE. The annual income of HNVP farmers (Rs.8,12,936.25) was higher than NHNVP farmers (Rs.6,90,961.30). The partial budgeting analysis revealed that, net additional benefit of Rs.70,841, Rs.47,063 and Rs.1,586 was realized by HNVP farmers from rose, tomato and ragi, respectively. The PCA results indicated that the positive externalities (0.66) outweighed the negative externalities (0.22). Hence, projects of this kind would help to increase water table, farmer's income and livelihood if implemented with more care.

## An Analysis of Behavioural Dimensions on Groundwater Conservation Practices by Farmers of Eastern Dry Zone of Karnataka

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THE study was undertaken to measure the attitude, knowledge and extent of adoption of various groundwater conservation practices by the farmers. Data was collected from 180 farmers from Kolar and Chikkaballapur district of Eastern dry zone of Karnataka using the structured pre-tested interview schedule. Standardized the scale to measure the attitude of farmers towards groundwater conservation practices. Seven factors of groundwater conservation practices such as socio-economic, environmental, agronomic, quality factors, management, institutional, future availability factors were taken to measure the attitude of the farmers towards groundwater conservation. The results revealed that the marginal farmers have shown less favourable (40.00 %), small farmers have expressed more favourable (43.33 %) and large farmers have expressed equal per cent of favourable and more favourable attitude (38.33 %) towards groundwater conservation practices. The marginal and small farmers have higher knowledge (38.33 and 41.67 %), while the large farmers have medium (40.00 %) level of knowledge on groundwater conservation practices. Various groundwater conservation practices taken into consideration in the study are *in-situ* conservation practices, *Ex-situ* conservation practices, Micro irrigation techniques and groundwater quality management practices. Marginal farmers were low adopters (36.67 %), small farmers have medium adoption level (41.67 %) and little more than one third of the large farmers have medium level of adoption (36.67 %) of groundwater conservation practices. Constraints like unprecedented monsoon leads to less scope to collect rainwater and lack of appreciation and incentives for using water judiciously are majorly expressed by farmers in adopting groundwater conservation practices in their field. Government should come up with initiatives keeping in view of demand side of Groundwater, Tank irrigation needs more concentration are the suggestion expressed by majority of the farmers to improve the groundwater source.

## **Impact of Migration on the Livelihood Status of the Migrated Labourers in Coffee Plantations of Kodagu and Chikkamagaluru Districts of Karnataka State**

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THE present investigation was carried out during the year 2020-21 in Kodagu and Chikkamagaluru districts of Karnataka state with a sample size of 180 migrant farm labourers. Simple random sampling technique was adopted to select the respondents and the data was collected with the help of structured and pre-tested interview schedule. In the present study the livelihood status of the migrant farm labourers was studied as a collective outcome of the five livelihood factors *viz.*, human factor, physical factor, social factor, financial factor and natural factor and the results depicted that nearly half of the migrant farm labourers had partially improved their level of livelihood status, followed by 19.45 per cent of the respondents who had improved livelihood status. Further, dimensions like nature of work (0.88), remigration intention (0.87), place of migration (0.85), migration network (0.82) and nature of migration (0.80) had great influence on their migration behaviour. As a result, majority of the migrant farm labours in Kodagu (46.67 %) and Chikkamagaluru (51.11 %) exhibited medium level of migration behaviour. Small landholding (85.00 %) followed by unstable income (78.33 %), lesser employment (71.67 %) and debt (70.56 %) were the major push factors. Whereas, higher wages (96.11 %), availability of employment opportunities (89.44 %), better social linkage (65.56 %) and better standard of living (65.00 %) were the key pull factors. The major constraints faced by the migrant farm labourers were language barrier (81.11 %) followed by difficulty in mixing with locals (68.89%) and delayed payment (54.44 %). Children's higher education has to be supported by the government (67.22 %), followed by formal structure to identify the migrant family and to avail benefits (63.89 %) were the major suggestions to improve their livelihood status in a sustainable way.

## **Study on Farmers Knowledge and Attitude towards Pradhan Mantri Fasal Bima Yojana in Tumkur District of Karnataka**

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THE present study was conducted during the year 2020-21 in Tumkur district of Karnataka. A total sample of 120 farmers comprising of 60 beneficiaries and 60 non-beneficiaries were randomly selected from 6 villages of Pavagada and Sira taluk. Data was collected using pre-tested interview schedule. The results of knowledge level of beneficiaries revealed that about 53.33 per cent had medium level of knowledge whereas, 43.33 per cent of non-beneficiaries had low level of knowledge. With respect to overall attitude of beneficiaries revealed that 48.33 per cent had favourable attitude whereas, 46.67 per cent of the non-beneficiaries had less favourable attitude. This clearly indicates that beneficiaries had more knowledge and favourable attitude than non-beneficiaries. Variables like education, annual income, land holding, farming experience, risk orientation and economic motivation were significant at five per cent level. Major problems faced by beneficiaries were delay in getting the claims, inadequate compensation and official bias in loss assessment. Whereas, major reasons for not availing benefits by non-beneficiaries were cut-off date problem to fill the application and to pay the premium amount, lack of awareness about the PMFBY and due in submission of appropriate documents. The major suggestions given by beneficiaries were dispersal of claim before starting of the next season, more number of trainings need to be organized on PMFBY and information should reach at the ground level in order to avail the benefits from PMFBY.

## **Impact Analysis of Pashu Bhagya Scheme on Livelihood Status of the Farmers in Vijayapura District of Karnataka State**

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THE present study was carried out to analyse the impact of Pashu Bhagya Scheme on livelihood status of farmers at Vijayapura district of Karnataka state during 2020-2021. A total sample of 120 farmers, comprising 90 beneficiaries and 30 non-beneficiaries were randomly selected from 6 villages of Vijayapura and Indi taluks. Data was collected through personal contact method using structured interview schedule. Results of the study revealed that beneficiaries (51.11 %) and non-beneficiaries (53.33 %) had medium and low level of livelihood status, respectively. Z- test results indicated significant difference in livelihood status (1.73\*), assets (1.74\*), capabilities (1.85\*) and food and nutritional status (1.67\*) of beneficiaries and non-beneficiaries. About 56.67 per cent of beneficiaries purchased cow/ buffaloes, 43.33 per cent claimed sheep/ goat units and 75.55 per cent attended the training programme under the scheme. The livelihood status of beneficiaries was significantly and positively associated with education, livestock rearing experience, scientific orientation, risk orientation, achievement motivation, economic motivation, credit orientation, extension participation, land holding, training received and cosmopolitanism at five percent level. Major problems faced by the beneficiaries were inadequate support received from the government organizations and departments, numerous and lengthy procedures to avail benefits. Lack of awareness about the scheme and inadequate information received from officials were the reasons for which the respondents were not able to get benefits from the scheme. Major suggestions given by respondents were to conduct training programs on management of diseases, digital technology to improve skill and knowledge of farmers and efficient extension advisory services.

## **Comparative Analysis of Adoption Level and Economic Performance of Chrysanthemum Growers in Tumakuru District of Karnataka**

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THE study was conducted in Tumakuru district of Karnataka state during 2020-2021 to analyze the adoption level and economic performance of chrysanthemum growers. Out of ten taluks in the district, two taluks namely Tumakuru and Koratagere were selected based on the criteria of maximum area under chrysanthemum cultivation. A total of 120 chrysanthemum growers were selected by using simple random sampling technique. Personal interview method was used to collect data and appropriate statistical tools were applied to analyze the data. Among overall growers half (50.83 %), 26.67 and 22.50 per cent of the chrysanthemum growers belonged to medium, low and high level of adoption, respectively. Among overall growers less than half (42.50 %), 30.83 and 26.67 per cent of the chrysanthemum growers belonged to low, medium and high level of economic performance, respectively. Variables such as education, annual income, extension agency contact, extension participation, cosmopolitanism, mass media, innovativeness, risk orientation, management orientation and knowledge level were found to have significant association with the adoption level and economic performance of chrysanthemum growers. The major problems related to chrysanthemum production were high cost of planting materials followed by non-availability of credit in time, non-availability of labours and low price were major financial, labour and marketing constraints. Majority of the chrysanthemum growers expressed, timely and adequate information regarding availability of inputs, prices and arrivals, providing suitable market information, infrastructure, transportation and promoting labour saving technologies like mulching, drip irrigation, harvesting as major suggestions.

## Analysis of Agri Startup Ecosystem in Karnataka

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STARTUPS play a key role in promoting innovation in a society. It is well known fact that startups are small companies but they play a significant role in the economic growth. The study was conducted in four southern districts of Karnataka state viz., Bengaluru urban, Bengaluru rural, Tumakuru and Mysuru during 2020-2021 to analyze the Agri startup ecosystem in the state. Twenty eight entrepreneurs from Bengaluru urban, 13 from Bengaluru rural, five from Tumakuru and four from Mysuru district were selected across different sectors to constitute a total sample of 50 entrepreneurs. The findings revealed that 72 per cent of the agristartup entrepreneurs exhibited high level, followed by 18 per cent and ten per cent of the respondents had medium and low entrepreneurial behaviour, respectively. The study also enunciated that education, specialization in higher studies, rural urban background, entrepreneurial experience, trainings undergone, volume of business, mass media exposure, social participation, source of finance and market orientation had contributed significantly towards entrepreneurial behaviour. The R<sup>2</sup> value indicated that all the 16 independent variables had contributed to the tune of 74.5 per cent of variation in entrepreneurial behavior. Tedious registration and licensing procedures (88 %), lack of early stage funding (84 %) and inadequate market information (76 %) were the major constraints faced by entrepreneurs during establishment and operating their startup.

