# Studies on Pollination Potentiality of Honey Bee (*Apis cerana* Fab.) in Sunflower Hybrid (RSFH 130) Seed Production

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### **AUTHORS CONTRIBUTION**

Abstract

SANGANNA M. SAJJANAR : Conducted the research and wrote manuscript M. THIPPAIAH : Guidance, manuscript correction A. PRABHURAJ : Supervision of research work and correction of manuscript K. S. JAGADISH : Suggestions for conducting research and manuscript correction

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Received : June 2022 Accepted : August 2022 The study was conducted at Main Agricultural Research Station, UAS, Raichur during rabi 2020-21 to utilize honey bees in sunflower hybrid seed production. Totally, thirty species of floral visitors were recorded on parental lines of sunflower hybrid (RSFH-130), among them 15 were hymenopterans, 7 lepidopterans, 5 coleopterans and 3 dipterans. Apis dorsata was predominant pollinator  $(22.41 \pm 11.51 \text{ bees} / 5 \text{ capitula} / 5$ min.) on CMS parental line (CMS-104 A), followed by A. cerana and least was A. florea. The peak foraging activity of A. dorsata was recorded at 1100-1200 hr with 37 bees, A. cerana recorded two peak foraging activity, one at 1000-1100 hr (22 bees) and another at 1500-1600 hr (18 bees) and A. florea recorded at 1000-1100 hr (8 bees). The maximum Shannon-Wiener index of diversity (H=1.34) of floral visitors on CMS line under open condition was recorded at 0800-0900 hr of the day and least (H=0.84) was at 1700-1800 hr. The same trend was also recorded in restorer parental line (R-630). A. cerana under caged condition (three colonies per acre) without pollen load on CMS line were recorded highest mean number of nectar foragers (72.33±23.01 bees), followed by A. dorsata, A. cerana and A. florea foragers from natural colonies in open pollination condition. Whereas, highest mean number of nectar foragers ( $46.41\pm29.23$ ) of A. cerana under caged condition (three colonies per acre) with pollen load were recorded, followed by A. dorsata, A. cerana and A. florea. Nectar foragers with pollen load on CMS line play an important role in transfer of pollen grains from restorer line to CMS line and carry out effective pollination. A. florea with pollen load spent more foraging duration (8.70 sec/ capitulum) on CMS line, followed by A. cerana and least in case of A. dorsata in open pollination. A. cerana without pollen load spent more nectar foraging duration (23.94) in open pollination compared with A. florea and A. dorsata. Bees without pollen load spent significantly greater nectar foraging duration as compared with bees with pollen load, both in caged and open pollination situations. Significantly highest seed yield per acre (1131.98 kg), per cent filled seeds (97.20 %), test weight (7.52 g), volume weight (40.48 g/100 ml), kernel percentage (79.78 %), highest kernel to husk ratio (3.87) and highest seed quality & associated parameters were recorded in hand + open pollinated plots, followed by the A. cerana @ 3 colonies per acre treated plots, hand pollination, open pollination and least seed quality parameters were recorded in control treatment.

Keywords : Honey bees, Foraging activity, Sunflower pollination

SUNFLOWER hybrid seed production through hand pollination is very difficult task due to shortage of agricultural labour and it is very expensive when there is availability of labour. Thus using honey bees as pollinators for good quality and quantity of sunflower hybrid seed production as an alternative to hand pollination is need of the hour.

Honey bees are the most important insect pollinators of cultivated crops world wide. While some insects visit the flowers of only a small number of plant species. Honey bees will visit all flower from which they can harvest reward. Honey bees are one of the few pollinating insects that can be managed. They can be delivered to a crop when required and various management options available to influence the honey bees' flower visiting behavior (Anonymous, 2012). Bees certainly are essential in seed setting of sunflower because pollen must be transferred from male-fertile to male sterile plants (Hoffman and Chambers, 2006). Many experiments have consistently confined that commercial yield levels could be increased to an extent of 45 to 50 per cent in sunflower, sesamum and niger (Melnichenko and Khalifman, 1960).

The species richness and foraging behavior of honey bees on sunflower is genotype specific and is influenced by morphometric variations of the plant, which includes flower shape, flower structure, head size, floret length, corolla length, stigma pigmentation and many other factors. Whereas, nectar and pollen are the source of food for honey bees, which attract or restrict bee visitation to the host plant flower (Rinku et al., 2017). Sunflower is an allogamic plant which needs insects during flowering for pollination especially honey bees for seed production. Pollen grains of sunflower are heavy and sticky in nature due to this wind plays a very minor role in transfer of pollen from one flower head to other, which can be done by using honey bees (Free, 1963). Both bee colony density and visitation rates increased all the productivity variables. However, effects were nonlinear for visitation rates, there is an optimum (mean of 8-10 visits per flower), beyond which more honey bees are not beneficial (and can even be detrimental) for crop productivity (Rollin and Garibaldi, 2019).

One acre of sunflower hybrid seed production required around 42 skilled labourers for transfer of pollen from male capitulum to cytoplamsic male sterile capitulum. The present study aims to address this important issue in sunflower hybrid seed production by utilizing Indian honey bee, *Apis cerana* Fab. and calculation of benefit:cost ratio of honey bee pollination v/s hand pollination.

### MATERIAL AND METHODS

The present investigation on 'Studies on pollination potentiality of honey bee (*Apis cerana* Fab.) in sunflower hybrid (RSFH 130) seed production' was carried out during 2020-21 at Main Agricultural Research Station, UAS, Raichur (Latitude16<sup>o</sup>12'N and Longitude 77<sup>o</sup>20'E) which is located in the II zone of Karnataka state with following five treatments in six replications, T<sub>1</sub>: Crop caged with three colonies of *Apis cerana* (six frame strength) per acre; T<sub>2</sub>: Hand pollination; T<sub>3</sub>: Open pollination; T<sub>4</sub>: Hand + open pollination; T<sub>5</sub>: Control (crop covered with mosquito nylon net). The parental lines CMS 104A (3 lines) and R-630 (1 line) of sunflower hybrid RSFH-130 were used for hybrid seed production.

