

Role of ways of insect visitors foraging for pollination in yield contributing traits of mustard

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Abstract

The experiment was conducted at the research farm of Sher-e-Bangla Agricultural University, Dhaka-1207 during the period from November 2016 to February 2017. The experiment consisted of three different ways of insect visitors foraging for pollination of mustard flowers *viz.*, T₁= Open field (Control), *i.e.*, the mustard field was fully open for free movement by the insect pollinators, T₂= Netting with honey bee, *i.e.*, the mustard plots was caged with muslin net and the bee hive was placed inside the cage and T₃= Netting without honey bee, *i.e.*, the mustard plots was caged with muslin net but the bee hive was not placed inside the cage. BARI Sharisha-8 [*Brassica juncea* (L.) Czernajew] was used as planting material. Randomized Complete Block Design was selected to lay

out the present experiment with 7 replicates. Study showed that honey bee was the most abundant hymenopterans in the mustard field as a pollinator. The yield and yield contributing traits were significantly influenced by different ways of insect visitors foraging for pollination. The maximum (3.50 g) 1000-seed weight was recorded from treatment T₂ followed by T₃ and the lowest 1000-seed weight (2.68 g) was recorded from T₃. The highest seed yield (2.45 t ha⁻¹) was exhibited from treatment T₂ followed by T₁ whereas the lowest seed yield (1.67 t ha⁻¹) was recorded from T₃. Finally, it can be concluded that providing honey bee colonies to the flowering mustard field can substantially contribute to the yield.

Introduction

Mustard; *Brassica juncea* (L.) Czernajew, tori sharisha and *Brassica napus* (L.), rye sharisha are the most cultivated species in Bangladesh. They ranked second most important edible oilseed crops of the world after soybean and the oldest cultivated amphidiploids¹ which are a dicotyledonous angiosperm plant belongs to Brassicaceae or Cruciferaceae family.^{2,3} Edible oil plays a very important role in human nutrition. As a high-energy component of food, edible oils are important for meeting the calorie requirements. Fats and oils are also the sources of essential fatty acids. The main essential fatty acids of vegetable oils are linoleic and linolenic acids. Fats and oils are used to synthesize phospholipid, which are important components of active tissues *viz.*, brain, nerve, and liver of human beings and other animals. These crops are grown under a wide range of agro-climatic conditions. Indian mustard is the most important member of this group, accounting for more than 70% of the area under rapeseed-mustard, followed by toria, yellow sarson and brown sarson.^{2,3} The oil content varies from 37 to 49% in *Brassica juncea* (Indian mustard).⁴ Rapeseed-Mustard is a principal oilseed crop, which plays a significant role in the national economy of Bangladesh. But seed yield/ha is very low compared to other mustard (tori/rye) growing countries of the world because the productivity of this crop is very low. The area for mustard production in Bangladesh has been on increase but the productivity has been diminishing hence the indigenous oil production of the state could not match the growing demand of population.⁵

The low average yield of mustard is due to cultivation of traditional varieties, non-availability of seeds of high yielding varieties and delayed sowing.⁵ Although such decline could be attributed to pests, diseases damage, poor soil fertility or water stress, there is evidence that insufficient pollination can also significantly minimize the crop yield.⁶ According to Sushil *et al.*,⁷ low seed yield due to inadequate pollination is often faced as a major problem of *Brassica* seed production. Inadequate pollination is caused by sev-

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Dedication: the authors' would like to dedicate their present piece of research to the farming communities of hunger world.

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eral factors and the most important of which include the lack of adequate number and diversity of pollinators. Thus, there is a need to ensure pollination by conserving the pollinators and attracting them towards the crop land. Pollination by insects is inevitable for *Brassica*, since they are generally incompatible (does not take part in fertilization process by pollen rain with ovule) in self pollination⁸ and the pollen is heavier and sticky, which is unable to be easily wind-borne. The blooming phase of any plant is the most crucial stage as the diversity of insects is higher than any other phase of plant species.^{9,10} Besides contributing to the preservation of natural ecosystems, bee pollination is one of the main alternatives for the improvement of crop productivity.^{11,12} Thus, honey bees, *Apis mellifera* L. are considered the most important flower visitors¹³ of most efficient *B. napus* L.^{6,14} However, the role of hymenopterans insect, especially honey bee on the productivity of mustard through cross pollination as an insect vectors is not well investigated to the researchers'. So, keeping the fact in view the present study was undertaken to study the effect of the managed bee on the yield and yield contributing characters of mustard.