## Knowledge, Adoption and Perception on Climate Smart Technologies by Small and Big Farm Redgram Growers in Southern Karnataka

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THE study was conducted in Magadi taluk of Ramanagara and Sira taluk of Tumakuru districts of Southern Karnataka during 2020-2021 to analyze the knowledge, adoption and perception on climate smart technologies by small and big farm redgram growers. A total of 120 (60 from each district) redgram growers were selected by using simple random sampling technique. Personal interview method was used to collect data and appropriate statistical tools were applied to analyze the data. Cent per cent of the respondents had correct knowledge on redgram varieties recommended by UAS-B. Majority of the respondents possess incorrect knowledge on use of 1% KCl (91.67%). 95.83 per cent adopted the varieties recommended by UAS-B, among the varieties majority of the redgram growers adopted BRG1 variety. 75 per cent and 66.67 per cent respondents are taken up earthing up and nipping technologies. Cent per cent of redgram growers not adopted transplanting technology. Majority strongly agreed earthing up helps to conserve water (75.83%). Variables such as education, annual income, risk orientation, mass media exposure, scientific orientation, Innovative proneness, cosmopolitaness and economic motivation were found to have 5 per cent significant association with the knowledge, adoption and perception of redgram growers. Farming experience, Extension contact and Extension participation shows 1 per cent significant association with the knowledge, adoption and perception of redgram growers. The major constraints related to redgram growers were lack of knowledge about climate smart technologies. Majority of the redgram growers expressed conducting training programmes on climate smart technologies as major suggestions.



## **Entrepreneurial Behaviour of Women Producing Value-Added Products of Millets in Bengaluru Rural and Urban Districts**

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THE study was conducted in Bengaluru rural and Bengaluru urban districts of Karnataka state during 2020-21 to analyze the entrepreneurial behaviour of women entrepreneurs actively involved in producing value-added products of millets. From each district, thirty women entrepreneurs were randomly selected constituting a sample of 60 respondents for the study. Personal interview method was used to collect data and appropriate statistical tools were applied to analyze the data. The findings revealed that higher percent of the women entrepreneurs were having medium level of innovativeness (38.33%), decision making ability (50.00%), level of aspiration (40.00%), entrepreneurial knowledge (40.00%), management orientation (50.00%), leadership ability (40.00%), competition orientation (36.67%) and self-confidence (50.00%) followed by high level risk orientation (36.67%), achievement motivation (53.33%), scientific orientation (35.00%) and credit orientation (50.00%). Most of the women entrepreneurs (35.00%) belong to medium level of entrepreneurial behaviour. Variables such as annual income, mass media exposure, financial support, education and extension participation had significant association with entrepreneurial behaviour of women entrepreneurs. Major constraints faced by women entrepreneurs were securing working capital, ineffective government consultation services, competition from well-established and large units, non-availability of raw materials and health problems. Interest-free credit, necessity for improvement in market intelligence and market facilities and organizing effective training programmes on millets are some of the major suggestions given by the women entrepreneurs.

## **A Critical Analysis of Knowledge Management Behaviour of Arecanut Growers in Davanagere District of Karnataka State**

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KNOWLEDGE management is concerned about approaches to exchange information among the individuals who can foster it and the people who can utilize it. The study was conducted in Davanagere district of Karnataka state during 2021-22 with sample size 90 to know the knowledge level and knowledge management behaviour of arecanut growers. The data was collected through personal interview method and analyzed by using suitable statistical tools. The findings of the study revealed that 41.00 per cent of respondents had medium level of knowledge about recommended arecanut production technologies and 41.10 per cent belonged to medium knowledge management behaviour category. In case of knowledge management dimensions respondents belonged to medium knowledge acquisition (43.33%), medium knowledge retention (46.67%), medium knowledge retrieving (56.67%), medium knowledge evaluation (48.89%) and medium knowledge dissemination (42.22%) category. Profile characteristics of respondents uncover that, among respondents, (46.67%) belonged to the middle age (35-50 years), (27.75%) educated up to high school (8 to 10<sup>th</sup> std), (41.11%) medium land holding (5.01-10.00 acre), (42.22%) medium annual income group (Rs. 7,80,354-16,17,201), medium extension participation (38.89%), medium mass media participation (44.44%), high level cosmopolitaness (71.11%), high risk orientation (53.33%), low credit orientation (45.56%). Out of eighteen independent variables, three variables namely, land holding, annual income and credit orientation showed non significant relationship with knowledge management behaviour. Majority (95.55%) expressed limited exposure visits and 96.66 per cent of respondents suggested to provide need based technical guidance from agricultural/ line departments. The aforementioned findings revealed the necessity of specialists working diligently and effectively at the grassroot level.

## **Impact of Soil Health Card Scheme on Enhancing Farm Productivity and Farmers Income in Bagalkot District of Karnataka – An Economic Analysis**

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THE study was undertaken in Bagalkot district of Karnataka to estimate. The impact of soil health card scheme on enhancing farm productivity and farmers income in Bagalkot district of Karnataka. Primary data was collected from 120 farmers in that 60 farmers were soil health card holders and 60 control farmers. The cultivation of major crops under soil health card holders was profitable as it generates higher net returns (sugarcane Rs.98583.47/acre, maize Rs.20432.40/acre and wheat Rs.12075.65/acre) compare to control farmers (sugarcane Rs.78272.36/acre, maize Rs.12218.30/acre and wheat Rs.6335.89/acre). Soil health card holders used the recommended fertilizers and FYM as per as mentioned in the soil health card. And control farmers used more fertilizers and underutilization of FYM leads to less yield and income compare to soil health card holders. Soil health card holders cultivating farms were more technically and economically efficient compared to the control farmers cultivating farms. Variation in rainfall, Agricultural extension services, Crop pests and diseases were major constraints in soil health card holders crops cultivation. And increases the awareness to control farmers about soil health card for better yield and income. Also reduced the more fertilizers usages in the farm and increases the soil fertility also. This made farmers economically stable.

## **Supply Chain Disruption of Jasmine Flowers in Ballari District of Karnataka**

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THE present study was undertaken in Huvina Hadagali taluk of Ballari district of Karnataka state to analyze economics and supply chain management of jasmine flowers. Thirty jasmine farmers, 15 wholesalers, 15 retailers and 15 consumers were interviewed during 2020-21 using pre-tested interview schedules. Descriptive statistics, shepherd's index and garrett's ranking technique were adopted to analyze the data. Per acre establishment cost, maintenance cost and cost of cultivation were Rs.50,453, Rs.2,92,773, Rs.5,29,947, respectively. Gross returns and net returns realized from six year old jasmine garden was Rs.7,68,375 and Rs.2,38,428, respectively. Net present value of investment was Rs.2,07,020 (at 15 % discount rate), payback period was 4.04 years, BC ratio was 5.10 and internal rate of return was 67.21 per cent. Quantity of flowers traded during lockdown period was reduced by 90.71 per cent among farmers, 75 per cent and 58.25 per cent among wholesalers and retailers, respectively. Three marketing channels were, channel I: producer-wholesaler-cum-commission agents-retailers-consumers; channel II: producers-retailers-consumers; channel III: producers-consumers. Producer's share in consumer's rupee was high in channel-III (100 %) followed by channel-II (42.71 %) and channel-I (30.92 %). Channel-III was the most efficient channel with Shepherd's index of 1. High commission charges (12-14 %), incidence of pests and disease, scarcity of labour were major constraints in production. High marketing cost, price and demand fluctuation and absence of regulated market were major marketing constraints encountered by farmers and intermediaries.

## Value Chain Analysis of Broiler Poultry Farms – A Study along North Transect of Bengaluru

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THE present study was conducted in north transect of Bengaluru including Doddaballapur and Devanahalli districts. The study aimed at mapping the broiler value chain; assess the financial viability of broiler enterprise; analyze the production efficiency of broiler farms, examine the marketing of broiler birds; analyze the consumer preference for broiler meat; and examine the production and marketing constraints in broiler farming. The primary data was collected from 40 broiler farmers and 60 consumers. The major actors in broiler value chain were input suppliers, broiler poultry farmers/producers, processors, traders, retailers and consumers. The total cost incurred and gross returns realized per batch with a flock size of 19,688 birds was Rs.2,17,692/- and Rs.4,85,118/-, respectively, resulting in a net return of Rs.2,67,425.50/-. At 12 per cent discount rate, the net present worth, benefit-cost ratio and internal rate of return were found to be Rs.94,16,648/-, 2.46 and 62 per cent, respectively, indicating financial viability of broiler farming. The water cost and bedding material cost were the important factors which had positive impact on returns from broiler farming. Two marketing channels were identified in the study area viz., Channel I: Producers – Integrators – Traders – Retailers – Consumers and Channel II: Producers – Integrators – Processors – Consumers. Easy availability of broiler meat was the major factor influencing the consumption. Disease outbreak was the major production constraint while price fluctuation was the major marketing constraint in broiler farming.

## Value Chain Analysis of Chilli in Guntur District of Andhra Pradesh

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THE study was conducted in Pedanandhipadu Mandal of Guntur district with the objective of analyzing the value chain of chilli in Guntur district. The primary data for the study was collected from 90 respondents which included farmers (30), commission agents (10), wholesalers (10), processors (05), exporters (05) and consumers (30). The secondary data was obtained from the FAOSTAT and INDIASTAT database. The analytical tools employed to conduct the study were descriptive statistics, value chain analysis and Garrett's ranking. The important stakeholders in the value chain of chilli in the Guntur district were input suppliers, chilli growers, commission agents, wholesalers, processors, exporters and consumers. Nearly 43.33 per cent of the chilli growers preferred to sell chilli through agricultural market committees. *Teja* and *Byadagi* chilli varieties were most preferred by the processors since they mix these two varieties to bring out the chilli powder which has a good demand in the local markets as well as outside the state. The degree of the value addition for dry chilli was highest at the exporter level (10%). In case of chilli powder, the value addition at the processors level was highest (20.81%). The study revealed that there is a good scope for value addition since there is a good demand for value-added products. Hence, the government needs to take up the initiatives in strengthening the value chain of chilli. Exploring new markets, promoting value-added products by the exporters will certainly help in strengthening the chilli value chain.

## **Consumer Preference and Price Behaviour of selected Vegetables in Bengaluru Rural District of Karnataka**

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THE present research which aimed at studying the consumer preference and price behaviour of selected vegetables was conducted in Bengaluru Rural district of Karnataka. The study used both the primary data (40 farmers, 30 market intermediaries and 30 consumers) and secondary data (arrivals and price from krishimaratavahini website). The analysis of the data using suitable tools indicated returns per rupee of expenditure of 1.62 for tomato, 1.35 for cole crops and 1.50 for gourd family vegetables. The time series analysis showed the highest arrivals and prices for tomato during 2010 (544.71) and 2021 (1370.01) and the corresponding values for chilli were 2010 (70.86) and 2021 (3305.5). Seasonal indices for arrivals was found to be the highest during the month of June (108.07) for tomato, during July (108.08) for chilli, during August (116.78) for cabbage cauliflower and ridge gourd and January (127.93) for cucumber. Majority (66.67%) of the consumers purchased during morning hours and spent around 12 per cent of their consumption expenditure on vegetables. Among the different channels identified in marketing of vegetables, Channel III (Farmer-Commission agent-Wholesaler-cum-traders-Retailer-Consumer) was found to be the most efficient for tomato, chilli, cabbage and cauliflower, while for gourd vegetables (cucumber and ridge gourd) channel IV (Farmer- Commission agent-Wholesaler-cum-traders-Vegetable vendor-Consumer) was found to be the most efficient based on the Acharya's and Shepherd's measures of marketing efficiency. The major constraints faced by the farmers in marketing of vegetables included high price fluctuations, delayed receipt of payment for vegetables and lack of storage facility.

