

Pollination Potentiality of Honey Bee (*Apis cerana* Fab.) in Sunflower Hybrid (DRSH-1) Seed Production

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Received 7 April 2023, Accepted 4 July 2023, Published on 20 September 2023

ABSTRACT

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 (DRSH-1), among them 15 were hymenopterans, 7 lepidopterans, 5 coleopterans and 3 dipterans. *Apis cerana* was predominant pollinator (17.91±6.72 bees/5 capitula/ 5 min) on CMS parental line (ARM-243), followed by *A. dorsata* and least was *A. florea*. The peak foraging activity of *A. dorsata* was recorded at 0900-1000 hr with 24 bees, *A. cerana* recorded two peak foraging activity, one at 1100-1200 hr (29 bees)

and another at 1600-1700 hr (18 bees) and *A. florea* recorded at 1000-1100 hr (10 bees). The maximum Shannon-Wiener index of diversity (H=1.32) 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 1600-1700 hr. In case of fertility restorer line (6D-1) maximum Shannon-Wiener index was recorded at 1600-1700 hr (1.24). *A. cerana* under caged condition (three colonies per acre) without pollen load on CMS line were recorded highest mean number of nectar foragers (65±22.60 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 (42.25±23.59) with pollen load in *A. cerana* under caged condition (three colonies per acre) 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 and without pollen load spent more foraging duration (13.17 and 25.20 sec/capitulum respectively) on CMS line, followed by *A. cerana* and least in case of *A. dorsata* in open pollination. 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 hectare (839.53 kg), per cent filled seeds (95.80 %), test weight (5.41 g), volume weight (46.41 g/100 ml), kernel percentage (66.09 %), high-

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est kernel to husk ratio (1.94) and highest seed quality and 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, *Apis cerana*.

INTRODUCTION

Sunflower hybrid seed production through hand pollination is very difficult task due to shortage of agricultural labor and it is very expensive when there is availability of labor. 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 (Anon 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).

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. Both bee colony density and visitation rates increased all the productivity variables. However, effects were non-linear 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 laborers for transfer of pollen from male capitulum to cytoplasmic 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.

MATERIALS AND METHODS

The present investigation on "Pollination potentiality of honey bee (*Apis cerana* Fab.) in sunflower hybrid (DRSH-1) seed production" was carried out during 2020-21 at Main Agricultural Research Station, UAS, Raichur (Latitude 16°12'N and Longitude 77°20'E) which is located in the II zone of Karnataka state with following five treatments in six replications, T₁: Crop caged with three colonies of *Apis cerana* (six frame strength) per acre, T₂: Hand pollination, T₃: Open pollination, T₄: Hand + open pollination, T₅: Control (crop covered with mosquito nylon net). The parental lines ARM 243 (3 lines) and 6D-1 (1 line) of sunflower hybrid DRSH-1 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 five 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 (Belavadi and Ganeshiah 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 :

$$H = -\sum p_i \times \ln p_i$$

Where, p_i is the proportion of the i^{th} 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

$$d = n_i / NT$$

Where, n_i is the number of individuals of the i^{th} 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 600 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 liter (μl)/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 vigor Index : Ten seedlings from each replication from all treatments were randomly taken to record root and shoot length. Seed vigor 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 (DRSH-1) produced from different treatments were analyzed 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% 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 (DRSH-1), of which 15 species were from five families of Hymenoptera (Apidae, Vespidae, Halictidae, Megachilidae and Sphecidae), seven species from four families of Lepidoptera (Nymphalidae, Erebididae, Pieridae and Spingidae), five species from three families of Coleoptera (Chrysomelidae, Coccinellidae and Scarabaeidae) and one each species from three families of Diptera (Syrphidae, Sarcophagidae and Muscidae) (Table 1). Similarly, at Pantanagar recorded 12 species of insect visitors, of which nine belonged to Hymenoptera, one to Diptera, one to

Table 1. List of floral visitors of parental lines of sunflower hybrid (DRSH-1).

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

Table 2. Abundance, diversity and dominance of floral visitors on CMS parental line (ARM-243) of sunflower hybrid (DRSH-1) under open condition.