Materials and Methods

Site description of research areas

Research farm of Sher-e-Bangla Agricultural University, Dhaka-1207 was used to conduct the present study during the period from November 2016 to February, 2017 which covered the full growing period of mustard in Bangladesh. The experimental field was located at 90° 33 E longitude and 23°71 N latitude at a height of 8 m above the sea level. The land of the research area was medium high in topography. The pH of the research plot was 5.57 and soil texture was silty clay in nature which contained 24% sand, 48% silt, and 28% clay. This value seemed the general observation of experimental area for normal growth of mustard plant but the analysis was not done based on the emphasis for seeing the variation of experimental plots. The physico-chemical properties of the soil are presented in Table 1. The research area belonged to the Madhupur Tract Agro-Ecological Zone (AEZ-28). Sub-tropical climate is characterized by the research areas with three distinct seasons, *kharij-1* (April-June), *kharij-2* (July-September) and *robi*/winter (October-March). This weather is favorable for crop production due to lower rainfall found during this period and it did not hampered the crop cycle. The average maximum temperature during the period of the experiment was 29.19°C and the average minimum temperature was 9.16°C.

Details of the meteorological data related to the temperature, relative humidity, and rainfalls during the period of the experiment were collected from the Bangladesh Meteorological Department, Dhaka and presented in Table 2.

Table 2. Monthly meteorological information during the period from November 2016 to February 2017.

Year	Month	Air temperature (°C)		Relative humidity (%)	Total rainfall (mm)
		Maximum	Minimum		
2016-2017	November	29.19	12.23	57.28	49
	December	26.00	9.16	70.43	1
	January	24.92	9.87	72.79	Trace
	February	25.85	14.54	78.42	Trace

Source: Metrological Centre (Climate Division), Agargaon, Dhaka.

Experimental details

Crop

BARI Sharisha-8 (*Brassica juncea* L.) was used as planting material. It is herbaceous growing to a height of 0.9 to 1.10 meters with tap root system and it has the potentiality to give 2.0-2.5 tons seed per hectare at well-managed condition.¹⁵

Experimental design and treatments

Randomized Complete Block Design (RCBD) was selected to lay out the present experiment. The unit plot size was 2.5 m × 2.0 m. Seven replicates were taken having 1.0 m space between the replicates and 0.75 m between the plots. Each plot contained two rows having 40 cm distance between the row and that between plants was 10 cm. The experiment consisted on three treatments such as, T₁= Open field (Control), *i.e.*, the mustard field was fully open for free movement by the insect pollinators, T₂= Netting with honey bee, *i.e.*, the mustard plots was caged with muslin net and the bee hive was placed inside the cage and T₃= Netting without honey bee, *i.e.*, the mustard plots was caged with muslin net but the bee hive was not placed inside the cage. Each treatment was randomly placed in each replicate. Two frame nucleus boxes having 200 individuals were used as rearing for honey bees and sugar candy binding with a stick was placed in cage as supplemental food for the honey bee (Figure 1).

Crop husbandry

A power tiller was used to open the plots selected for the research in the last week of October 2016 and exposed to the sun for a week, after which the land was harrowed, ploughed and cross-ploughed four times followed by laddering to obtain a good tilt condition of soil with the removal of weeds and stubbles. After collecting the seeds were treated with Provex 200 WP @ 2 g per

Table 1. The physicochemical properties of the soil of research plot (0-15 cm depth).