## **Business Performance of Tur Processing Unit in Kalburgi District of Karnataka**

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THE present study was planned to assess the business performance of tur processing unit and it was carried out in two processing units *i.e.*, Gubbi Dal and Sri Vasavi Dal Industry Kalburgi district from Karnataka. The liquidity position of Gubbi dal industry was 3.69 and 6.52 for Sri Vasavi dal industry and was well over the acceptable norms. The leverage ratio – debt to equity was 1.35 for Gubbi dal industry and 0.02 for Sri Vasavi dal industry, capital employed to net worth ratio is 1.03 for Gubbi dal industry and 2.02 for Sri Vasavi dal industry, Interest coverage ratio for Gubbi dal industry was 1.47, whereas 8.00 for Sri Vasavi dal industry, the units are making enough money to pay the interest. The Gross and Net profit margins of Gubbi dal industry were 0.53 and 0.13 and for Vasavi dal industry 0.33 and 0.14, respectively. Around 68.33 per cent of growers sell their produce through channel I (producer → wholesaler → retailer → consumer) and 31.67 per cent in channel II (producer → processor → wholesaler → retailer → consumer). Price spread was higher in channel I (Rs.10,700). Damage due to pest and diseases, scarcity of labour, water and lodging due to wind were the major production constraints. The major marketing constraints were fluctuation in prices, high transportation cost and lack of storage facilities. The major constraints faced by the processing units are difficulty in availability of raw material, fluctuation in prices of raw materials, wage rate and labour problem.

## Value Chain Analysis of Bangalore Blue Grapes in Chikkaballapur District

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THE study was conducted to assess the value chain analysis of Bangalore blue grapes in Chikkaballapur district. The primary data was collected from 90 respondents which include farmers (60), traders (10), wholesalers (10) processors (5) and consumers (5). The secondary data about district wise production of grapes was collected from Directorate of Horticulture. The analytical tools employed include compound growth rate analysis and descriptive statistics. Area and production of Chikkaballapur district was increased by 2.43 and 14.86 per cent over ten years. The study revealed that the cost of cultivation per acre was Rs.227130.06. The average yield per acre was 179 quintal which accrued a gross return of Rs.320768 per acre and net return per acre was Rs.93637.94. The return per rupee investment was Rs.1.4. The process of value addition starts at the stage of trading, because farmers in the chain will not add any value to grapes at farm level. Degree of value addition takes place during pulp extraction (41.80%) and processing of grapes into wine (312.29%). Lack of proper marketing system, non-availability of storage and processing unit, poor transportation facilities, absence of price support scheme are the major constraints. Farmers' Producers' Organization should be formed at village level. The infrastructures like pack houses, cold storages etc should be built on cooperative/community basis and all members farmers should be able to use them.

## Supply Chain Analysis of Mango in Dharwad District of Karnataka

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THE study was conducted in Dharwad district of Karnataka during 2020-21 to analyze the economics of production, marketing, supply chain, post-harvest losses, constraints in production and marketing of mango. Total cost of cultivation of mango in Hubballi was Rs.55,688.52/ acre and in Dharwad it was Rs.50,855.19/ acre and overall average was Rs.53271.85 /acre. The cost of chemical fertilizers (Rs.5600.21 in Hubballi, Rs. 5000.12 in Dharwad with a pooled average of Rs.5300) was the major item of variable cost and the rental value of land was the major fixed cost item (Rs.17,348 in Hubballi and Rs.16,547 in Dharwad with a pooled average of Rs.16, 497). The average yield of mango was 5.8 tonnes /acre in Hubballi taluk and 5.45 tonnes/ acre in Dharwad taluk, the gross and net returns were Rs.1, 35,175 and Rs.81, 903, respectively per acre. Producer's share of mango was 44.13 per cent in SC-I (involving CA), 50.48 per cent in SC-II (involving village trader) 90.74 per cent in SC-III (Direct sale to consumer). Marketing efficiency index (MEI) in SC-I was 0.79, 1.28 in SC-II and 10.80 in SC-III. Total post-harvest losses (PHL) in mango was 21.48 per cent in Hubballi and 18.83 per cent in Dharwad. Pests and diseases and fluctuating market price were the major production and marketing constraints. The study recommended the adoption of improved technologies to enhance the productivity and to minimize the losses and maximize the efficiency.

## **A Study on Supply Chain Management of Mango in Nalgonda District of Telangana**

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THE study was conducted to examine the forward and backward linkages for processing products, costs and returns of different methods of mango production and constraints in the supply chain. The primary data was collected from 20 traditional and high-density mango growers and 20 intermediaries. The study revealed that 80 per acre of the processing units procure raw material from pre-harvest contractors, 60 per acre of the packaging material and 40 per acre of the chemical agents from the manufacturers. The annual maintenance cost per acre was Rs. 45,029.08 for traditional orchards and Rs. 66,446.52 for the high-density orchards. The average yield per acre was 5.62 tonnes from traditional orchards and 12.54 tonnes from high-density orchards which accrues a gross return of Rs. 1,60,170 for traditional orchards and Rs. 3,57,390 for high-density orchards. Around 80 per cent of the mango growers sold produce through channel-I (Producer - Pre-harvest contractor – Wholesaler – Retailer - Consumer) with a marketing cost of Rs. 1240.18 per quintal. Around 40 per cent of the processing units dispose their produce through channel-I (Producer-Processor-other firms-Retailers-Consumer) and Channel-II (Producer - Pre-harvest contractor – Processor-Wholesaler – Retailer - Consumer). Flower fall due to wind, damage due to pests and diseases and high cost of input were the major production constraints. Fluctuations in prices, lack of storage facilities and delays in payment were the major marketing problems faced by growers. Damage due to improper transportation, poor harvesting practices followed by farmers and high labour charges were the major problems faced by the processing units.

## **A Study on Supply Chain Management of Arecanut in Chickmagaluru District of Karnataka**

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ARECANUT is one of the most important commercial crop and the world's most traded commodity. The main objectives of the study are to analyse the supply chain management of Arecanut, to assess profitability and to document the constraints in the arecanut supply chain. The study was conducted in Chickmagaluru district of Karnataka. Primary data was collected from farmers (60), traders (5), pre harvest contractor (5), retailers (5), consumers (5). The data was subjected to tabular analysis, process mapping technique, descriptive statistics and Garrett's ranking technique. The supply chain of arecanut consisted of farmer, traders, wholesaler, pre harvest contractors and consumer. About 63.33 per cent of farmers sold their produce through pre harvest contractor, 20 per cent of farmers sold their produce through traders, 6.67 percent of the farmers sold their produce through commission agents. Since B:C ratio was greater than one the investment was financially viable. The study reveals that low price for the produce and late payment by the traders were the major constraints faced by the farmers in marketing of arecanut. The study also indicated that price fluctuation and delivery problem was the major constraints faced by the intermediaries in the supply chain.

## **Performance Analysis of Hassan Co-Operative Milk Producers' Societies Union Limited (HAMUL)**

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THE study was conducted in Hassan district of Karnataka state during 2020-2021 to analyze the performance analysis of Hassan Co-operative Milk Producers' Societies Union Limited (HAMUL). A total of 60 farmers who are the members of the union and supplying milk to HAMUL were selected by using simple random sampling technique. Personal interview method was used to collect data and appropriate statistical tools were applied to analyze the data. The physical performance analysis revealed that the growth in milk procurement was positive with a growth rate of 7.70 per cent per annum. Majority (62.16 %) of the dairy farmers preferred HAMUL for the service of artificial insemination. Among the facilities created by HAMUL milk lactometer ranked I (53.90 %). The Union does not achieved 100 per cent target from 2011 to 2021 but highest recorded in the year 2018-19 with 98.45 per cent. The cost of transportation, milk procurement, milk sale and milk loss during handling, transportation have increased by Rupees 0.18 per litre 4,79,751 LDP, 4,78,688 LDP and 1,066 LPD, respectively during the period from 2011-12 to 2020-21. Among top ten sold products, standardized milk 3,62,24,868 kg per year rank I. The major problems faced by HAMUL in marketing of milk and milk products were, competition from local dairies and other private firms.

## **Adoption and Marketing Behaviour of Tamato Growers in Madanapalle Division of Chittoor District**

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THE present study was carried out in Madanapalle division of Chittoor district in Andhra Pradesh to analyse the adoption and marketing behavior of tomato growers. Sixty tomato growers were interviewed in ten villages of Madanapalle division. The results revealed that as high as 41.68 per cent of the tomato growers were belonging to high overall adoption level of recommended cultivation practices, while 31.66 and 26.66 per cent of the tomato growers were falling under the medium and low category of overall adoption level of recommended cultivation practices, respectively. Annual income, tomato farming experience, extension agency contact, education, land holding, tomato farming experience, cosmopolitaness, achievement motivation, management orientation, innovativeness, mass media participation, and extension participation of tomato growers had significant to highly significant association with the adoption of recommended cultivation practices. All the tomato growers had studied the available resources and facilities in the area before cultivating tomato (100.00%), understood the customer's needs before cultivating tomato (100.00%), understood the market distribution system of tomato (100.00%) and collected information about institution/ persons engaged in the marketing of tomato (100.00%), While, a greater majority of the tomato growers (90.00%) had decided the marketing channel for marketing tomato that will give maximum profit.