Pollinator fauna and their foraging activity on parental lines of sunflower hybrid: In order to record the species abundance and diversity of flower visitor, ad-libitum sampling of five capitula was done for 5 minutes duration hourly intervals, commencing from 0600 hr to 1800 hr.

Diversity of flower visitors on parental lines of sunflower hybrid: Representative samples of flower visitors were collected by various methods of collection viz., visual scanning, sweep net sampling and bee bowls as per the methodology suggested by Belavadi and Ganeshaiah (2013).

The frequency of visits by each species was recorded in order to identify the most abundant species visiting sunflower capitulum. Pollinator population count was used to compute Shannon-Weiner index of diversity (H) by using the following formula:-

### $H = \sum pi x lnpi$

Where, pi is the proportion of the i<sup>th</sup> species

The dominant species on any given sampling day was determined by the Berger-Parker dominance index 'd' which gives the proportion of the total numbers of individuals in a sample that is due to the dominant species and was calculated by

d = ni / NT

Where, ni is the number of individuals of the i<sup>th</sup> species on sampling date and NT is the total number of individuals in the sample (Southwood, 1988).

Foraging activity of honey bee: The pollen and nectar foraging activity of different honey bee species in open plot and foraging activity of Apis cerana Fab. in caged pollinated plots were determined on the capitulum during flowering period for 5 min. The forager bees with pollen pellets in their corbicula (even small loads) were classified as pollen foragers. Similarly, the forager bees without pollen load in their corbicula were classified as nectar foragers and were recorded throughout the study period from 0600 to 1800 hr of the day at hourly interval in three replications and were expressed as number of pollen or nectar foragers per 5 capitula per 5 minutes.

*Time spent by bees on the flower heads:* The time when forager bee landed on the capitulum till leaving was recorded by using a digital stop watch and considered as time spent by the bee/capitulum in open pollination plots for Apis species. Similarly, separate observations were made on nectar and pollen collection in CMS and R line from 0600 up to 1800 hr at an hourly interval during the flowering period. The mean time spent by bee per capitulum is expressed as time (sec) spent/capitulum.

Nectar yield and total soluble solids (TSS): Quantity of nectar and TSS in the nectar produced by the disc florets (n=25) of CMS and R line was estimated. The randomly selected capitula were enclosed with butter paper cover to avoid the visit of pollinators. Next day the covers were removed from the capitula and the quantity of nectar produced per floret was measured from 0600 to 1800 hr at two hourly intervals by using calibrated capillaries/micro syringe. The quantity of nectar collected was expressed in micro litre ( $\mu$ l)/floret (Belavadi and Ganeshaiah, 2013). The quality of nectar in terms of sugar (TSS) content was assessed by using hand refractometer and expressed in percentage.

*Effect of different pollination conditions on seed quality, seed yield and yield attributing characters:* In the field experiment, from each treatment thirty sunflower heads were harvested separately to record following yield attributes.

*Per cent seed filling:* In each head, total numbers of filled and unfilled seeds were counted. The ratio of number of filled seeds to the total number of seeds per head was expressed in percentage.

*Test weight:* Hundred filled seeds were counted from each treatment in all replications and its mean weight was expressed in grams.

*Volume weight:* Hundred ml of filled seeds were collected from each treatment in all replications and its mean weight was expressed in g/100ml.

Kernel to Husk (K/H) Ratio, Husk and Kernel per cent: Collected hundred seeds from each treatments and were manually dehusked. Both weight of kernel and husk were determined to calculate the husk percentage, kernel percentage and kernel to husk ratio.

*Germination per cent:* Hundred seeds were placed on moist blotting paper and kept for observation. The germination counts were taken five days later and per cent seed germination was computed.

*Seed vigour Index:* Ten seedlings from each replication from all treatments were randomly taken to record root and shoot length. Seed vigour index (VI) was calculated by using the following formula

$$VI = RL + SL \times GP$$

Where in,

RL = Root length;

- SL = Shoot length;
- GP = Germination percentage

*Percent oil content:* Oil content of seeds of hybrid sunflower (RSFH-130) produced from different treatments were analysed by using NMR facility at AICRP (Sunflower), Main Agricultural Research Station, UAS, Raichur.

*Statistical analysis:* The data from the field experiment was analyzed statistically for comparing treatments following ANOVA for Randomized Block Design and results were interpreted at 5 per cent level of significance in order to compare the dependency of sunflower hybrid on honey bee pollination for per cent seed filling, test weight, germination percentage and oil content over other treatments.

Calculation of cost benefit ratio of honey bee pollination v/s hand pollination: The benefit cost ratio of the different modes of pollination was calculated for hybrid seed production by considering the current cost of inputs.

### **RESULTS AND DISCUSSION**

*Floral visitor on parental lines:* Thirty species of floral visitors were recorded on parental lines of the sunflower hybrid (RSFH-130), of which 15 species were from five families of Hymenoptera (Apidae, Vespidae, Halictidae, Megachilidae and Sphecidae), seven species from four families of Lepidoptera

Order	Family	Sl. No.	Scientific name	Forage collected
Hymenoptera	Apidae	1	Apis dorsata Fabricius	N+P
		2	Apis cerana indica Fabricius	N+P
		3	Apis florea Fabricus	N+P
		4	Xylocopa aestuans (Linnaeus)	N+P
		5	Xylocopa fenestrata (Fabricius)	N+P
		6	Amegilla sp.	N+P
		7	Unidentified sp.	Ν
Halictidae		8	Lassooglossum sp.	Ν
		9	Unidentified sp.	Ν
Vespidae		10	Vespa tropica (Linnaeus)	Ν
		11	Ropalidia marginata (Lepeletier)	Ν
		12	Poslistes sp.	Ν
Sphecidae		13	Unidentified sp.	Ν
Megachilidae		14	Megachile disjuncta (Fabricius)	N+P
		15	Megachile lanata (Fabricius)	N+P
Lepidoptera	Sphingidae	16	Unidentified sp.	Ν
Nymphalidae		17	Danaus chrysippus Linnaeus	Ν
		18	Junonia lemonias (Linnaeus)	Ν
		19	Tirumala limniace (Cramer)	Ν
Pieridae		20	Catopsilia sp.	Ν
Erebidae		21	Amata passalis (Fabricius)	Ν
		22	Amata cyssea (Stoll)	Ν
Coleoptera	Scarabaeidae	23	Gametis versicolor (Fabricius)	N+P
Coccinellidae		24	Coccinella transversalis Fabricius	N+P
		25	Chilomenes sexmaculata Fabricius	N+P
Chrysomelidae		26	Monolepta sp.	N+P
		27	Leptisma sp.	N+P
Diptera	Syrphidae	28	Eristalinus sp.	N+P
Sarcophagidae		29	Unidentified sp.	Ν
Muscidae		30	Unidentified sp.	Ν