Soil parameters	Observed values
pH	5.57
Total N (%)	0.0679
Phosphorus	21.25 µg/g soil
Sulphur	24.29 µg/g soil
Boron	0.52 µg/g soil
Copper	3.38 µg/g soil
Zinc	2.98 µg/g soil

Source: Soil Resources Development Institute (SRDI), Khamarbari, Dhaka.

kg seed to protect seedlings against foot and root rot diseases and from later *Alternaria* blight. During land preparation, 8 t/ha decomposed cow dung was mixed. Urea, TSP, MoP, Gypsum, Zinc sulphate and Boric acid were applied as the source of Nitrogen (N), Phosphorus (P_2O_5), Potassium (K_2O), S, Zn and Boron (B) at the rate of 250, 180, 100, 150, 7 and 10 kg per hectare. At November 7, 2016, seeds are sown continuously in 2-2.5 cm deep furrows made by hand iron tine maintaining desired row spacing. Before seed placement in rows light water was applied in furrows for the easy emergence of seeds. After placement of seeds were covered with soil by hand. Within ten days of sowing the germination was satisfactory. Operations like thinning, weeding, watering and drainage and pest Management were done as and when necessary for proper growth and development of the crop.

Parameters taken

Overall parameters studied

At physiological maturity stage, the crop was harvested. Data on, plant height (cm), number of primary branches plant⁻¹, pod formation at 45, 55 and 65 DAS (Days after sowing), number of siliqua plant⁻¹, number of seeds siliqua⁻¹, 1000-seed weight (g), seed yield (t ha⁻¹).

Finally, overall increases of seed yield over control were seen, and the correlation coefficient was calculated by using Excel spreadsheet among the number of siliqua plant⁻¹, number of seeds siliqua⁻¹ and seed yield were calculated to show the importance of honey bee pollination of mustard flowers.

Counting the visiting insects order

Hymenopterans pollinator and other foraging insects were counted on this cultivar by seeing up on the net and open plots at morning and afternoon throughout the blooming period and counting the number of hymenopterans pollinator and other foraging insects actively collecting nectar or pollen. The visiting insects were identified on the basis of internet browsing and also by using the knowledge available in research expertise of Bangladesh localities under mustard research.

Flower visitors, along with the number of flowers visited and the total number of flowers per patch were recorded. This allowed calculation of abundances and visitation rates for different species groups, calculated as the proportion of flowers visited per species in a 5 min period during both morning and afternoon (where that species was present).

Statistics

After collection of all the data, those were analyzed following the ANOVA techniques by using the Web Agri Stat Package-WASP (Version-1) computer package program. The means were separated by using Least Significant Difference (LSD) at 5% level of probability. Graphs and figures were created by using MS Excel spreadsheet. Correlation line was created by using MS Excel spreadsheet, also.

Results and Discussion

Diversity of visiting insect

Different insects visited experimental plots during experimentation period (Figure 2A). Among different insects, the hymenopterans dominated the mustard field followed by lepidopterans, coleopterans, dipterans insect and lowest insect visitors were homopterans (1%). Mahfouz *et al.*¹⁶ found the similar pattern of insects foraging for pollination of sesame and they found that, hymenopterans were the most forgeable insects order in rapeseed plant. Stanley *et al.*¹⁷ stated that the pollinators can differ in terms of their efficiency of crop pollination. They also found that the honeybee, *Eristalis* hoverflies and bumblebees were the most useful pollinators of winter oilseed rape based on the amount of pollen carried, visitation rates and their abundance. Rader *et al.*¹⁸ reported that honey bees are not the only insects that pollinate crops. Apart from a few managed bee taxa, the great majority of other pollinators are free-living or wild, providing an ecosystem service to crops. Wild pollinators other than honey bees recently have been recognized for their role in increasing and stabilizing crop-pollination services. Increasingly, studies indicated that insect pollinators other than bees, such as flies, beetles, moths, and butterflies, are equally if not more important for the production of some crops.¹⁸

Plant height

The plant height of mustard was not significantly ($p > 0.05$, df: 2, sd: 0.87) differed due to different pollination condition (Figure 2B). Numerically the tallest plant (96.80 cm) was recorded from netting with honey bee (T_2) and the shortest plant (94.85 cm) was recorded from netting without honey bee (T_3). The honey bee has no any effects on plant height due to it is a varietal trait. So, under present study, no significant differences among pollination modes were found.