## Small Ruminant Value Chain Analysis - A Study of Sheep Rearing along South Transect of Bengaluru

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THE present study was conducted in South transect of Bengaluru (Bengaluru urban and Ramanagara districts) to map the sheep value chain; to assess the financial viability, production efficiency of sheep enterprise and marketing of sheep; to analyse the consumer preference for sheep meat; and to identify the constraints in sheep rearing. The major actors in sheep value chain in the study area were input suppliers, sheep rearers, butchers and consumers. The total cost incurred and the gross returns realised per annum for rearing a flock size of 51 sheep was Rs.2,86,384.62/- and Rs.4,29,364.83/-, respectively, resulting in a net returns of Rs.1,42,980.21/-. Labour was the major cost accounting for about 49 per cent of the total variable cost. At 12 per cent discount rate, the NPW, BCR and IRR were found to be Rs.3,36,569.23/-, 1.24 and 36 per cent, respectively, indicating the financial viability of sheep rearing. In the study area, two channels were prevalent for marketing of sheep, viz., channel I: Farmer – Farmer and channel II: Farmer – Butcher – Consumer. Majority (50 %) of the sample sheep farmers sold their sheep exclusively through Channel I while 37.50 per cent of the sheep farmers sold their sheep exclusively through channel II. Tenderness was the most important factor influencing the purchase of sheep meat by sample consumers. The incidence of diseases and lack of organised marketing facility were the major constraints faced by sheep rearers.

## A Study on Seasonality and Volatility in Arrivals and Prices of Soybean in Selected Markets of Karnataka

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SOYBEAN is a popular crop all over the world. Soybean is used to make food, cooking, industrial oils and animal feed. India ranks fourth in area and fifth in production in the world. Karnataka has the fourth position in area and production of soybean in the country. To study the trend, volatility analysis and for forecasting, the monthly data on arrivals and prices of soybean were collected from Belagavi, Bidar and Dharwad APMCs for the period of 15 years (2005-2019). Two years weekly price data (2018-2019) was collected for studying the co-integration of soybean markets. To analyze the trend in arrivals and prices of soybean, linear and nonlinear models were fitted. MAPE and  $R^2$  values were considered to check the adequacy of the fitted models. Among the fitted models cubic model was best fitted to arrivals of soybean for Belagavi market and prices of Belagavi and Bidar market. The exponential model was best fitted among the fitted models for arrival and prices of soybean in Dharwad market and arrival of Bidar market. Seasonal ARIMA models and GARCH models were fitted for estimating and forecasting the prices of soybean. The fitted SARIMA models for the prices of the three markets, Belagavi, Bidar and Dharwad were SARIMA (2,1,1)(1,0,0)<sub>12</sub>, SARIMA (0,1,0)(0,0,1)<sub>12</sub> and SARIMA (0,1,0)(1,0,0)<sub>12</sub>, respectively and the volatility was absent in all the price series of markets. Cointegration test revealed that all the selected markets were co-integrated and they have one cointegration equation.



## A Statistical Analysis of Arrivals and Prices of Mango in selected Markets of Karnataka

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ANALYSIS of prices and market arrivals over time is important in order to understand price fluctuations and for establishing effective methods and means for reducing price fluctuations of agricultural commodities. In the present study, trends in mango crop arrivals and prices over time from Binny Mill, Chintamani and Srinivaspur markets of Karnataka state were chosen from a pool of all market places in Karnataka based on the highest amount of mango arrivals. Monthly secondary data on arrivals and prices of mango were obtained from the respective APMCs over the period of 18 years from 2002 to 2019. The linear model predicted the trend in mango arrivals as the best in Binny Mill market while cubic model predicted the best in both Chintamani and Srinivaspur markets. Further, cubic model explained the trend in mango prices as the best in both Binny Mill and Chintamani markets while Quadratic model explained best for Srinivaspur market. Box-Jenkin's method was used to forecast mango prices in selected markets. Since, there was seasonality in the data, seasonal ARIMA model were fitted. The data's stationarity was tested using the Augmented Dickey-Fuller test, which revealed that all the markets' price series were non-stationary. From the analysis, it was revealed that SARIMA(0,1,1)(0,0,1)[3], SARIMA(2,0,0)(0,0,0)[3] and SARIMA(0,1,0)(1,0,1)[3] models were the best fitted SARIMA models for Binny Mill, Chintamani and Srinivaspur markets, respectively.

## Organic Weed Management in Black Gram (*Vigna mungo* L.)

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A field experiment was conducted during *rabi* 2020-21 at RIOF field unit, UAS, GKVK, Bengaluru. The experiment was laid out in RCBD comprising twelve treatments replicated thrice. The treatments were consisting of inter cultivation, stale seedbed technique straw mulching, fodder cowpea as an intercrop, smothering crop and *in-situ* incorporation, cycle weeding, spraying of cucumber and *Ageratum conyzoides* leaf extracts, hand weeding and weedy check. Major weeds observed were *Cyperus rotundus*, *Cynodon dactylon*, *Eleusine indica*, *Dactyloctenium aegyptium*, *Ageratum conyzoides*, *Alternanthera sessilis* and *Borreria hispida*. Among different weed management practices stale seed bed technique + inter cultivation twice at 25 and 45 DAS recorded lower total weed density, weed dry weight, weed index and higher weed control efficiency (38.67 m<sup>2</sup>, 5.21 g m<sup>-2</sup>, 3.49 % and 57.27 %, respectively) and registered significantly higher seed and haulm yield (1089 and 4514 kg ha<sup>-1</sup>, respectively), which may be attributed to significantly higher plant height (34.04 cm), number of branches plant<sup>-1</sup> (7.55) and total dry weight of plant (24.22 g) at harvest, leaf area at 60 DAS (811.66 cm<sup>2</sup> plant<sup>-1</sup>), number of productive branches (5.87), number of pod plant<sup>-1</sup> (29.71), test weight (47.80 g), seed yield per plant (6.71 g) and protein content (25.08 %) and this treatment also resulted in higher net returns (Rs.25698 ha<sup>-1</sup>) and B-C ratio (2.44) and found to be most economical and comparable with the treatment of hand weeding at 20 and 40 DAS.

## Studies on Influence of Nano Nitrogen and Phosphorus on Growth and Yield of Maize (*Zea mays* L.)

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A field experiment was conducted at Zonal Agricultural Research Station, GKVK, UAS, Bangalore during *khariif* 2021 to study the effect of nano nitrogen and phosphorus on growth and yield of maize. The experiment was laid out in RCBD with 9 treatments, replicated thrice and the cultivar used was BRMH-8. The treatment includes *viz.*, RDF + FYM ( $T_1$ ), RDF ( $T_2$ ), 25% RDN + Nano-N ( $T_3$ ), 50% RDN + Nano-N ( $T_4$ ), 75% RDN + Nano-N ( $T_5$ ), 25% RDNP + Nano-NP ( $T_6$ ), 50% RDNP + Nano-NP ( $T_7$ ), 75% RDNP + Nano-NP ( $T_8$ ) and absolute control ( $T_9$ ). Nano-N ( $2 \text{ ml L}^{-1}$ ) and Nano-NP ( $1.25 \text{ ml L}^{-1}$ ) was sprayed at 30 and 60 DAS. The results revealed that application of 75% RDN + Nano-N recorded significantly higher plant height (272.2 cm), number of leaves plant<sup>-1</sup> (8.5), leaf area ( $7098 \text{ cm}^2 \text{ plant}^{-1}$ ), LAI (3.9), total dry matter production ( $491.8 \text{ g plant}^{-1}$ ), cob length (22.9 cm), cob girth (19.9 cm), number of rows cob<sup>-1</sup> (15.8), number of kernels row<sup>-1</sup> (36.9), number of kernels cob<sup>-1</sup> (625.3) and weight of kernels cob<sup>-1</sup> (255.2 g), ultimately resulted in higher kernel and stover yield (9654 and 9515 kg ha<sup>-1</sup>, respectively) and accounted 10.20 and 9.29 per cent higher kernel and stover yield, respectively as compared to RDF + FYM as per package of practices. Application of 75% RDN + Nano-N also recorded higher nutrient uptake ( $299.22 \text{ kg ha}^{-1} \text{ N}$ ,  $55.56 \text{ kg ha}^{-1} \text{ P}$  and  $208.26 \text{ kg ha}^{-1} \text{ K}$ ) and net returns ( $143370 \text{ }^1 \text{ ha}^{-1}$ ) with higher B:C ratio (4.20).

## Effect of Organic Mulches on Growth and Yield of Maize (*Zea mays* L.)

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A field experiment was conducted at College of Agriculture, V.C. Farm, Mandya during *khariif* 2021 to study the 'Effect of organic mulches on growth and yield of maize (*Zea mays* L.)'. The experiment was laid out in RCBD comprising nine treatments replicated thrice. Application of pongamia leaves as organic mulch @  $5 \text{ t ha}^{-1}$  resulted in better soil moisture content at 30 days after mulching at 0 - 15 and 15 - 30 cm soil depth (14.85 and 16.03%, respectively). Higher soil temperature at 5, 10 and 15 cm depth was recorded during morning while lower was recorded during noon. At harvest, the higher infiltration rate of soil ( $5.59 \text{ cm hr}^{-1}$ ), lower bulk density ( $1.43 \text{ g cc}^{-1}$ ), higher maximum water holding capacity (48.12%), organic carbon (0.48%), available nitrogen, phosphorous and potassium ( $299.05$ ,  $42.84$  and  $254.11 \text{ kg ha}^{-1}$ , respectively), nutrient uptake ( $214.68$ ,  $30.37$  and  $210.93 \text{ kg ha}^{-1}$  nitrogen, phosphorous and potassium uptake, respectively) nutrient use efficiency, water use efficiency and microbial population was also recorded in the same treatment. Further, mulching with pongamia leaves @  $5 \text{ t ha}^{-1}$  recorded higher kernel yield ( $6180 \text{ kg ha}^{-1}$ ), stalk yield ( $9161 \text{ kg ha}^{-1}$ ), gross returns (Rs.101861 ha<sup>-1</sup>), net returns (Rs.66606 ha<sup>-1</sup>) and B:C ratio (2.89). Hence, it can be inferred that the application of pongamia leaves as organic mulch found to be superior in improving physical, chemical and biological properties of the soil, enhanced growth and yield of maize which is economically superior.