# TABLE 1 List of floral visitors on parental lines of sunflower hybrid

(Nymphalidae, Erebidae, Pieridae and Spingidae), five species from three families of Coleoptera (Chrysomelidae, Coccinellidae and Scarabaeidae) and one each species from three families of Diptera (Syrphidae, Sacrophagidae and Muscidae) (Table 1). Similarly, Jadhav *et al.* (2011) from Thirupathi, who recorded two families of Hymenoptera, four families of Lepidoptera and three families of Coleoptera and one family of Diptera visiting sunflower capitulum. Goswami *et al.* (2013) recorded 12 species of insect visitors, of which nine belonged to Hymenoptera, one to Diptera, one to Lepidoptera and one was a Coleopteran species at Pantanagar.

Abundance of floral visitors on parental lines RSFH-130: On CMS-104A line, A. dorsata was predominant pollinator (22.41±11.51 bees / 5 capitula / 5 min.), followed by the A. cerana and least was A. florea. The peak foraging activity of A. dorsata was recorded at 1100-1200 hr with 37 bees / 5 capitula / 5 min, A. cerana recorded two peak foraging activity pattern, one at 1000-1100 hr and another at 1500-1600 hr and A. florea recorded peak foraging activity at 1000-1100 hr (Table 2). Among non-Apis species, hymenopterans were the predominant (3.66±2.90 bees/ 5 capitula / 5 min.) floral visitors and their peak foraging activity was at 1000-1100 hr, followed by lepidopterans, dipterans and lowest was recorded in case of coleopterans. In case of fertility restorer line (R-630) A. dorsata was the predominant pollinator  $(35\pm13.84 \text{ bees} / 5 \text{ capitula} / 5 \text{ min.})$ , followed by the A. cerana and least was A. florea. The peak foraging activity of A. dorsata was recorded at 1200-1300 hr with 57 bees / 5 capitula / 5 min, A. cerana recorded two peak foraging activity at 1100-1200 hr and at 1500-1700 hr and A. florea recorded peak foraging activity at 1100-1200 hr (Table 3). Among non-Apis species, hymenopterans were predominant  $(1.75\pm1.13 \text{ bees } /$ 5 capitula / 5 min.) and peak foraging activity was observed at 1000-1100 hr, followed by lepidopterans, coleopterans and lowest was dipterans. The corolla length of disc florets of CMS line was 4.52 mm and R line was 5.32 mm. The variation in abundance, diversity and dominance of pollinators on parental lines could be due to the variation in availability of rewards (pollen and nectar) and variation in corolla length of disc florets. The present findings on abundance and foraging hours are in agreement with the findings of Rangarajan *et al.* (1974) who showed that, *A. cerana* and *A. florea* were the most frequent visitors of sunflower capitula and maximum visitation was made during 0600-1100 hr, whereas comparatively lowest activity was observed during 1200-1430 hr of the day. The activity of the pollinators was more frequent in the forenoon (0900-1100 hr) and in the later afternoon (1600-1700 hr) (Delaude *et al.*, 1978).

Diversity and dominance of floral visitors on parental lines of RSFH-130 : The maximum Shannon-Wiener index of diversity (H) of floral visitors (H=1.34) under open condition was recorded at 0800-0900 hr of the day and least (H=0.84) was recorded at 1700-1800 hr. The highest (d=0.503) Berger Parker dominance index between the floral visitors were recorded in case of A. dorsata and lowest was in case of coleopterans (d=0.005), whereas between the hours of the day, highest dominance was recorded at 1300-1400 hr (0.67) and lowest was recorded at 0800-0900 hr (0.43) on CMS parental line (CMS-104A) (Table 2). In case of fertility of restorer parental line (R-630), maximum Shannon-Wiener index of diversity (H) of floral visitors (H=1.27) on fertility restorer parental line in open condition was recorded at 0900-1000 hr of the day and least (H=0.89) was recorded at 1700-1800 hr. The highest (d=0.44) Berger Parker dominance index between the floral visitors of R-630 was recorded with A. dorsata and lowest with dipterans (d=0.003), whereas highest dominance between the hours of the day was recorded at 1400-1500 hr (0.53) and lowest at 1000-1100 hr (0.40) (Table 3). The findings of the present study on diversity indices are similar to the findings of Biswanath and Kakali (2015) who reported the Shannon-Wiener diversity index H was calculated and found to be 1.49 for order Hymenoptera, 1.4 for the species of order Diptera and 1 for the species under order Lepidoptera in West Bengal.

Foraging activity of Apis species on CMS parental line (CMS-104A) : The highest mean number of nectar foragers (72.33±23.01 bees/5 capitula/5 min.)