Figure 1. Experimental overviews.

Number of primary branches plant⁻¹ (no.)

Here we have simply noticed the differences found under different plots due to their normal growth and varietal potentiality. The number of primary branches plant⁻¹ of mustard varied under different pollination condition (Figure 3A). The results of the experiment revealed that the maximum number of primary branches plant⁻¹ was recorded when the plots netting with honey bee (T₂) followed by T₁ and the minimum number of primary branches plant⁻¹ was recorded from when the plots netting without honey bee (T₃).

Number of siliquae setting plant⁻¹ (no.)

The number of siliquae setting plant⁻¹ of mustard was significant due to different pollination condition at 45, 55 and 65 DAS (Table 3). The maximum number of siliquae setting plant⁻¹ (124.30, 133.30, 137.00 at 45, 55 and 65 DAS, respectively) were recorded from treatment T₂ followed by T₁ at 45, 55 and 65 DAS and the minimum number of siliquae setting plant⁻¹ (92.25, 98.25 and 100.83 at 45, 55 and 65 DAS, respectively) was produced by T₃ treatment. Kumar *et al.*¹⁹ found that the seed set was highest (75.5%) with hand + insect pollination than open pollination and

Table 3. Effect of different pollination condition on the number of siliquae setting plant⁻¹ of mustard at different days after sowing.

Treatments	No. of siliquae setting plant ⁻¹ at different days after sowing (DAS)		
	45	55	65
T ₁	108.5 b	115.57 b	119.0 b
T ₂	124.3 a	133.3 a	137.0 a
T ₃	92.25 c	98.25 c	100.83 c
LSD (0.05)	14.32	16.33	17.41

T₁=Open field (Control), T₂=Netting with honey bee, T₃=Netting without honey bee.

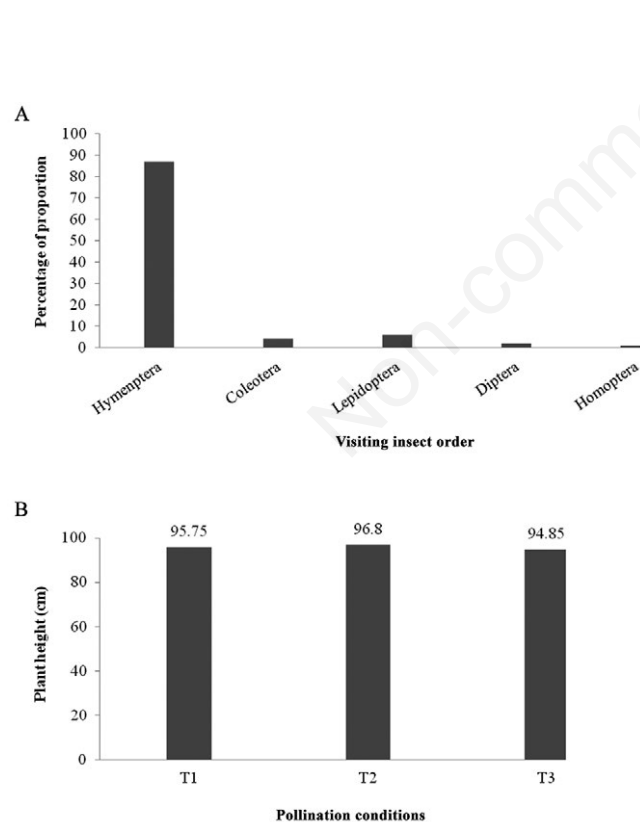


Figure 2. (A) Percentage of the major insect orders visited in mustard field during the flowering period; (B) Effect of different pollination condition on the plant height of mustard (LSD(0.05)=NS). T₁=Open field (Control), T₂=Netting with honey bee and T₃=Netting without honey bee.