## Field Evaluation of Organic Weed Management Methods in *Kharif* Kodo Millet (*Paspalum scrobiculatum* L.)

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A field experiment was conducted at Research Institute of Organic Farming field unit, UAS, GKVK, Bengaluru during 2021 to evaluate different organic weed management methods in *kharif* kodo millet. The experiment was laid out in RCBD with 12 treatments, replicated thrice and the cultivar used was RK 390-25. The treatments consisting of intercultivation, stale seedbed technique, straw mulching, fodder cowpea as an intercrop, smothering crop and *in-situ* incorporation, cycle weeding, spraying of cucumber and *Ageratum conyzoides* leaf extracts, hand weeding and weedy check. Major weeds observed were *Cyperus rotundus*, *Cynodon dactylon*, *Eleusine indica*, *Dactyloctenium aegyptium*, *Digitaria marginata*, *Ageratum conyzoides*, *Alternanthera sessilis* and *Borreria hispida*. Among various weed management practices stale seed bed technique + inter cultivation twice at 25 and 45 DAS recorded lower total weed density, weed dry weight, weed index and higher weed control efficiency (30.0 m<sup>2</sup>, 7.49 g m<sup>-2</sup>, 17.6 % and 49.6 %, respectively) and significantly higher number of tillers (4.0 plant<sup>-1</sup>), dry matter production (21.67 g plant<sup>-1</sup>), productive tillers (3.33 plant<sup>-1</sup>), number of panicles (3.67 plant<sup>-1</sup>) and panicle length (6.38 cm) at harvest stage, which attributed to significantly higher grain and straw yield (763.4 and 4167 kg ha<sup>-1</sup>, respectively). The same treatment resulted in higher net returns (Rs.28,373 ha<sup>-1</sup>) and B:C ratio (2.34) and it was on par with intercultivation at 25 DAS with one hand weeding at 45 DAS treatment.

## Optimization of Spacing and Nutrient Levels in Sesame Genotypes (*Sesamum indicum* L.)

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A field experiment was conducted at Zonal Agricultural Research Station, UAS, GKVK, Bengaluru during *kharif* 2021 for optimization of spacing and nutrient levels in sesame genotypes. The experiment was laid out in factorial randomized complete block design consisting of three factors *viz.*, two genotypes (GT-1 and GKVK-1), two spacings (30 cm × 10 cm and 45 cm × 10 cm) and three nutrient levels (75 % RDF, 100 % RDF and 125 % RDF) with 12 treatment combinations replicated thrice. Among genotypes, GKVK-1 recorded significantly higher plant height (102.73 cm), dry matter accumulation (21.08 g plant<sup>-1</sup>), seed yield (680 kg ha<sup>-1</sup>), oil yield (297 kg ha<sup>-1</sup>), uptake of nitrogen (33.78 kg ha<sup>-1</sup>), phosphorous (11.55 kg ha<sup>-1</sup>) and potassium (36.46 kg ha<sup>-1</sup>). Narrow spacing of 30 cm × 10 cm recorded significantly higher plant height (101.11 cm), seed yield (662 kg ha<sup>-1</sup>), oil yield (291 kg ha<sup>-1</sup>), uptake of nitrogen (31.71 kg ha<sup>-1</sup>), phosphorous (10.95 kg ha<sup>-1</sup>) and potassium (33.78 kg ha<sup>-1</sup>). Application of 125 per cent RDF recorded significantly higher plant height (107.23 cm), dry matter accumulation (22.28 g plant<sup>-1</sup>), seed yield (719 kg ha<sup>-1</sup>), oil yield (318 kg ha<sup>-1</sup>), uptake of nitrogen (34.73 kg ha<sup>-1</sup>), phosphorous (11.86 kg ha<sup>-1</sup>) and potassium (36.46 kg ha<sup>-1</sup>). Higher gross returns (Rs.127878 ha<sup>-1</sup>), net returns (Rs.96223 ha<sup>-1</sup>) and cost to benefit ratio (4.04) was observed in GKVK-1 genotype with spacing of 30 cm × 10 cm under 125 per cent RDF.

## Sensor Based Irrigation and Nutrient Management through Nano Fertilizers in Aerobic Rice (*Oryza sativa* L.)

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A field experiment on above title was conducted during *kharif* 2021 at ZARS, GKVK, Bengaluru allocating three irrigation techniques in main plot (drip irrigation as per UAS-B PoP, sensor based automated drip irrigation at 25% DASM and at 50% DASM) and five nutrient management treatments in sub plots (absolute control, 100% RDF, foliar spray of nano-urea @ 2 ml litre<sup>-1</sup> at 30 and 60 DAS, foliar spray of nano-urea @ 2 ml litre<sup>-1</sup> at 30, 60 and 90 DAS and NDVI based nitrogen management through nano-urea @ 2 ml litre<sup>-1</sup>) replicated thrice in split plot design. The results showed that, sensor based automated drip irrigation at 25% DASM recorded significantly higher grain yield (7091 kg ha<sup>-1</sup>), straw yield (7782 kg ha<sup>-1</sup>), gross returns (Rs.138614 ha<sup>-1</sup>), net returns (Rs.80955 ha<sup>-1</sup>) and B:C ratio (2.4) apart saving water to the tune of 18.58 per cent. Among nutrient management, foliar spray of nano-urea at 30, 60 and 90 DAS recorded significantly higher grain yield (7953 kg ha<sup>-1</sup>), straw yield (8787 kg ha<sup>-1</sup>), NUE (63.28 kg kg<sup>-1</sup>), gross returns (Rs.155493 ha<sup>-1</sup>), net returns (Rs. 94616 ha<sup>-1</sup>) and B:C ratio (2.6). Among interaction, sensor based automated drip irrigation at 25% DASM with foliar spray of nano-N at 30, 60 and 90 DAS recorded significantly higher grain yield (9343 kg ha<sup>-1</sup>) and straw yield (9574 kg ha<sup>-1</sup>).

## Performance of Niger Genotypes under Different Levels of Nutrients and Spacing

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A field experiment was conducted at Zonal Agricultural Research Station, GKVK, UAS, Bangalore during *kharif* 2021 for performance of niger genotypes under different levels of nutrients and spacing. The experiment was laid out in factorial randomized complete block design consisting of three factors *viz.*, two genotypes (KBN-1 and KBN-2), two spacings (30 cm × 10 cm and 45 cm × 10 cm) and three nutrient levels (75% RDF, 100% RDF and 125% RDF) with 12 treatment combinations replicated thrice. Among genotypes, KBN-2 recorded significantly higher number of branches (22.14 plant<sup>-1</sup>), total dry matter accumulation (51.35 g plant<sup>-1</sup>), seed yield (634 kg ha<sup>-1</sup>), oil yield (267 kg ha<sup>-1</sup>), uptake of nitrogen (43.76 kg ha<sup>-1</sup>), phosphorus (8.63 kg ha<sup>-1</sup>), potassium (9.91 kg ha<sup>-1</sup>). Wider spacing of 45 cm × 10 cm recorded significantly higher number of branches (21.24 plant<sup>-1</sup>), total dry matter accumulation (53.33 g plant<sup>-1</sup>), seed yield (613 kg ha<sup>-1</sup>), oil yield (249 kg ha<sup>-1</sup>), uptake of nitrogen (43.44 kg ha<sup>-1</sup>), phosphorus (8.56 kg ha<sup>-1</sup>), potassium (9.83 kg ha<sup>-1</sup>). Application of 125 per cent RDF recorded significantly higher number of branches (22.48 plant<sup>-1</sup>), total dry matter accumulation (52.59 g plant<sup>-1</sup>), seed yield (664 kg ha<sup>-1</sup>), oil yield (269 kg ha<sup>-1</sup>), uptake of nitrogen (44.26 kg ha<sup>-1</sup>), phosphorus (8.75 kg ha<sup>-1</sup>), potassium (10.07 kg ha<sup>-1</sup>). Higher gross returns (Rs.59880ha<sup>-1</sup>), net returns (Rs.29475 ha<sup>-1</sup>) and B:C ratio (1.97) was observed in KBN-2 genotype with spacing of 45 cm × 10 cm under 125 per cent RDF.

## Studies on Efficacy of Pre and Post Emergent Herbicides for Weed Management in Maize (*Zea mays* L.)

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A field experiment was carried out at College of Agriculture, V.C. Farm, Mandya during *kharif* 2021 to study the efficacy of pre and post emergent herbicides for weed management in maize (*Zea mays* L.). The experiment was laid out in randomized complete block design (RCBD) with fourteen treatments replicated thrice. The treatments comprised of sole and combined application of pre-emergence herbicides (atrazine and pendimethalin) at 3 DAS and post emergence herbicides (2,4-D, topramezone and tembotrione) at 3-4 weed leaf stages were compared with two hand weedings (at 20 DAS and 40 DAS), weed free check and unweeded check. Among chemical weed management practices, application of atrazine (50% WP) @ 1 kg *a.i.* ha<sup>-1</sup> as pre-emergence at 3 DAS followed by topramezone (33.6% SC) @ 50 g *a.i.* ha<sup>-1</sup> as post emergence application at 3-4 weed leaf stages recorded significantly lower total weed density (2.61, 2.34 and 4.26 no. 0.25 m<sup>-2</sup> at 20, 40DAS and at harvest, respectively) and lower total weed dry weight (0.90, 1.31 and 1.49 g. 0.25 m<sup>-2</sup> at 20, 40DAS and at harvest, respectively) with weed control efficiency of 70.4 per cent and weed index of 1.5 per cent at harvest. The same treatment recorded higher kernel yield (11469 kg ha<sup>-1</sup>), stover yield (13542 kg ha<sup>-1</sup>), net monetary returns (Rs.132141 ha<sup>-1</sup>) and B:C ratio (3.47). Hence, it was found to be suitable and economical for effective control of weeds in maize.