Time (hrs)	Apis species			Number of floral visitors/5 capitula/5 min.				Total	“H” value	“D” value
	<i>Apis cerana</i>	<i>Apis dorsata</i>	<i>Apis florea</i>	Hymenoptera	Diptera	Lepidoptera	Coleoptera			
0600-0700	9	2	2	0	0	0	1	14	1.03	0.64
0700-0800	18	10	3	2	0	0	1	34	1.18	0.52
0800-0900	20	18	4	2	1	1	1	47	1.32	0.42
0900-1000	24	24	6	3	0	0	0	57	1.12	0.42
1000-1100	27	23	10	4	0	1	0	65	1.26	0.41
1100-1200	29	20	7	4	1	0	0	61	1.21	0.47
1200-1300	21	17	6	3	0	0	0	47	1.17	0.44
1300-1400	12	17	5	2	0	1	0	37	1.25	0.45
1400-1500	13	12	3	3	0	0	0	31	1.18	0.41
1500-1600	16	10	2	4	1	0	0	33	1.24	0.48
1600-1700	18	6	1	1	0	0	0	26	0.84	0.69
1700-1800	8	2	0	1	0	1	0	12	0.98	0.66
Total	215	161	49	29	3	4	3	464		
Mean±SD	17.91±6.72	13.41±7.59	4.08±2.84	2.41±1.31	0.25±0.45	0.33±0.49	0.25±0.45			
“d” value	0.463	0.346	0.105	0.062	0.006	0.008	0.006			

Lepidoptera and one was a Coleopteran species (Goswami *et al.* 2013).

Abundance of floral visitors on parental lines DRSH-1 : On ARM-243 line, *A. cerana* was predominant pollinator (17.91 ±6.72 bees/ 5 capitula/ 5 min), followed by the *A. dorsata* and least was *A.*

florea. The peak foraging activity of *A. dorsata* was recorded at 0900-1000 hr with 24 bees/ 5 capitula/ 5 min, *A. cerana* recorded two peak foraging activity pattern, one at 1100-1200 hr and another at 1600-1700 hr and *A. florea* recorded peak foraging activity at 1000-1100 hr (Table 2). Among non-*Apis* species, hymenopterans were the predominant (2.41±1.31

Table 3. Abundance, diversity and dominance of floral visitors on fertility restorer parental line (6D-1) of sunflower hybrid (DRSH-1) under open condition.

Time (hrs)	Apis species			Number of floral visitors/5 capitulum/5 min				Total	“H” value	“D” value
	<i>Apis cerana</i>	<i>Apis dorsata</i>	<i>Apis florea</i>	Hymenoptera	Diptera	Lepidoptera	Coleoptera			
0600-0700	21	26	8	1	0	0	1	57	1.14	0.45
0700-0800	33	38	10	2	0	1	1	85	1.17	0.44
0800-0900	38	42	16	1	1	0	1	99	1.17	0.42
0900-1000	44	54	18	3	0	0	1	120	1.14	0.45
1000-1100	46	58	20	4	0	1	1	130	1.20	0.44
1100-1200	47	60	19	1	1	0	0	128	1.08	0.46
1200-1300	40	57	18	1	1	0	0	117	1.09	0.48
1300-1400	22	48	14	1	0	0	0	85	1.02	0.56
1400-1500	22	31	8	1	0	0	0	62	1.05	0.50
1500-1600	23	26	10	1	0	1	0	61	1.16	0.42
1600-1700	25	22	10	1	1	1	0	60	1.24	0.41
1700-1800	20	10	4	0	0	1	0	35	1.03	0.57
Total	381	472	155	17	4	5	5	1039		
Mean ± SD	31.75±	39.33±	12.91±	1.41±	0.33±	0.41±	0.41±			
	10.70	16.43	5.24	1.08	0.49	0.51	0.51			
“d” value	0.366	0.454	0.149	0.016	0.003	0.004	0.004			

Table 4. Nectar foragers of *Apis* species on CMS parental line (ARM-243) of sunflower hybrid (DRSH-1) under caged (3 colonies / acre) pollination as compared with open condition. NB:*Significant at $p<0.05$, NS- Non significant at $p<0.05$.