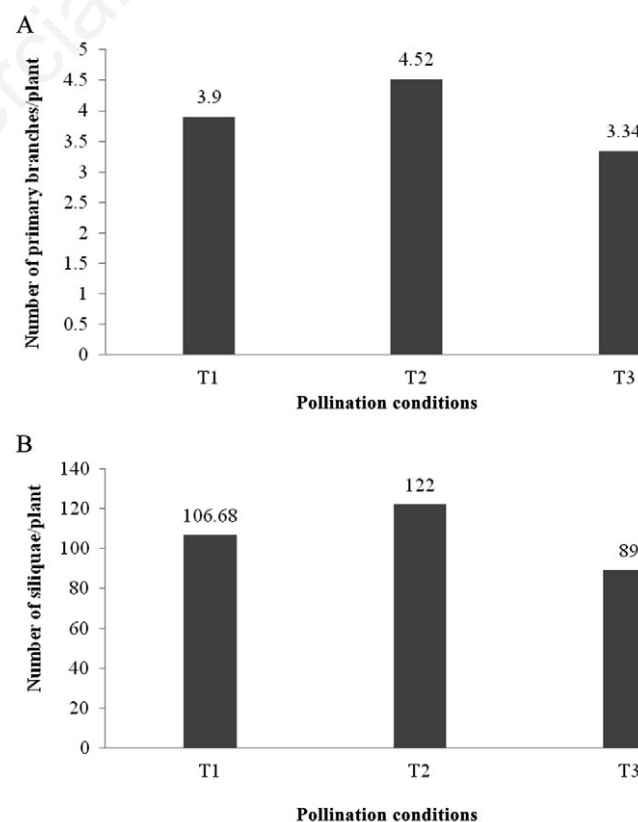


Figure 3. (A) Effect of different pollination condition on the number of primary branches plant⁻¹ of mustard (LSD (0.05)=0.54); (B) Effect of different pollination condition on the number of siliquae plant⁻¹ of mustard (LSD (0.05)=15.15). T₁=Open field (Control), T₂=Netting with honey bee and T₃=Netting without honey bee.

the crop netted without bees in sunflowers. Varma *et al.*²⁰ also compared the honey bee pollination with other insects' pollination on mustard bloom and they found that the honey bee pollination increased the pod set by 74.3 per cent.

Number of siliquae plant⁻¹ (no.)

The number of siliquae plant⁻¹ of mustard varied significantly due to different pollination condition (Figure 3B). The result revealed that the highest number of siliquae plant⁻¹ (122.00) was found from treatment T₂ followed by T₁ and the lowest number of siliquae plant⁻¹ (89.00) was found from T₃. Atmowidi *et al.*²¹ reported that the number of pods increased by three times in insect pollinated plots of mustard (*B. rapa*). Kumar *et al.*¹⁹ also found that the number of filled seeds per capitulum (728.2) was highest with hand + insect pollination than open pollination and the crop netted without bees in sunflowers.

Number of seeds siliqua⁻¹

The number of seeds siliqua⁻¹ of mustard significantly varied due to different pollination condition (Figure 4A). The result revealed that the highest number of seeds siliqua⁻¹ (28.50) was recorded from treatment T₂ and the lowest number of seeds siliqua⁻¹ (15.75) was recorded from T₃. The present findings were in agreement with Kumar *et al.*¹⁹ who observed that *B. juncea* plants caged with bees produced 25 percent more seed than plants caged without bees. Munawar *et al.*²² found that the number of filled seeds per capitulum was highest with hand + insect pollination than open pollination and the crop netted without bees in sunflowers Kumar