		Abund	ance, diver	sity and dominan of sunflower hybı	TABLE 2 ce of floral vi id (RSFH-13	2 sitors on CMS 0) under open	parental line ( condition	CMS-1	04A)		
			Numb	er of floral visitors/5	capitula/5 min.						
Time (hrs)		Apis species			Non-Apis	species		Total	",H"Value	% composition	"d" value
	Apis cerana	Apis dors ata	Apis florea	Hymenoptera	Diptera	Lepidoptera	Coleoptera				
0600-0700	6	10	1	0	0	0	1	21	1.01	3.93	0.47
0200-0800	18	19	3	2	0	0		<del>4</del>	1.14	8.05	0.44
0800-0900	20	25	5	4	-		TU	57	1.34	10.67	0.43
0900-1000	21	30	7	5	7	6/	0	65	1.29	12.17	0.46
1000-1100	8	36	8	8	2 /15 L	1	0	<u>7</u> 6	1.30	14.23	0.47
1100-1200	13	37	7	9	0	0	0	63	1.11	11.80	0.58
1200-1300	10	32	L	5 L 3	0	0	0	<b>3</b> 6	1.15	10.49	0.57
1300-1400	7	31	2	9	0	0	0	4	0.95	8.61	0.67
1400-1500	10	33	1	4	1		0	<del>9</del>	1.17	7.49	0.57
1500-1600	18	16	1		0	1	0	37	1.01	6.93	0.48
1600-1700	11	8	1		0	0	0	21	66.0	3.93	0.52
1700-1800	9	2	1	0	0	0	0	6	0.84	1.69	0.66
Total	165	269	4	44	4	5	3	534			
$Mean \pm SD 1$	3.75 ± 5.72.	$22.41 \pm 11.513$	$.66 \pm 2.90$	$3.66\pm2.80$	$0.33\pm0.49$	$0.41\pm0.51$	$0.25\pm0.45$				
%Compositi	on30.89	50.37	8.23	8.23	0.74	0.93	0.56			100	
"d" value	0.308	0.503	0.082	0.082	0.007	0.00	0.005				

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30)		/alue % composition "d" value		.10 4.93 0.42	.20 6.39 0.42	.25 8.28 0.43	.27 10.48 0.42	.20 12.89 0.40	.10 13.63 0.43	.05 12.37 0.48	.14 8.81 0.46	.14 6.71 0.53	.11 6.71 0.46	.05 5.87 0.46	0.89 2.93 0.50			100
line (R-6		"Total "H"		47	61	62	100	123	130	118	22	2	2	56	38	954		
restorer parental condition			Coleoptera	0			07	0	0	0	0		0	0	0	4	$0.33\pm0.49$	0.41 0.004
s ors on fertility 0) under open o		· species	Lepidoptera	0	7		E Y		0	0		0	0	15.5	0	9	$0.50 \pm 0.52$	0.62 0.006
TABLE 3 of floral visiti id (RSFH-13	capitula/5 min.	Non-Apis	Diptera	0	0	0	° /=		0	0	0	0		0	0	3	$0.25\pm0.45$	0.31 0.003
nd dominance sunflower hybr	of floral visitors/5	1	Hymenoptera	2		ю	3	4	2		大部長いと	2		2	0	21	$1.75 \pm 1.13$	2.20 0.022
ce, diversity a of s	Number o		Apis florea	5	8	12	16	18	22	18	15	6	8	4	2	137	$11.41 \pm 6.37$	14.36 0.143
Abundanc		Apis species	Apis dorsata	8	26	\$	36	50	56	57	39	\$	30	26	12	420	$35 \pm 13.84$	44.02 0.440
			Apis cerana	20	24	28	4	49	50	43	28	18	24	24	14	363	.25 ± 12.30	on38.05 0.380
		Time (hrs)		0600-0700	0700-0800	0060-0080	0900-1000	1000-1100	1100-1200	1200-1300	1300-1400	1400-1500	1500-1600	1600-1700	1700-1800	Total	Mean $\pm$ SD30	% Compositic "d" value

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of A. cerana under caged condition (three colonies per acre) without pollen load on cytoplasmic male sterile parental line (CMS-104A) of sunflower hybrid (RSFH-130) were recorded, followed by A. dorsata, A. cerana and A. florea foragers from natural colonies in open pollination condition, whereas, highest mean number of nectar foragers (46.41±29.23 bees / 5 capitula / 5 min.) of A. cerana under caged condition (three colonies per acre) with pollen load were recorded, followed by A. dorsata (3.33±2.96 bees / 5 capitula / 5 min.), A. cerana (2.91±2.19 bees /5 capitula / 5 min.) and A. florea foragers from natural colonies in open pollination condition (Table 4). The highest number of nectar foragers of A. cerana with and without pollen load in caged condition (three colonies per acre) was recorded between 0800-0900 hr (97 and 101 bees / 5 capitula / 5 min., respectively) and

1400-1600 hr (38 and 96 bees / 5 capitula / 5 min., respectively) of the day. But there was no significant difference in number of nectar foragers with and without pollen load in caged condition. Foragers from natural colonies of A. cerana with pollen load recorded two peaks of nectar foraging activity at 0700-0800 hr (7 bees / 5 capitula / 5 min.) and 1500-1600 hr (4 bees / 5 capitula / 5 min.). Whereas, A. cerana without pollen load recorded two peaks at 1000-1100 hr and 1500-1600 hr of the day. A. dorsata and A. florea with pollen load recorded peak nectar foraging activity at 0900-1100 hr and A. dorsata and A. florea without pollen load recorded peak foraging activity at 1100-1200 hr of the day. Nectar foragers with pollen load on CMS lines are plays important role in transferring of pollen grains from restorer line to cytoplasmic male sterile line and carried out effective

#### TABLE 4

Nectar foragers of Apis species on CMS parental line (CMS-104A) of sunflower hybrid (RSFH-130) under caged (3 colonies /acre) pollination as compared with open condition