Time (hrs)	Number of floral visitors/5 capitula/5 min							
	Foragers of <i>A.cerana</i> under caged condition (3 colonies/acre)		Foragers of <i>Apis</i> species from natural colonies					
	Bees with pollen load	Bees without pollen load	<i>Apis cerana</i>		<i>Apis dorsata</i>		<i>Apis florea</i>	
		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	44	51	3	6	0	2	1	1
0700-0800	69	89	4	14	2	8	1	2
0800-0900	79	94	4	16	2	16	2	2
0900-1000	68	88	6	18	4	20	2	4
1000-1100	65	76	8	19	5	18	4	6
1100-1200	46	49	6	23	4	16	2	5
1200-1300	28	50	3	18	3	14	2	4
1300-1400	12	77	2	10	2	15	0	5
1400-1500	14	87	5	8	0	12	0	3
1500-1600	32	48	8	8	0	10	0	2
1600-1700	36	46	4	14	0	6	0	1
1700-1800	14	25	2	6	0	2	0	0
Total	507	780	55	160	22	139	14	35
Mean \pm SD	42.25 \pm 23.59	65 \pm 22.60	4.58 \pm 2.06	13.33 \pm 5.66	1.83 \pm 1.85	11.58 \pm 6.02	1.16 \pm 1.26	2.91 \pm 1.88
t-test @ 5%		*		*		*		*

bees/ 5 capitula/ 5 min) floral visitors and their peak foraging activity was at 1000-1200 hr, followed by lepidopterans, dipterans and coleopterans. In case of fertility restorer line (6D-1) *A. dorsata* was the predominant pollinator (39.33 \pm 16.43 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 60 bees/5 capitula/5 min, *A. cerana* recorded two peak foraging activity at 1100-1200 hr and at 1600-1700 hr and *A. florea* also

Table 5. Pollen and nectar foragers of *Apis* species on fertility restorer parental line (6D-1) under caged (3 colonies/acre) and open condition. NB:*Significant at $p<0.05$, NS-Non significant at $p<0.05$.

Time (hrs)	Number of bees/5 capitula/5 min							
	Foragers of <i>A. cerana</i> under caged condition (3 colonies/acre)		Foragers of <i>Apis</i> species from natural colonies					
	Pollen	Nectar	<i>Apis cerana</i>		<i>Apis dorsata</i>		<i>Apis florea</i>	
		Pollen	Nectar	Pollen	Nectar	Pollen	Nectar	
0600-0700	100	64	15	6	16	10	6	2
0700-0800	140	78	25	8	26	12	8	2
0800-0900	152	80	30	8	28	14	12	4
0900-1000	146	82	35	9	36	18	14	4
1000-1100	135	77	36	10	38	20	15	5
1100-1200	99	76	38	9	38	22	13	6
1200-1300	53	43	32	8	36	21	12	6
1300-1400	21	31	14	8	32	16	10	4
1400-1500	15	28	16	6	16	15	6	2
1500-1600	64	36	18	5	14	12	8	2
1600-1700	102	48	20	5	12	10	10	0
1700-1800	60	18	16	4	6	4	4	0
Total	1087	661	295	80	298	174	118	37
Mean \pm SD	90.58 \pm 47.82	55.08 \pm 23.58	24.58 \pm 9.13	7.16 \pm 1.89	24.83 \pm 11.48	14.50 \pm 5.28	9.83 \pm 3.48	3.08 \pm 2.06
t-test @ 5%		*		*		*		*

Table 6. Nectar foraging duration of *Apis* species on CMS parental line (ARM-243) under caged pollination as compared with open pollination. The values with same superscript in a column do not differ significantly by DMRT. NB : *Significant at $p < 0.05$.