## Calibration and Validation of Ceres Maize Model for Maize Hybrids under Varied Growing Environments in Southern Dry Zone of Karnataka

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A field experiment was conducted at ZARS, V.C. Farm, Mandya during *kharif* 2021, entitled 'Calibration and validation of CERES maize model for maize hybrids under varied growing environments in southern dry zone of Karnataka'. The field experiment was laid out in split plot design consisting of four date of sowing as main plot and three maize hybrids as a sub plot, with three replication. Among different date of sowing, D<sub>1</sub> (August 1<sup>st</sup> fortnight) resulted in significantly higher leaf area index (1.76), dry matter production (331.56 g plant<sup>-1</sup>), grain yield (7355 kg ha<sup>-1</sup>) and stalk yield (10647 kg ha<sup>-1</sup>). Among different hybrids leaf area index (1.70), dry matter production (321.39 g plant<sup>-1</sup>), grain yield (6967 kg ha<sup>-1</sup>) and stalk yield (10075 kg ha<sup>-1</sup>) were significantly higher in MAH 14-5. CERES-Maize model was calibrated and validated by using the data collected from the field experiments. The model satisfactorily predicted the total dry matter production, leaf area index, number of grains per cob and grain yield with RMSE of 11.74 g plant<sup>-1</sup>, 0.23, 17.36 and 315.74 kg ha<sup>-1</sup>. The highest gross returns (Rs.1,29,952), net returns (Rs.89013 ha<sup>-1</sup>) and B:C ratio (3.17) was observed when MAH 14-5 was sown during August 1<sup>st</sup> fortnight. From this investigation, it can be inferred that the CERES-Maize model was successfully validated for southern dry zone of Karnataka with 95 per cent efficiency and hence, model can be used as a research tool to make strategic decisions.

## Effect of Pre-Emergence Herbicides on Growth and Yield of Direct Seeded Rice (*Oryza sativa* L.)

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A field experiment was conducted during *khariif* 2021 at College of Agriculture, V.C. Farm, Mandya to study the effect of pre-emergence herbicides on growth and yield of direct seeded rice (*Oryza sativa* L.). The experiment was laid out in RCBD with three replications consisting of nine treatments *viz.*, pretilachlor 30% + pyrazosulfuron ethyl 0.75% WG @ 1500, 2000, 2500 g ha<sup>-1</sup>, respectively, pyrazosulfuron ethyl 10% WP @ 250 g ha<sup>-1</sup>, pretilachlor 50% EC @ 1500 ml ha<sup>-1</sup>, pretilachlor 30% EC + 10% safener @ 1000 ml ha<sup>-1</sup>, hand weeding (20 & 40 DAS), weed free check and weedy check. The predominant weed flora observed were *Echinochloa colona*, *Cynodon dactylon*, *Eleusine indica*, *Dinebra retroflexa*, *Dactyloctenium aegyptium*, *Chloris barbata*, *Physalis minima*, *Ageratum conyzoides*, *Portulaca olearacea*, *Parthenium hysterophoru*, *Gomphrena decumbens*, *Cyperus rotundus*, *Cyperusiria*. The results revealed that pre-emergence application of pretilachlor 30% + pyrazosulfuron ethyl 0.75% WG @ 2500 g ha<sup>-1</sup> at 3 DAS recorded significantly lower total weed count (0.25, 0.72, 2.72, 15.00 and 19.33 No. m<sup>-2</sup>), weed dry weight (1.22, 3.09, 4.18, 5.50 and 8.08 g m<sup>-2</sup>) and weed control efficiency of 84.36, 82.86, 80.43, 79.00 and 78.82 per cent, respectively, at 15, 30, 45, 60 DASP and at harvest. Due to efficient weed control, the same treatment recorded higher grain yield (6491 kg ha<sup>-1</sup>), straw yield (9584 kg ha<sup>-1</sup>), net monetary returns (Rs.92120 ha<sup>-1</sup>) and B: C ratio (3.54) and was statistically comparable with weed free check.

## Effect of Different Organic Sources of Nutrients on Growth and Yield of Grain Amaranth (*Amaranthus hypochondriacus* L.)

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A field experiment was conducted at Research Institute on Organic Farming field unit, GKVK, UAS, Bangalore during late *khariif* 2021 entitled 'Effect of organic sources of nutrients on growth and yield of Grain Amaranth (*Amaranthus hypochondriacus* L.)'. The experiment was laid out in randomised complete block design which was replicated thrice. Treatments included different combinations of organic sources of nutrients. The results revealed that application of 50% N equivalent through farmyard manure + 50% N equivalent through pongamia cake recorded significantly higher plant height (165.7 cm plant<sup>-1</sup>), number of leaves (26.33 plant<sup>-1</sup>), leaf area (2435 cm<sup>2</sup> plant<sup>-1</sup>), total dry matter (71.13 g plant<sup>-1</sup>), yield components like panicle length (54.33 cm), fingers (37.67 panicle<sup>-1</sup>), grain yield (23.20 g plant<sup>-1</sup>), grain yield (2127 kg ha<sup>-1</sup>) and stover yield (3596 kg ha<sup>-1</sup>). Significantly higher total N, P and K uptake (58.67, 17.4, 57.17 kg ha<sup>-1</sup>, respectively) were observed with application of 50% N equivalent through farmyard manure + 50% N equivalent through pongamia cake. Higher available nitrogen, phosphorus and potassium (324.49, 35.60, 246.67 kg ha<sup>-1</sup>, respectively) were recorded with application of 100% N equivalent through farmyard manure. Application of 50% N equivalent through pongamia cake + 50% N equivalent through bio-digester liquid manure gave maximum net returns of Rs. 89,511 along with highest B:C ratio (2.43)

## Exploration of Underutilized Greens for $\beta$ -Carotene Enhanced Products

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MICRONUTRIENT deficiencies have become a serious global problem, resulting in poor health, low work productivity and high rates of mortality and morbidity. Therefore, the study aimed at exploration of underutilized greens for  $\beta$ -carotene enhanced products. Total of 67 respondents from six villages of Chamarajanagara district were interviewed in which 59.7 per cent of respondents were young. Majority were married (88.1%) and female (71.6%). Maximum respondents (85.1%) have been using underutilised green leafy vegetables (UGLVs) in their daily diet and 40.3 per cent cultivates UGLVs. Forty-two UGLVs were documented which belonged to 25 plant families, 3 divisions, 34 genera and 42 different plant species. Most of them were available throughout the year. UGLVs found to be used for culinary purposes and as ethnomedicinal plants to treat several diseases and disorders. They contained (g/100g) moisture- 77.7 to 93.2 per cent, protein 1.24 to 8.00, fat 0.31 to 2.20, crude fiber 1.91 to 8.83 and ash 0.93 to 2.89. The mineral content (mg/100g) were iron 0.43-17.46, zinc 0.07-1.08, copper 0.04-0.55, manganese 0.05-2.09, calcium 39.7-421.3 and phosphorus 13.2-104.6 mg/100g.  $\beta$ -carotene in UGLVs ranged from 634 to 15870  $\mu\text{g}/100\text{g}$ . Based on quality parameters of UGLVs, *Basella alba* and *Moringa oleifera* were selected for  $\beta$ -carotene enhanced products namely basella pasta, moringa pasta, moringa dark chocolates and moringa green chocolates were developed. Moringa and basella added pasta and chocolates showed better nutritional composition. A significant increase in mineral and  $\beta$ -carotene was found in moringa added products. Biochemical changes, microbial and sensory scores were within the acceptable limit in two packaging material tested at two storage temperatures. These products need to be introduced to the market at an affordable cost to reach maximum population, especially children. Therefore, UGLVs need a lot of attention from policymaker and researcher to make them available and bring diversity to meal plate.

## Evaluation and Standardization of *Tinospora cordifolia* (*Amrutha balli*) Vine for Product Development

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ONE among medicinal plants in Indian system of medication is *Tinospora cordifolia* (*amrutha balli*). The plant is a large, smooth surfaced climbing shrub. The present study aims at utilizing the benefits of the plant by standardizing the process of dehydration, proximate analysis of the dehydrated vine and product development. Dehydration was done using hot air oven for, blanched and non-blanched samples, at different temperatures of 40, 50 and 60°C. The colour analysis values indicated that vine blanch-dried at 60°C had more colour retention compared to other temperatures *i.e.*,  $L^*$ ,  $a^*$ ,  $b^*$  values were 29.01, -0.98, 7.25 and 53.21, 0.95, 16.17 for leaves and stems, respectively. Proximate analysis of blanched leaves and stem indicated moisture (%), carbohydrate (g), protein (g), fat (g), ash (g), fibre (g), energy (kcal) content as 5.87, 68.43, 7.37, 0.29, 5.04, 12.99, 306 and 6.98, 56.64, 8.64, 0.09, 12.64, 15.01, 262, respectively per 100 g. The mineral analysis showed that vine contains adequate amount of essential minerals *viz.*, calcium, magnesium, iron, phosphorus, sodium and zinc. The findings revealed that aqueous vine extract possess good antimicrobial activity against commonly associated food contaminants *viz.*, *Staphylococcus aureus* and *Serratia marcescens*. Value added products such as green tea powder (15 %) and crackers (40 % replaced with cumin seeds) were acceptable. Vine powder can be incorporated into many traditional products to achieve healthier products, other than consumption in crude form. Thus the production and consumption of commercial *T. cordifolia* incorporated food products can be encouraged among all sectors of population.

## Development of Ready-to-Drink (RTD) Beverage Mix from Finger millet (*Eleusine coracana*) and It's Nutritional Evaluation

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RTD beverage mix was developed from finger millet by using spray drying technology. Finger millet variety (GPU-66) was popped to get popped finger millet flour that was blended with homogenized milk and subjected to spray drying. Three blends at 5, 10 and 15 per cent slurry concentration were prepared with popped finger millet flour which was then spray dried with different inlet temperatures and outlet temperatures. Adding powdered sugar and different flavours to the spray dried powder was done. Sensory evaluation of reconstituted RTD beverage mix indicated that 10 per cent slurry concentration with vanilla flavour was best among all variations. The spray drying conditions for 10 per cent ragi slurry concentration viz., inlet air temperature 170°C, pressure 2.0 bar, blower speed 1300rpm, flow rate 13rpm and yield 145 g/kg of spray dried finger millet powder. Proximate composition of RTD mix indicated protein (11.09±0.21 %), fat (5.61± 0.14 %), carbohydrate (73.93±0.25 %), energy (Kcal/100 g) (390.77± 2.00), moisture (2.64±0.02 %). It was found that RTD beverage mix had highest amount of potassium (457 mg/100 g) followed by calcium (380 mg/100 g), phosphorous (202 mg/100g) and magnesium (112 mg/100 g) observed. The bulk density (g/mL) and solubility index (%) was found to be 0.28 g/mL and 2.2 per cent, respectively. This beverage mix is stable during storage both in metalized polyester pouch and glass jar till three months with microbial counts under permissible limits. Hence, RTD beverage mix was found to be a satisfactory reconstituted product with good sensory properties and storage period.