		30	Numbe	r of floral visito	rs/5 capitula/5	5 capitula/5 min.					
Time (hrs)	Foragers under cag	of <i>A.cerana</i> ed condition		Forager	s of <i>Apis</i> speci	ies from natural	colonies				
	(3 colo	nies/acre)	Apis	cerana	Apis	dorsata	Api	s florea			
	Bees with Pollen load	Bees without pollen load	Bees with Pollen load	Bees without pollen load	Bees with Pollen load	Bees without pollen load	Bees with Pollen load	Bees without pollen load			
0600-0700	55	56	3	6	2	8	0	1			
0700-0800	86	96	7	11	4	15	1	2			
0800-0900	97	101	6	14	6	19	2	3			
0900-1000	75	96	5	16	6	24	3	4			
1000-1100	64	83	3	19	8	28	2	6			
1100-1200	44	56	2	11	7	30	1	6			
1200-1300	32	58	1	9	4	28	1	6			
1300-1400	21	85	1	6	3	28	1	1			
1400-1500	11	96	2	8	0	23	0	1			
1500-1600	38	56	4	14	0	16	0	1			
1600-1700	26	54	1	10	0	8	0	1			
1700-1800	8	31	0	6	0	2	0	1			
Total	557	868	35	130	40	229	11	33			
Mean $\pm$ SD	$46.41 \pm 29.23$	$72.33 \pm 23.01$	2.91±2.19	$10.83 \pm 4.21$	$3.33 \pm 2.96$	$19.08 \pm 9.34$	$0.91\pm0.99$	$2.75 \pm 2.17$			
t-test @ 5%	1	NS	*		×	¢		*			

NB : \* Significant at p<0.05; NS - Non significant at p<0.05

pollination. There are two peak nectar foraging activity of honey bees during 0800-1100 hr in the morning and 1400-1600 hr in the evening. There is significant difference in number of nectar foragers with and without pollen load in open pollination. But there is no significant difference in number of nectar foragers of A. cerana with and without pollen load inside the cage with 3 colonies per acre. That leads to effective transfer of pollen from restorer line to the cytoplasmic male sterile lines.

Foraging activity of Apis species on fertility restorer parental line (R-630): Pollen and nectar foragers (no. of bees / 5 capitula / 5 min.) on restorer line under caged (3 colony / acre) and open condition was recorded. *A. cerana* recorded highest mean number of pollen foragers (99.66±48.13 bees / 5 capitula / 5

min.) compared with nectar foragers (60.25±23.11 bees / 5 capitula / 5 min.) under caged condition (three colonies per acre), whereas in open condition A. *cerana* was the more abundant pollen forager and A. dorsata was the more abundant nectar forager compared with other Apis species on restorer line R-630 and least pollen and nectar foragers were recorded with A. florea (Table 5). A. cerana recorded two peaks of pollen foraging activity in caged at 0800-0900 hr and 1600-1700 hr (160 bees and 125 bees / 5 capitula / 5 min., respectively) and open condition at 1000-1100 hr and 1600-1700 hr (36 bees and 18 bees /5 capitula/5 min., respectively), whereas, two nectar foraging activity of A. cerana in caged condition at 0900-1000 hr and 1600-1700 hr (86 bees and 51 bees /5 capitula/5 min., respectively) and in open condition only one peak in nectar foraging activity was recorded at 1100-1200 hr with 16 bees / 5 capitula / 5 min. A.

I ABLE 5
Pollen and nectar foragers of Apis species on fertility restorer parental line (R-630)
under caged (3 colonies/acre) and open condition

			Nu	mber of bees /	5 capitula / 5 n	nin.		
Time (hrs)	Forager	s of A.cerana	Vana	Forag	ers of Apis spec	cies from natura	al colonies	
	under ca (3 col	ged condition onies/acre)	Api	s cerana	Api.	s dorsata	Apis	sflorea
	Pollen	Nectar	Pollen	Nectar	Pollen	Nectar	Pollen	Nectar
0600-0700	101	68	16	4	10	10	4	1
0700-0800	150	83	18	6	12	14	6	2
0800-0900	160	84	20	8	16	18	8	4
0900-1000	150	86	32	10	18	18	10	6
1000-1100	143	83	36	13	28	22	12	6
1100-1200	108	80	34	16	32	24	14	8
1200-1300	65	49	30	12	36	21	12	6
1300-1400	26	40	18	10	21	18	10	5
1400-1500	21	34	12	6	18	16	6	3
1500-1600	73	45	16	8	16	14	6	2
1600-1700	125	51	18	6	12	14	4	0
1700-1800	74	20	10	4	6	6	2	0
Total	1196	723	260	103	225	195	94	43
Mean± SD	99.66±48.13	$360.25 \pm 23.11$	$21.66 \pm 8.89$	$8.58 \pm 3.72$	$18.75 \pm 9.09$	$16.25\pm5.08$	$7.83 \pm 3.76$	$3.58 \pm 2.64$
t-test @ 5%		*	*	k	1	VS		*
t-test (a) 5%		* NB :	* Significant a	nt p<0.05; NS-	Non significan	v.5 t at p<0.05		Ύ

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dorsata and A. florea recorded only one peak in pollen and nectar foraging activity between 1100-1300 hr of the day. There were significant differences in the number of nectar and pollen foragers of A. cerana on R-630 parental line in caged condition and A. cerana, A. florea of natural colonies in open pollination. But, there was no significant difference between number of pollen and nectar foragers of A. dorsata in open pollination.

The variation in the abundance of insect pollinators, nectar foragers and pollen foragers on restorer lines of selected sunflower hybrids was mainly due to variation in corolla length of disc florets, variation in the availability of nectar quantity and sugar concentration, availability of pollen in disc florets. Similar findings were also reported by Rajasri *et al.* (2012) wherein female parental (CMS lines) of sunflower hybrids were visited mainly by major nectar collectors due to greater availability of nectar. Greenleaf and Kremen (2006) observed that, pollen foragers of *A. mellifera* were more abundant on male fertile flowers and the nectar foragers were more abundant on male sterile flowers. The higher activity of the bees in the morning may be attributed to the abundant availability of pollen and also nectar in the sunflower heads. Pollen availability gradually decreased due to pollen foraging by honey bees,

TABLE 6 Nectar foraging duration of Apis species on CMS parental line (CMS-104A) under caged pollination as compared with open pollination