Time (hrs)	Mean time (sec/capitulum) spent for nectar collection							
	Foragers of <i>A. cerana</i> under caged condition (3 colonies/acre)		Foragers of <i>Apis</i> species from natural colonies					
	Bees with pollen load	Bees without pollen load	<i>Apis cerana</i>		<i>Apis dorsata</i>		<i>Apis florea</i>	
			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	2.80 ^k	2.46 ^l	1.10 ^g	4.50 ^e	0	2.09 ^h	0	4.92 ⁱ
0700-0800	5.53 ⁱ	7.40 ^k	3.98 ^f	10.86 ^f	1.51 ^f	7.36 ^g	3.25 ^e	11.96 ^h
0800-0900	13.00 ^g	18.26 ^j	6.90 ^{de}	16.31 ^e	2.90 ^e	11.85 ^f	10.65 ^d	18.82 ^f
0900-1000	29.26 ^a	49.06 ^a	11.26 ^b	26.22 ^c	5.42 ^d	21.80 ^d	15.06 ^{bc}	30.04 ^d
1000-1100	29.34 ^a	43.73 ^b	13.84 ^a	37.83 ^a	7.84 ^c	31.84 ^a	18.24 ^a	39.46 ^a
1100-1200	27.33 ^b	42.06 ^c	12.34 ^{ab}	35.54 ^a	9.46 ^{ab}	30.42 ^{ab}	16.28 ^b	37.12 ^{ab}
1200-1300	23.80 ^c	40.60 ^d	11.86 ^b	34.56 ^a	10.22 ^a	28.25 ^b	13.57 ^c	35.30 ^b
1300-1400	18.20 ^d	38.93 ^e	10.06 ^c	30.84 ^b	8.76 ^b	25.24 ^c	0	32.63 ^c
1400-1500	15.33 ^e	37.13 ^f	9.06 ^c	29.44 ^{bc}	0	21.19 ^d	0	29.13 ^d
1500-1600	14.60 ^f	36.14 ^g	8.40 ^{cd}	22.86 ^d	0	18.06 ^e	0	22.16 ^e
1600-1700	9.80 ^h	34.20 ^h	6.73 ^{de}	19.06 ^e	0	10.04 ^f	0	15.49 ^g
1700-1800	4.80 ^j	26.54 ⁱ	6.04 ^e	11.76 ^f	0	7.53 ^g	0	0
Mean	16.14	31.37	8.48	23.31	6.58	17.97	13.17	25.20
SEm±	0.08	0.23	0.58	1.13	0.30	0.81	0.54	0.82
CD @ 5%	0.24	0.69	1.73	3.34	0.89	2.41	1.61	2.44
t-test @ 5%		*		*		*		*

recorded two peak foraging activity at 1000-1100 hr and 1500-1700 hr (Table 3). Among non-*Apis* species, hymenopterans were predominant (1.41±1.08 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 4.40 mm and R line was 6.61 mm. The variation in abundance,

Table 7. Foraging duration of *Apis* species on fertility restorer parental line (6D-1) 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.

Time (hrs)	Mean time (sec/capitulum) spent for pollen nectar collection							
	Foragers of <i>A. cerana</i> under caged condition (3 colonies/acre)		Foragers of <i>Apis</i> species from natural colonies					
	Pollen	Nectar	<i>Apis cerana</i>		<i>Apis dorsata</i>		<i>Apis florea</i>	
			Pollen	Nectar	Pollen	Nectar	Pollen	Nectar
0600-0700	23.93 ^g	3.40 ^l	2.19 ^g	3.20 ⁱ	2.01 ^f	1.27 ⁱ	2.15 ^h	3.82 ^g
0700-0800	26.40 ^e	8.40 ^k	5.80 ^f	8.65 ^h	3.72 ^{ef}	5.48 ^h	8.91 ^f	10.38 ^f
0800-0900	28.00 ^d	24.13 ^j	10.44 ^e	14.00 ^{fg}	5.80 ^{de}	10.17 ^g	13.83 ^d	16.86 ^e
0900-1000	29.13 ^c	50.94 ^a	15.91 ^{cd}	24.62 ^d	9.15 ^c	18.18 ^e	17.61 ^{bc}	26.12 ^d
1000-1100	31.73 ^b	50.00 ^b	21.57 ^a	36.14 ^a	10.74 ^c	31.77 ^a	20.66 ^a	38.10 ^a
1100-1200	34.60 ^a	46.87 ^c	20.57 ^a	34.33 ^{ab}	13.48 ^{ab}	27.81 ^{bc}	19.15 ^{ab}	37.10 ^{ab}
1200-1300	28.53 ^{cd}	45.40 ^d	16.85 ^b	32.86 ^b	15.10 ^a	28.06 ^b	18.71 ^{bc}	34.65 ^{bc}
1300-1400	26.40 ^e	41.86 ^c	14.86 ^{cd}	30.08 ^c	11.11 ^{bc}	23.16 ^c	17.15 ^c	32.87 ^c
1400-1500	25.06 ^f	40.67 ^f	13.75 ^d	25.72 ^d	11.20 ^{bc}	20.54 ^d	14.24 ^d	28.17 ^d
1500-1600	21.53 ^h	38.13 ^g	10.53 ^c	21.00 ^e	8.82 ^c	14.82 ^f	11.89 ^e	19.30 ^e
1600-1700	16.60 ⁱ	35.40 ^h	7.94 ^f	16.33 ^f	6.17 ^d	7.89 ^g	7.81 ^f	9.10 ^f
1700-1800	11.20 ^j	25.20 ⁱ	5.88 ^f	12.33 ^g	3.39 ^{ef}	5.44 ^h	5.34 ^g	0
Mean	25.26	34.20	12.19	21.60	8.39	16.21	13.12	23.31
Sem±	0.29	0.17	0.83	0.83	0.82	0.79	0.63	0.86
CD @ 5%	0.86	0.51	2.46	2.47	2.42	2.34	1.86	2.55
t-test @ 5%		NS		*		*		NS