## Induction of Seed Dormancy and its Impact on Seed Quality in Groundnut (*Arachis hypogaea* L.)

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FIELD and laboratory experiments were conducted to induce dormancy in nondormant groundnut variety GKVK-5 by foliar spraying of different dormancy inducing chemicals of various concentrations viz., maleic hydrazide @ 1250, 1500 ppm, abscisic acid @ 100, 200 ppm and cycocel @ 2000, 3000 ppm at 85 DAS during *kharif* 2020. The results revealed that there was a significant difference between the treatments of which the highest germination (82.00%) was recorded from control. While, the least germination (28.66 %) was recorded from maleic hydrazide @ 1500 ppm immediately after harvest. The dissipation of dormancy was studied both in field and laboratory condition. The seeds from the dormancy induced plots showed dormancy of upto 8 to 12 days whereas, the seeds from control showed a dormancy period of mere 1 to 2 days in field condition. The seeds under lab condition recorded the germination of 74.66 and 75.33 per cent from MH @ 1500 ppm and cycocel @ 3000 ppm, respectively on 16 DAH. Whereas, the control was recorded 82.00 per cent germination immediately after harvest. The harvested seeds from each of the plot was carried forward to break the dormancy with different dormancy breaking chemicals at various concentrations viz., KNO<sub>3</sub> @ 0.4, 0.6 %, GA<sub>3</sub> @ 250, 500 ppm, Ethrel @ 250, 500 ppm, hydration and dehydration along with control. It was recorded that GA<sub>3</sub> @ 500 ppm recorded the highest germination (98.83%) followed by hydration and dehydration. Whereas, the lowest germination (28.66 %) was recorded from control.



## Genome-wide Association Studies for Seed Longevity in Soybean [*Glycine max* (L.) Merrill]

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SIXTY diverse genotypes with different seed coat colour and seed size was evaluated for seed longevity under natural ageing for 20 months and accelerated ageing as per international seed testing association (ISTA) guidelines over two season using fresh seeds. Seed germination was recorded at bimonthly interval after storage in natural ageing and another set after accelerated ageing. The correlation between accelerated and bimonthly natural ageing was highly significant. Two black seed coat colour genotypes, Acc No.369 and Acc No.39 consistently showed higher longevity over the ageing treatment and serve as a source of higher seed longevity in soybean. A panel of 96 genotypes was combined for association mapping and the genetic diversity as well as population structure was studied using SSR, Genotyping-by-Sequencing (GBS)-SNP and agro-morphological traits. Bayesian-model based population STRUCTURE, UPGMA and principal component analysis approaches using SSR, SNP and agro-morphological traits exhibited a complementary pattern of population structure. The genotypes with higher seed longevity and small seed size formed a single cluster. Higher coefficient of variability of seven productivity and nine seed longevity related traits confirmed the diversity in assembled population and its suitability for QTL mapping. Genome-wide association mapping using 29,955 GBS-SNP markers and sixteen traits produced 30 marker trait associations. One co-localized SNP (SNW\_024464723.1\_95172) was identified for seed longevity traits viz., seed germination, seedling vigour index-II and per cent reduction in vigour index-II. The biparental linkage mapping population for seed longevity was also developed by crossing JS93-05 and Local black soybean using Wet cotton and Kolot method with wet cotton method producing higher success rate. High genetic variability was observed among 352 F<sub>2</sub> plants. The GBS-SNP resources, association mapping panel and F<sub>2</sub> population developed in this study serve as a valuable resource for soybean crop improvement programme.

## Influence of Seed Invigoration and Pro-Tray Media on Seedling Emergence and Establishment in Hybrid Tomato under Nursery Condition

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THE experiment was conducted to study the 'Influence of seed invigoration and pro-tray media on seedling emergence and establishment in hybrid tomato under nursery condition' during 2020-2021. Among ten different priming treatments and ten pro-tray media by considering all seed quality parameters tested, the best seed priming treatment was hydropriming for 12 hours and pro-tray media was coco peat (95 %) + zeolite (5 %), which were showed improved seed germination (93.00 and 89.33 %), pro-tray seed germination (92.33 and 91.33 %), days taken for 1<sup>st</sup> emergence (5.33 and 6.33) and days taken for complete emergence (11.33 and 12.00), speed of emergence (37.6 and 38.5), seedling length (34.88 and 36.540 cm), stem girth (3.14 and 3.23 cm), number of leaves (18.52 and 19.75), leaf area (12.64 and 12.91 cm<sup>2</sup>), chlorophyll content (38.50 and 30.40 mSPU), root mass (328.27 and 340.33 mg), dry root mass (44.90 and 46.91 mg), shoot mass (2634.90 and 2799.8 mg), dry shoot mass (283.35 and 297.35 mg), seedling mass (2963.17 and 3128.03 mg), dry seedling mass (328.25 and 344.26 mg) and number of transplantable (93 and 91.33 %), respectively, followed by KNO<sub>3</sub> (1 %) for 12 hours, GA<sub>3</sub> (100 ppm) for 24 hours in priming treatment and coco peat (95 %) + perlite (5 %), coco peat (95 %) + vermiculite (10 %) in pro-tray media. In conclusion, 12 hour hydropriming with media coco peat (95 %) + zeolite (5 %) could be used commercially for raising tomato seedlings in pro-tray nursery conditions.

## Standardization of Seed Production Techniques in Newly Developed Single Cross Maize Hybrid; MAH 14-138

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THE present investigation was carried out in two field experiments, one being the staggered sowing to study synchronization of flowering between female (MAI-729) and male (MAI-105) parents of single cross hybrid maize, MAH 14-138 during *rabi* 2020. The experiment was laid out in RCBD design with five treatments and four replications. The female and male parents sown on same day ( $S_2$ ) showed better synchronization in flowering which resulted in significant enhancement of yield parameters like cob length (15.80 cm), cob weight (146.45 g), number of seeds row<sup>-1</sup> (28.44), seed yield plant<sup>-1</sup> (163.65 g), seed yield plot<sup>-1</sup> (2.79 kg) and seed yield ha<sup>-1</sup> (38.77 q) in female parent. To optimize the planting ratios an experiment was laid out in split plot design with five main plot treatments (planting ratios) and four sub plot treatments (agronomic practices) during *kharif* 2021. The interaction of planting ratio and agronomic practice  $P_4T_3$  (4:2 planting ratio, 2 per cent urea spray before flowering) registered significantly higher values for cob weight (166.33 g), seed weight cob<sup>-1</sup> (151.42 g) and seed yield plant<sup>-1</sup> (233.02 g) in seed parent. However, significant differences for seed yield plot<sup>-1</sup> (15.61 kg) and seed yield ha<sup>-1</sup> (59.34 q) were observed in  $P_2T_3$  (4:1 planting ratio, 2 % urea spray before flowering). Hence, the study concluded that the sowing of female and male parents on same day with planting ratio 4:1 and 2 per cent urea spray before flowering could achieve better synchronisation of flowering in parents, higher seed yield and quality of seed parent.

## Alternative Media Composition for Production of Micro-Propagated Disease-Free Seedlings of Sugarcane (*Saccharum officinarum* L.)

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EXPERIMENT was conducted to standardize an alternative media composition for production of micro-propagated disease-free seedlings of sugarcane. The study consisted of two experiments which includes, shoot and root growth under *in-vitro* condition and hardening of seedlings under greenhouse. Significant differences were noticed between the media with standardized and alternative sources of components. Among the alternative gelling agents used, MS media with China grass at 25g/l produced a mean of 88.04 per cent uncontaminated explants with minimum days taken for sprouting (10.47) and maximum per cent survival of explants (87.67) in varieties *viz.*, VCF0517, Co86032, CoVC18061, CoVC16061 and CoVC16062. All the varieties studied have developed maximum number of shoots per explant (11.45) with maximum shoot length (8.47 cm) in MS media with table sugar 20 g/l. Whereas, maximum number of roots (8.35) and root length (6.43 cm) were noticed in tender coconut water at 100 ml/l media in all the varieties. Maximum per cent survival of seedlings (93.33) was recorded in composted pressmud along with sand and soil in the ratio of 1:1:2 after 21 days of hardening.

## Micropropagation of Disease-Free Seedlings of Ginger (*Zingiber officinale* Rosc.)

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THE experiment was conducted to standardize an alternative media for micro propagation of ginger. The study consisted of two experiments viz., shoot and root growth under *in vitro* condition and hardening of seedlings under greenhouse. Significant differences were noticed among the media with standardized and alternative sources of components. Among the alternative gelling agents used, MS media with China grass at 25 g/l produced 86.67 per cent uncontaminated explants with prominent sprouting of 91.67 per cent in Maran and 86.67 per cent in Rio de Janerio and Himachal varieties within 11 days of inoculation. Table sugar used at 20 g/l media as a substitute to sucrose produced greater number of average shoots (1.73) per explant with average shoot length of 6.85, 6.68 and 6.79 cm in Rio de Janerio, Himachal and Maran varieties, respectively. All the varieties have produced maximum number of roots along with maximum root length in tender coconut water at 100 ml/l media. Rio de Janerio, Maran and Himachal have produced average number of 2.5 roots with root length of 2.35, 3.20 and 2.49 cm, respectively. In general, in all the three varieties, cocopeat along with sand and soil in the ratio of 1:2:1 showed maximum of 92.59 per cent micro propagated seedling establishment after 21 days of hardening.

## Impact of Entomopathogens and Inert Dust Seed Treatment on Insect Pests and Seed Quality of Cowpea (*Vigna unguiculata* L.) during Storage

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BRUCHID, a pest that has survived from the field is a severe problem with stored cowpea seeds. Although there are various approaches to addressing this issue, utilising biological agents is the most practical, cost-effective and ecologically friendly choice. Laboratory study was conducted in a completely randomised manner to investigate the effect of entomopathogens on the storability of cowpea seeds. Cowpea seeds were treated with the entomopathogens *Beauveria bassiana* and *Metarhizium anisopliae*, diatomaceous earth and a chemical check called deltamethrin @ 1 ppm. The seeds were then stored in HDPE bags for nine months, from July 2021 to April 2022. The parameters of insect population and seed quality were recorded in order to assess the efficacy of entomopathogens. *Metarhizium anisopliae* (CFU:  $1.0 \times 10^8$ ) @ 20 g/kg seed + Diatomaceous earth @ 5g/kg seeds recorded the highest seed quality parameters viz., hundred seed weight (10.38 g), seed germination (82 %), shoot length (22.70 cm), root length (17.09 cm), mean seedling length (39.78 cm), mean seedling dry weight (55.09 mg), seedling vigour index I and II (3053 and 4244), total dehydrogenase activity @ 480 nm (1.97), total protein (21.26 %) and the lowest seed moisture content (9.64 %), the electrical conductivity of seed leachate ( $1.56 \text{ mS cm}^{-1}$ ), total soluble sugars (90.17  $\mu\text{g/ml}$ ), seed damage (1.42 %), seed infection (3.00 %), number of live insects/400 seeds (1.43), number of dead insects/400 seeds (3.14 %) and with C: B ratio of 1: 3.85 after nine months of storage. A successful seed storage management strategy that kept seed quality above MSCS level for up to nine months of storage was the *Metarhizium anisopliae* (CFU:  $1.0 \times 10^8$ ) @ 20 g/kg seed + Diatomaceous earth @ 5g/kg seeds seed treatment.