			Mean time	(sec/capitulum)	spent for nec	tar collection		
Time (hrs)	Foragers	of A.cerana	ISV I	Forager	s of <i>Apis</i> speci	ies from natural	colonies	
	(3 colo	nies/acre)	Apis	cerana	Apis	dorsata	Api	s florea
	Bees with Pollen load	Bees without pollen load	Bees with Pollen load	Bees without pollen load	Bees with Pollen load	Bees without pollen load	Bees with Pollen load	Bees without pollen load
0600-0700	3.60 <sup>j</sup>	3.60 <sup>k</sup>	1.20 <sup>g</sup>	4.73 <sup>h</sup>	1.42 f	2.13 h	0	5.59 <sup>i</sup>
0700-0800	7.06 <sup>i</sup>	9.00 j	4.13 f	11.36 g	1.68 f	7.48 g	5.92 °	12.51 <sup>h</sup>
0800-0900	14.80 g	23.60 <sup>i</sup>	7.04 °	16.68 f	3.13 °	12.18 f	11.25 <sup>d</sup>	19.13 <sup>f</sup>
0900-1000	30.93 a	53.60 ª	11.60 <sup>b</sup>	26.82 de	5.50 d	22.18 d	15.56 bc	30.54 <sup>d</sup>
1000-1100	30.53 b	48.00 <sup>b</sup>	14.24 ª	38.56 ª	7.90 °	32.36 ª	18.81 <sup>a</sup>	40.12 a
1100-1200	29.60 °	47.20 <sup>b</sup>	12.67 <sup>b</sup>	36.21 ª	9.50 <sup>b</sup>	30.47 ab	16.89 <sup>b</sup>	37.85 <sup>ab</sup>
1200-1300	25.60 d	45.00 °	11.53 <sup>b</sup>	35.10 ab	10.55 ª	28.42 <sup>b</sup>	14.19 °	35.90 <sup>b</sup>
1300-1400	20.34 °	43.20 d	10.36 °	31.58 bc	8.79 <sup>b</sup>	25.51 °	12.18 d	33.23 °
1400-1500	16.26 f	41.34 °	9.40 cd	30.14 <sup>cd</sup>	0	21.62 d	0	29.84 <sup>d</sup>
1500-1600	$16.60^{\rm \ f}$	<b>39.94</b> f	8.68 d	24.20 °	0	18.44 °	0	22.56 °
1600-1700	11.66 <sup>h</sup>	38.13 g	6.40 °	19.43 f	0	10.71 <sup>f</sup>	0	16.16 <sup>g</sup>
1700-1800	6.86 <sup>i</sup>	29.74 <sup>h</sup>	0	12.52 g	0	7.49 g	0	0
Mean	17.82	35.19	8.10	23.94	4.03	18.24	8.70	23.61
Sem±	0.12	0.33	0.54	1.32	0.25	0.80	0.58	0.88
CD @ 5%	0.35	0.98	1.61	3.91	0.75	2.38	1.73	2.60
CV(%)	1.16	1.63	11.65	9.59	11.01	7.66	12.91	6.46
t-test @ 5%		*		*		*		*

The values with same superscript in a column do not differ significantly by DMRT NB : \* Significant at p<0.05

			Nun	nber of bees / 5	capitula / 5 min	1.		
Time (har)	Foragers	of A.cerana		Forage	rs of Apis specie	es from natural	colonies	
Time (nrs)	under cag (3 colo	nies/acre)	Apis	cerana	Apis a	lorsata	Apis	florea
	Pollen	Nectar	Pollen	Nectar	Pollen	Nectar	Pollen	Nectar
0600-0700	21.80 f	2.80 <sup>j</sup>	1.85 <sup>h</sup>	2.66 <sup>i</sup>	1.64 °	1.60 <sup>h</sup>	2.28 h	3.55 g
0700-0800	24.13 °	7.20 <sup>i</sup>	5.80 <sup>g</sup>	8.09 h	3.25 cde	5.64 g	7.84 fg	9.86 f
0800-0900	25.93 d	21.40 h	9.44 f	13.50 fg	5.27 <sup>cd</sup>	10.61 f	13.06 °	15.86 °
0900-1000	27.54 °	48.06 a	15.24 <sup>cd</sup>	24.15 d	8.58 b	18.57 de	16.94 d	25.81 d
1000-1100	29.74 <sup>b</sup>	46.47 <sup>b</sup>	17.91 <sup>a</sup>	35.91 a	10.24 <sup>b</sup>	31.88 a	20.02 a	37.77 ª
1100-1200	31.73 a	43.60 °	17.24 ª	33.89 ab	13.01 <sup>a</sup>	28.23 b	18.77 <sup>b</sup>	36.82 ab
1200-1300	26.80 d	41.67 <sup>d</sup>	16.85 ab	32.20 b	14.53 <sup>a</sup>	28.17 <sup>b</sup>	18.71 bc	34.15 bc
1300-1400	24.40 °	38.93 °	15.53 bc	29.58 °	10.55 <sup>b</sup>	24.16 °	17.09 <sup>cd</sup>	32.30 °
1400-1500	22.20 f	38.60 °	14.08 d	25.28 d	10.54 <sup>b</sup>	21.27 <sup>cd</sup>	15.51 <sup>d</sup>	27.90 <sup>d</sup>
1500-1600	19.80 g	35.86 <sup>f</sup>	11.08 e	20.53 °	8.26 b	16.16 °	12.64 °	0
1600-1700	13.93 <sup>h</sup>	30.13 g	8.26 f	15.86 f	5.57 °	9.57 f	8.15 f	0
1700-1800	8.80 <sup>i</sup>	21.66 h	6.20 <sup>g</sup>	11.86 <sup>g</sup>	3.15 de	6.44 <sup>g</sup>	6.34 <sup>g</sup>	0
Mean	23.06	31.36	10.93	21.12	7.88	16.85	13.11	24.89
Sem±	0.34	0.39	0.45	0.75	0.79	1.05	0.56	0.91
CD@5%	1.03	1.15	1.33	2.24	2.34	3.12	1.65	2.69
CV (%)	2.61	2.15	6.75	6.22	17.43	10.87	7.40	7.72
t-test @ 5%		*		*	N	5		*

TABLE 7

Foraging duration of Apis species on fertility restorer parental line (R-630) under caged and open pollination

NB:\*Significant at p<0.05; NS-Non significant at p<0.05

The values with same superscript in a column do not differ significantly by DMRT

thereby resulting in decreased bee activity. In the evening probably the stigma would have pushed remaining pollen out of anther tube resulting in increased bee visits to sunflower heads (Singh, 1977).