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.

Diversity and dominance of floral visitors on parental lines of DRSH-1: The maximum Shannon-Wiener index of diversity (H) of floral visitors (H=1.32) under open condition was recorded at 0800-0900 hr of the day and least (H=0.84) was recorded at 1600-1700 hr. The highest (d=0.463) Berger Parker dominance index between the floral visitors were recorded in case of *A. cerana* and lowest was in case of coleopterans and dipterans (d=0.006), whereas between the hours of the day, highest dominance was recorded at 1600-1700 hr (0.69) and lowest was recorded at 1000-1100 and 1400-1500 hr (0.41) on CMS parental line (ARM-243) (Table 2). In case of fertility of restorer parental line (6D-1), maximum Shannon-Wiener index of diversity (H) of floral visitors (H=1.24) in open condition was recorded at 1600-1700 hr of the day and least (H=1.02) was recorded at 1300-1400 hr. The highest (d=0.454) Berger Parker dominance index between the floral visitors of 6D-1 was recorded with *A. dorsata* and lowest with dipterans (d=0.003), whereas highest dominance between the hours of the day was recorded at 1700-1800 hr (0.57) and lowest at 1600-1700 hr (0.41) (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 (ARM-243) : The highest mean number of nectar foragers (65±22.60 bees/5 capitula/5 min) of *A. cerana* under caged condition (three colonies per acre) without pollen load were recorded, followed by *A. cerana*, *A. dorsata* and *A. florea* foragers from natural colonies in open pollination condition, whereas, highest mean number of nectar foragers (42.25±23.59 bees/ 5 capitula/ 5 min) of *A. cerana* under caged condition (three colonies per acre) with pollen load were recorded, followed by *A. cerana* (4.58±2.06 bees/ 5 capitula/ 5 min), *A. dorsata* (1.83±1.85 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 (79 and 94 bees/ 5 capitula /5 min. respectively) and 1600-1700 hr and 1400-1500 hr (36 and 87 bees/5 capitula/5 min respectively) of the day. But there was 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 1000-1100 hr and 1500-1600 hr (8 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 1000-1100 hr and *A. dorsata* and *A. florea* without pollen load recorded peak foraging activity at 0900-1100 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 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. 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 (6D-1) : 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 (90.58±47.82 bees/5 capitula/5 min) compared with nectar foragers (55.08±23.58 bees/5 capitula/5 min.) under caged condition (three colonies per acre), whereas in open condition *A. dorsata* was the more abundant pollen and nectar forager compared with other *Apis* species (Table 5). *A. cerana* recorded two peaks of pollen foraging activity in caged at 0800-0900 hr and 1600-1700 hr (152 bees and 102 bees / 5 capitula/ 5 min respectively) and open condition at 1100-1200 hr and 1600-1700 hr (38 bees and 20 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

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.