## Effect of Nano Nitrogen Foliar Spray on Growth and Yield of Tree Mulberry and Cocoon Production

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A study on 'Effect of nano nitrogen foliar spray on growth and yield of tree mulberry and cocoon production' was conducted in the established V-1 tree mulberry garden at KVK, Hassan during 2020-21. On 45<sup>th</sup> days after pruning (days after pruning), significantly higher number of shoots per tree (46.83), number of leaves per tree (658.45) and leaf area (168.05 cm) were recorded in tree mulberry raised with foliar application of nano nitrogen at 4 ml L<sup>-1</sup> on 25<sup>th</sup> and 35<sup>th</sup> DAP+60 % N through soil application. The leaf yield (6340.52 g/tree on 60<sup>th</sup> DAP), leaf moisture (77.56 %), total chlorophyll (2.54 mg/g), leaf elemental except nitrogen and biochemical compositions at 45<sup>th</sup> DAP were highest in foliar spray of nano nitrogen at 4 ml L<sup>-1</sup> on 25<sup>th</sup> and 35<sup>th</sup> DAP+60 % N through soil application. However, significantly shorter fifth instar duration (7.29 days), higher fifth instar larval weight (54.69 g/10 larvae), ERR (97.33 %), single cocoon weight (2.58 g), single cocoon shell weight (0.58 g), cocoon shell ratio (22.55 %), filament length (1435.44 m) and filament weight (0.51 g) were recorded in treatment with foliar application of nano nitrogen at 4 ml L<sup>-1</sup> on 25<sup>th</sup> and 35<sup>th</sup> DAP+60 % N through soil application. Net returns per hectare of tree mulberry (Rs.1,63,340 ha/crop) and B:C ratio (3.17) were more with foliar application of nano nitrogen at 4 ml L<sup>-1</sup> on 25<sup>th</sup> and 35<sup>th</sup> DAP+60 per cent nitrogen through soil application and found to be cost effective as compared to other treatments.

## Assessment of Major Nutrients Requirement for Tree Mulberry under Irrigated Condition in Eastern Dry Zone of Karnataka

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A study on 'Assessment of major nutrients requirement for tree mulberry under irrigated condition in eastern dry zone of Karnataka' was conducted in the established V-1 tree mulberry garden at College of Sericulture, Chintamani during 2020-21. Application of 100 per cent recommended dose of fertilizers + FYM through soil application to tree mulberry garden recorded longest shoot length (48.33, 78.90 and 100.93 cm), number of shoots/tree (30.23, 41.83 and 43), number of leaves/tree (280.10, 485.63 and 738.13) and leaf area (89.20, 121.27 and 136.80 cm<sup>2</sup>), respectively on 15<sup>th</sup>, 30<sup>th</sup> and 45<sup>th</sup> days after application of fertilizers. The leaf yield (4340 g/tree), leaf moisture (75.80%), moisture retention after 6 h. (95.30%), total chlorophyll (2.52 mg/g), leaf elemental and biochemical composition were also highest in the same treatment. However, these parameters were on par with the treatment receiving 75 per cent RDF through water soluble fertilizers + FYM. Further, significantly shorter 5<sup>th</sup> instar larval duration (7.17 days), average moulting duration (96.35 h) and higher grown up larval weight (35.10 g/10 larvae), ERR (94.59%), cocoon weight (2.38 g/cocoon), shell weight (0.53 g/cocoon) and shell percentage (23.55), longest filament length (1185.92 m) and higher Denier (2.63) were recorded in the batches of silkworms reared on the leaves from treatment with 75 per cent NPK through WSF + FYM. The net returns per hectare of mulberry was more (Rs.109725 ha<sup>-1</sup> crop<sup>-1</sup>) in same treatment and was found to be cost effective with higher B:C ratio (2.23) compared to other treatments.

## Effect of Nitrogen Management Practices on Soil Nitrogen Forms, Crop Growth and Yield of Maize and Greengram in *Alfisols*

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THE effect of nitrogen management practices using different organic manures along with inorganic N levels on soil N forms, growth and yield of maize and subsequent greengram crop were studied in the laboratory condition and field during 2019-20 and 2020-21. The field experiment was laid out in randomized complete block design with ten treatments replicated thrice. Incubation study revealed that the content of  $\text{NH}_4^+\text{-N}$  and  $\text{NO}_3^-\text{-N}$  (24.83 and 37.87  $\text{mg kg}^{-1}$ , respectively) was higher in the treatment receiving 75% RDN + 50% RD FYM + 25% RDN through vermicompost in 60 DAI and hydrolysable  $\text{NH}_4^+\text{-N}$ , hexosamine nitrogen, amino acid nitrogen, unidentified hydrolysable-N, total hydrolysable-N, non hydrolysable-N and total nitrogen was found significantly higher in treatment  $T_7$  (50% RDN + 25% RD FYM + 50% RDN through vermicompost) than  $T_1$  and  $T_2$  treatments. The field experiment revealed that significantly higher plant height (207.03 cm), number of leaves (14.63), kernel yield (84.88  $\text{q ha}^{-1}$ ), stover yield (90.00  $\text{q ha}^{-1}$ ), nutrient content and uptake by maize crop were significantly higher in treatment  $T_6$  (75% RDN + 50% RD FYM + 25% RDN through vermicompost). The pooled analysis of post harvest soils of maize and greengram recorded significantly higher available major and secondary nutrients in  $T_7$  treatment. The nitrogen fractions *viz.*,  $\text{NO}_3^-\text{-N}$  and  $\text{NH}_4^+\text{-N}$  content of 13.68  $\text{mg kg}^{-1}$  and 22.11  $\text{mg kg}^{-1}$ , respectively was recorded in treatment  $T_6$  and organic fractions were found significantly higher in treatment ( $T_7$ ). The higher greengram seed yield (8.80  $\text{q ha}^{-1}$ ) and nutrients uptake was recorded due to residual effect of the same treatment ( $T_6$ ). Economic analysis revealed that higher total net returns (Rs.1,47,267) was recorded in  $T_6$  treatment receiving 75% RDN + 50% RD FYM + 25% RDN through vermicompost in maize and greengram crop.

## Evaluation of Potassium Feldspar as an Alternative to Conventional Potassium Sources and its Effect on Growth and Yield of Paddy

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IN this study, the efficiency of potassium feldspar (KF) in nutrient release and its contribution to growth and performance of paddy was compared with conventional potassium sources *viz.*, muriate of potash (MOP), sulphate of potash (SOP) and certified reference material (JF2). Incubation and pot culture experiments were conducted in three different soils *i.e.*, acidic, neutral and alkaline. Potassium feldspar at four different levels (50, 75, 100 and 150  $\text{kg K}_2\text{O ha}^{-1}$ ) were compared with MOP, SOP and JF2 @ 50  $\text{kg K}_2\text{O ha}^{-1}$ . The incubation study revealed that, the available  $\text{K}_2\text{O}$  and available Si (Acetic acid and  $\text{CaCl}_2$  extractable silicon) content of soil increased with the increased levels of KF. However, application of KF@150  $\text{kg K}_2\text{O ha}^{-1}$  showed significantly increased available  $\text{K}_2\text{O}$  and Si with the incubation period and was observed maximum at 120 DAI irrespective of soils over other potassium sources. Among the soils, significantly higher available  $\text{K}_2\text{O}$  and Si were recorded in alkaline soil followed by neutral and acidic soils. Pot culture experiments conducted with paddy as test crop, revealed that application of KF @ 150  $\text{kg K}_2\text{O ha}^{-1}$  enhanced plant height, number of panicles, panicle length, test weight, grain and straw yield irrespective of the soils. Content and uptake of K and Si in crop were also observed to be enhanced with KF application @ 150  $\text{kg K}_2\text{O ha}^{-1}$  over other levels of KF and sources of potassium.

## **Quantification of Runoff and Identification of Suitable Sites for Rainwater Harvesting by Using Geospatial Techniques in Halayapura Micro-Watershed of Tumkur District**

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GEOSPATIAL technique were used for land and water management action plan for Halayapura micro-watershed lies between  $75^{\circ} 51' 37.585''$  to  $75^{\circ} 53' 29.93''$  E longitude and  $13^{\circ} 58' 59.959''$  to  $14^{\circ} 1' 3.722''$  N latitude in Tumkur district of Karnataka. Arc GIS software was used in evaluation of basemap and morphological characteristics in micro-watershed. The present study reveals that drainage pattern was dendritic with trunk order 4.00. The area, maximum length and width of the micro-watershed are 503.00 ha, 4.40 km and 2.20 km, respectively. The mean value of bifurcation ratio was 2.50, it indicates micro-watershed has been suffered less structural disturbance. The value of drainage density is  $0.851 \text{ km}^{-2}$  which indicated that, the region is having permeable subsoil material. The value of form factor indicates micro-watershed is approaching towards elongated shape of watershed. The study was used to estimate runoff and base map was prepared. The estimated runoff available to use is 72.90 mm for annual rainfall of 774.50 mm. The recharge factor found to be 7.00 % and utilizable groundwater is 41.70 mm (70.00 per cent of 59.60 mm recharge estimated). The total available water resource combining the soil moisture storage and utilizable runoff plus recharge is 256.00 mm. Currently about 19.40 per cent of the utilizable runoff is being used and 9.00 per cent of runoff excess is promoting by harvesting and conservation structures.

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