Foraging duration of Apis species on CMS parental line (CMS-104A): The mean nectar foraging duration with pollen load of A. florea spent more foraging duration (8.70 sec/flower) on the flowers of CMS parental line, followed by A. cerana and least duration was recorded in case of A. dorsata in open pollination. Whereas, maximum nectar foraging duration (17.82 sec/capitulum) was recorded by A. cerana in caged pollination (3 colonies per acre) (Table 6). A. cerana without pollen load spent more mean nectar foraging duration (23.94 sec/capitulum) in open pollination compared with *A. florea* (23.61 sec/capitulum) and *A. dorsata* (18.24 sec/capitulum). But in caged condition (3 colonies of *A. cerana* / acre) it recorded maximum nectar foraging duration of 35.19 sec/ capitulum. Bees without pollen load spent significantly greater nectar foraging duration as compared with bees with pollen load, both in caged and open pollination situations.

Foraging duration of Apis species on fertility restorer parental line (R-630): The maximum mean pollen foraging duration (13.11 sec/capitulum) was recorded by the *A. florea* in open pollination, followed by the A. cerana and least was recorded in case of A. dorsata on the flowers of fertility restorer parental line. The peak pollen foraging duration was recorded

		Sunflower hybrid	1 (RSFH-130)	
	Cytoplasmic male steril	e line (CMS-104A)	Restorer line	(R-630)
l ime (hrs)	Nectar (µl/floret)	TSS (%)	Nectar (µl/floret)	TSS (%)
0600	1.00 g (0.00)	1.00 <sup>f</sup> (0.00)	1.00 g (0.00)	1.00 <sup>f</sup> (0.00)
0800	1.09 <sup>f</sup> (0.18)	5.22 ° (26.26)	1.01 <sup>f</sup> (0.03)	5.01 ° (24.10)
1000	1.21 ° (0.45)	5.76 <sup>d</sup> (32.28)	1.08 ° (0.16)	5.54 <sup>d</sup> (29.72)
1200	1.59 ° (1.54)	6.30 ° (38.73)	1.19 <sup>a</sup> (0.40)	6.12 ° (36.54)
1400	1.45 <sup>b</sup> (1.11)	7.06 <sup>b</sup> (48.84)	1.16 <sup>b</sup> (0.34)	6.82 <sup>b</sup> (45.50)
1600	1.33 <sup>d</sup> (0.76)	7.30 ° (52.33)	1.11 <sup>d</sup> (0.24)	7.05 ° (48.71)
1800	1.41 ° (0.98)	7.04 <sup>b</sup> (48.65)	1.14 ° (0.29)	6.84 <sup>b</sup> (45.82)
Mean	1.29	5.66	1.09	5.48
Sem±	0.002	0.017	0.001	0.028
CD @ 5%	0.006	0.053	0.003	0.08
CV(%)	0.259	0.517	0.130	0.88

## TABLE 8 Nectar secretion and its total soluble solids (TSS) content in the flowers of parental lines of sunflower hybrid (n=25 florets)

The values with same superscript in a column do not differ significantly by DMRT Values outside the parenthesis are square root transformed values

### TABLE 9

# Effect of different modes of pollination on hybrid seed yield and yield attributing characters of sunflower hybrid (RSFH-130)

	Treatments	Seed yield (kg/ha)	Per cent filled seeds	Test weight (g)	Volume weight (g)	Kernel (%)	Husk (%)	K : H Ratio
T <sub>1</sub>	: <i>A. cerana</i> 3 colonies/acre	1082.30 <sup>b</sup>	94.6 <sup>b</sup>	7.19 <sup>b</sup>	37.68 <sup>b</sup>	75.75 в	24.25 d	3.12 <sup>b</sup>
T <sub>2</sub>	: Hand pollination	1033.83 °	94.2 <sup>b</sup>	6.86 °	38.78 <sup>b</sup>	73.58 °	26.42 °	2.81 °
T <sub>3</sub>	: Open pollination	638.24 <sup>d</sup>	88.6 °	4.24 <sup>d</sup>	30.12 °	69.76 <sup>d</sup>	30.24 ь	2.30 d
T <sub>4</sub>	: Hand +Open pollination	1131.98 ª	97.2 ª	7.52 ª	40.48 <sup>a</sup>	79.78 ª	20.22 °	3.87 <sup>a</sup>
T <sub>5</sub>	: Control	154.14 <sup>e</sup>	0 <sup>d</sup>	1.02 e	12.26 <sup>d</sup>	0 <sup>e</sup>	100 a	0 e
	Mean	808.09	74.92	5.36	31.86	59.70	40.30	2.43
	Sem±	9.62	0.40	0.06	0.41	0.70	0.67	0.11
	CD @ 5%	29.09	1.21	0.19	1.24	2.12	2.04	0.33
	CV(%)	2.66	1.20	2.66	2.89	2.62	3.75	10.28

The values with same superscript in a column do not differ significantly by DMRT

between 1000-1300 hr of the day (Table 7). The duration for nectar collection was also recorded in same trend with *A. florea* (24.89 sec/capitulum), *A. cerana* (21.12 sec/capitulum) and *A. dorsata* (16.85 sec/ capitulum). The pollen (23.06 sec/capitulum) and nectar (31.36 sec/capitulum) foraging duration by the *A. cerana* in caged pollination (3 colonies/acre) registered highest mean foraging duration as compared with the which spent time for collection of pollen and nectar in open pollination. The time spent by *A. cerana* in caged condition and A. florea in open pollination for nectar foraging was on par with pollen foraging. *A. dorsata* and *A. cerana* recorded significantly higher nectar foraging duration as compared with pollen duration in open pollination.