Time (hrs)	Sunflower hybrid (DRSH-1)			
	Cytoplasmic male sterile line (ARM-243)		Restorer line (6D-1)	
	Nectar ($\mu\text{l}/\text{florete}$)	TSS (%)	Nectar ($\mu\text{l}/\text{florete}$)	TSS (%)
0600	1.00 ^g (0.00)	1.00 ^g (0.00)	1.00 ^g (0.00)	1.00 ^f (0.00)
0800	1.08 ^f (0.16)	5.40 ^f (28.25)	1.01 ^f (0.01)	5.28 ^e (26.85)
1000	1.16 ^e (0.35)	6.14 ^e (36.74)	1.08 ^e (0.16)	5.93 ^d (34.19)
1200	1.45 ^a (1.11)	6.57 ^d (42.21)	1.22 ^a (0.49)	6.39 ^c (39.86)
1400	1.39 ^b (0.94)	7.19 ^b (50.82)	1.17 ^b (0.38)	7.00 ^b (48.00)
1600	1.25 ^d (0.55)	7.42 ^a (54.14)	1.10 ^d (0.21)	7.32 ^a (51.67)
1800	1.35 ^c (0.83)	7.05 ^c (48.71)	1.14 ^c (0.31)	7.04 ^b (48.59)
Mean	1.24	5.83	1.10	5.69
SEm \pm	0.002	0.015	0.002	0.014
CD @ 5%	0.005	0.046	0.008	0.044

(82 bees and 48 bees/5 capitula /5 min respectively) and in open condition only one peak in nectar foraging activity was recorded at 1000-1100 hr with 10 bees/5 capitula/5 min *A. dorsata* and *A. florea* recorded only one peak in pollen and nectar foraging activity between 1000-1200 hr of the day. There were significant differences in the number of nectar and pollen foragers of *Apis* species.

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, thereby resulting in decreased bee activity.

Foraging duration of *Apis* species on CMS parental line (ARM-243) : The mean nectar foraging duration with pollen load of *A. florea* spent more foraging

duration (13.17 sec/capitulum) 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, nectar foraging duration (16.14 sec/capitulum) was recorded by *A. cerana* in caged pollination (3 colonies per acre) (Table 6). *A. florea* without pollen load spent more mean nectar foraging duration (25.20 sec/capitulum) in open pollination compared with *A. cerana* (23.31 sec/capitulum) and *A. dorsata* (17.97 sec/capitulum). But in caged condition (3 colonies of *A. cerana*/ acre) maximum nectar foraging duration (31.37 sec/capitulum) was recorded. 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 (6D-1) : The maximum mean pollen foraging duration (13.12 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 between 1000-1300 hr of the day (Table 7). The duration for nectar collection was also recorded in same trend with *A. florea* (23.31 sec/capitulum), *A. cerana* (21.60 sec/capitulum) and *A. dorsata* (16.21 sec/capitulum). The pollen (25.26 sec/capitulum) and nectar (34.20 sec/capitulum) foraging duration by the *A. cerana* in caged pollination (3 colonies/ acre) registered highest mean foraging duration as

Table 9. Effect of different modes of pollination on hybrid seed yield and yield attributing characters of sunflower hybrid (DRSH-1). The values with same superscript in a column do not differ significantly by DMRT.

Treatments	Seed yield (kg/ha)	Per cent filled seeds	Test weight (g)	Volume weight (g)	Kernel (%)	Husk (%)	K:H ratio
T ₁ : <i>A. cerana</i> 3 colonies/acre	786.28 ^b	93.16 ^b	5.25 ^b	44.26 ^b	64.55 ^b	35.45 ^d	1.81 ^b
T ₂ : Hand pollination	659.86 ^c	91.60 ^c	4.55 ^c	42.01 ^c	62.82 ^c	37.18 ^c	1.68 ^c
T ₃ : Open pollination	421.21 ^d	79.40 ^d	2.93 ^d	33.56 ^d	57.89 ^d	43.41 ^b	1.33 ^d
T ₄ : Hand +Open pollination	839.53 ^a	95.80 ^a	5.41 ^a	46.41 ^a	66.09 ^a	33.91 ^e	1.94 ^a
T ₅ : Control	117.86 ^e	0.00 ^e	0.91 ^e	8.85 ^e	0.00 ^e	100 ^a	0.00 ^e
Mean	564.94	71.99	3.81	35.01	50.27	49.99	1.35
SEm±	4.94	0.23	0.04	0.25	0.37	0.47	0.02
CD @ 5%	14.96	0.70	0.12	0.76	1.12	1.43	0.07

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 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)

Table 10. Effect of different modes of pollination on seed quality, associated parameters and B:C ratio of sunflower hybrid (DRSH-1). The values with same superscript in a column do not differ significantly by DMRT.

Treatments	Germination (%)	Shoot length (cm)	Root length (cm)	Seed vigour index	Oil content (%)	B:C ratio
T ₁ : <i>A. cerana</i> 3 colonies/acre	93.16 ^b	11.31 ^a	18.63 ^b	1072.54 ^b	36.43 ^b	1.39:1
T ₂ : Hand pollination	91.60 ^c	10.39 ^b	17.02 ^c	969.37 ^c	35.66 ^c	1.09:1
T ₃ : Open pollination	79.40 ^d	9.44 ^c	15.45 ^d	768.70 ^d	25.94 ^d	1.02:1
T ₄ : Hand +Open pollination	95.80 ^a	11.33 ^a	19.42 ^a	1104.53 ^a	36.84 ^a	1.40:1
T ₅ : Control	0.00 ^e	0.00 ^d	0.00 ^e	0.00 ^e	10.00 ^e	0.28:1
Mean	71.99	8.49	14.10	783.02	28.97	1.03
SEm±	0.23	0.08	0.08	8.81	0.09	
CD @ 5%	0.70	0.26	0.26	26.63	0.27	

content in the nectar of parental lines : Nectar secretion/production by both the parental lines of sunflower hybrid was recorded. 1.24 and 1.10 µl/floret was produced by ARM-243 and 6D-1 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 ARM-243 and 6 D-1 lines was recorded 5.83 and 5.69 % 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 (839.53 kg), per cent filled seeds (95.80%), test weight (5.41 g), volume weight (46.41 g/100 ml), kernel percentage (66.09 %) and highest kernel to husk ratio (1.94) was recorded with hand+open pollination treatment, followed by *A. cerana* @ 3 colonies per acre (786.28 kg, 93.16%, 5.25 g, 44.26g/100 ml, 64.55% and 1.81 K:H ratio, respectively), hand pollination, open pollination and least was recorded with control (Table 9). The highest germination percentage (95.80 %), shoot length (11.33 cm), root length (19.42 cm), seed vigour index (1104.53) and oil content (36.84 %) was recorded in hand + open pollination plots, followed by the *A. cerana* @ 3 colonies per acre treated plots (93.16 %, 11.31cm, 18.63 cm, 1072.54 and 36.43 %), hand pollination, open and pollination and least seed quality and its associated parameters were recorded in control treatment (Table 10). The statistically significant highest oil percentage content (36.84 %) was recorded in open+ hand pollination, followed by *A. cerana* @ 3 colonies per acre treated plot (36.43 %), hand pollination (35.66 %), open pollination (25.94 %) and least was recorded with control (10.00%). 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* himalaya) pollination. Crop pollinated by honey bees enhanced seed set (23.43 %) compared to pollination without insects and 18.31% compared to open pollination. The increase in seed yield in bee pollinated treatment was 80.49% greater than pollinated without insects and 11.75% than open pollination. The germination of seeds from bee pollinated plants was 30.41% higher than seeds from plants without insects and 5.35% 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 hand + open pollination treatment (1.40:1), followed by *A. cerana* @ 3 colonies per acre

(1.39:1), hand pollination (1.09:1), open pollination (1.02:1) and control (0.28:1) 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.

CONCLUSION

Sunflower hybrid seed production through hand pollination is very difficult task due to shortage of agricultural labor and it is also very expensive when there is availability of labor. 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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