The time spent by A. cerana in caged condition and A. florea in open pollination for nectar foraging was on par with pollen foraging. A. dorsata and A. cerana recorded significantly greater nectar foraging duration compared with pollen foraging duration in open pollination. The present findings were in agreement with findings of Panda et al. (1988) reported that, Apis cerana was the most dominant pollinator with maximal foraging activity at 1100 hr and 1400 hr while A. dorsata and A. florea were present at lower density. A. florea spent highest time (44.1 sec), followed by A. cerana (34.54 sec) and A. dorsata (31.9 sec). Ramya et al. (2014) reported that A. dorsata spent only 1.5 minutes as these bees swiftly collected floral rewards. They could also visit several flowers in a shorter time, making them efficient pollen vectors. The floral handling time was more for nectar collection (90 sec) than pollen collection (30 sec).

Nectar secretion and its total soluble solids (TSS) content in the nectar of parental lines: Nectar secretion/production by both the parental lines of sunflower hybrid was recorded. 1.29 and 1.09 µl/floret was produced by CMS 104A and R 630, respectively. Cytoplasmic male sterile parental lines produce more quantity of nectar as compared with fertility restorer line. CMS parental line attract more number of nectar foragers compared with fertility restorer parental line (Table 8). The mean total soluble solids (TSS) of nectar produced by CMS 104-A and R 360 lines was recorded 5.66 and 5.48 per cent, respectively. Gowda *et al.* (2003) reported that the mean nectar yield per floret of various sunflower genotypes showed a significant differences. The mean nectar yield per floret ranged from 0.21 mg (265R) to 0.59 mg (586R). CMS lines were found to have more mean nectar content as compared with R lines of sunflower (0.4 and 0.37 mg/ floret, respectively). Bees have differential preference for one of the two parental lines. This difference can be explained by differences between lines for nectar production and for concentration and quality of sugars (Basualdo *et al.*, 1999)

Effect of different modes of pollination on hybrid seed yield, yield attributing characters, seed quality and its associated parameters: The significantly highest seed yield per hectare (1131.98 kg), per cent filled seeds (97.20%), test weight (7.52 g), volume weight (40.48 g/100 ml), kernel percentage (79.78%) and highest kernel to husk ratio (3.87) was recorded with hand+open pollination treatment, followed by A. cerana @ 3 colonies per acre (1082.30 kg, 94.60, 7.19g, 37.68g/100ml, 75.75% and 3.12 K:H ratio, respectively), hand pollination, open pollination and least was recorded with control (Table 9). The highest germination percentage (97.2%), shoot length (10.44 cm), root length (18.20 cm), seed vigour index (1033.49) and oil content (37.20%) was recorded in hand + open pollination plots, followed by the A. cerana @ 3 colonies per acre treated plots (94.6%, 9.98 cm, 17.15 cm, 962 and 37.78%), hand pollination, open and pollination and least seed quality and its associated parameters were recorded in control treatment (Table 10). The numerically highest oil percentage content (37.78%) was recorded in A. *cerana* (a) 3 colonies per acre treated plot, compared with open+hand pollination treatment (37.20%), these two were statistically on par with each other and superior over other treatments, followed by hand pollination (36.30%), open pollination (26.11%) and least was recorded with control (10.17%). Similar results were reported by Singh et al. (1998) who conducted an experiment on effect of bee pollination in sunflower with three treatment *i.e.*, pollination without insects, open pollination and bee (A. cerana

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	Treatments	Germination (%)	Shoot length (cm)	Root length (cm)	Seed vigour Index	Oil content (%)	B:C Ratio
T <sub>1</sub>	: A. cerana 3 colo	onies/acre 94.6 <sup>b</sup>	9.98 b	17.15 <sup>b</sup>	962.00 b	37.78 a	1.92:1
$T_2$	: Hand pollination	n 94.2 <sup>b</sup>	9.13 °	15.91 °	875.79 °	36.30 <sup>b</sup>	1.72:1
T <sub>3</sub>	: Open pollination	n 88.6 °	8.23 <sup>d</sup>	14.43 d	759.23 d	26.11 °	1.55:1
T <sub>4</sub>	: Hand +Open pol	lination 97.2 <sup>a</sup>	10.44 <sup>a</sup>	18.20 a	1033.49 ª	37.20 ª	1.89:1
T <sub>5</sub>	: Control	0 <sup>d</sup>	0 e	0 e	0 e	10.17 <sup>d</sup>	0.37:1
	Mean	74.92	7.55	13.13	726.10	29.51	1.49
	Sem±	0.40	0.10	0.30	12.41	0.27	
	CD @ 5%	1.21	0.31	0.90	37.52	0.83	
	CV(%)	1.20	3.00	5.10	3.82	2.10	

TABLE 10 Effect of different modes of pollination on seed quality, associated parameters and B:C ratio of sunflower hybrid (RSFH-130)

The values with same super script in a column do not differ significantly by DMRT

himalaya) pollination. Crop pollinated by honey bees enhanced seed set (23.43%) compared to pollination without insects and 18.31 per cent compared to open pollination. The increase in seed yield in bee pollinated treatment was 80.49 per cent greater than pollinated without insects and 11.75 per cent than open pollination. The germination of seeds from bee pollinated plants was 30.41 per cent higher than seeds from plants without insects and 5.35 per cent higher than open pollinated plants. Rajagopal *et al.* (1999) reported that in the case of the female parent of sunflower hybrid, the filled seed weight, seed filling percentage and seed oil content were highest in the plot pollinated only by honey bees.

Benefit: cost ratios of different modes of pollination: The numerically highest benefit: cost ratio was recorded in *A. cerana* (*a*) 3 colonies per acre (1.92:1), followed by hand + open pollination treatment (1.89:1), hand pollination (1.72:1), open pollination (1.55:1) and control (0.37:1) treatments (Table 10). Similar finding were recorded by Devkota *et al.* (2016) who reported that, the benefit cost from the beekeeping was 1.81 which indicates higher yield and less cost of production of beekeeping contributed higher gross return and benefit cost ratio.

Sunflower hybrid seed production through hand pollination is very difficult task due to shortage of agricultural labour and it is also very expensive when there is availability of labour. To overcome this problem, we have to use honey bee colonies required for higher quantity and good quality sunflower hybrid seed production as an alternative to hand pollination. From the results of this study, three colonies of *Apis cerana* per acre was assessed to be effective for the production of good quality sunflower hybrid seeds.

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