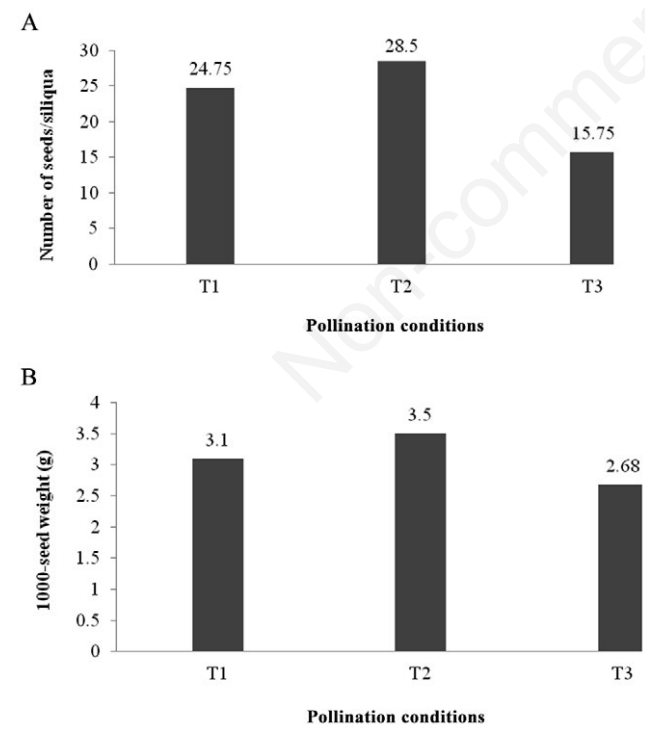


Figure 4. (A) Effect of different pollination condition on the number of seeds siliqua⁻¹ of mustard (LSD (0.05)=2.31). T₁=Open field (Control), T₂=Netting with honey bee and T₃=Netting without honey bee; (B) Effect of different pollination condition on the 1000-seed weight of mustard (LSD (0.05)=0.35). T₁=Open field (Control), T₂=Netting with honey bee and T₃=Netting without honey bee.

*et al.*¹⁹ reported that number of seeds per pod increased 98.2 % in insect pollinated plots of mustard (*B. rapa*).

1000-seed weight (g)

Different pollination condition significantly affected the 1000-seed weight of mustard (Figure 4B). The highest 1000-seed weight (3.50 g) was recorded from treatment T₂ followed by T₁ and the lowest 1000-seed weight (2.68 g) was recorded from T₃. The present result is in partial agreement with Atmowidi *et al.*²¹ who reported that the test weight of 1000-seed of canola, *Brassica napus* was with highest seed weight plants caged with honeybees (26.00g). The lowest was in plants caged without honeybees (9.30g). Latif *et al.*²³ reported that seed produced in cages with honey bees was heavier than the seeds from open plots of *B. juncea*. Singh *et al.*²⁴ reported that bee pollinated plots produced three times heavier seeds than self-pollinated plants in *B. campestris* var. toria.

Seed yield (t ha⁻¹)

Seed yield of mustard significantly influenced by different pollination condition (Figure 5). The highest seed yield (2.45 t ha⁻¹) was recorded from treatment T₂ followed by T₁ and the lowest seed yield (1.67 t ha⁻¹) was recorded from T₃. Kumar *et al.*¹⁹ who also recorded the higher seed yield of sunflower (849 kg/ha) in the intercropping system of sunflower + niger and lowest was in a sole crop of sunflower (747 kg/ha) indicating the role of pollinators in both cross-pollinated crops. The main reason for the higher seed yield per hectare may be the higher weight of seed per plant and higher 1000-seed weight may also be the reasons for highest seed yield. Singh *et al.*²⁴ emphasized upon the view that insect pollination led to the formation of well-shaped larger grains and more viable seeds than self-pollinated plants. Sanas *et al.*²⁵ who also reported that increased production of sunflower seed in an area with the introduction of honey bees related to areas without bees. Netting with honeybee (T₂) produced (6.52%) higher seed yield over control (T₁) and T₃ exhibited the negative response (-27.39%) on seed yield increase over control. Singh *et al.*²⁶ reported that, the honey bee introduced plots of cauliflowers gave the increasing percent of seed yield than that of open pollinated plots.

Correlation coefficient (r)

Data from harvested plant samples were used to calculate the correlation coefficient (r). Here we tried to show only the apparent normal relationship among these traits of mustard plant. Different

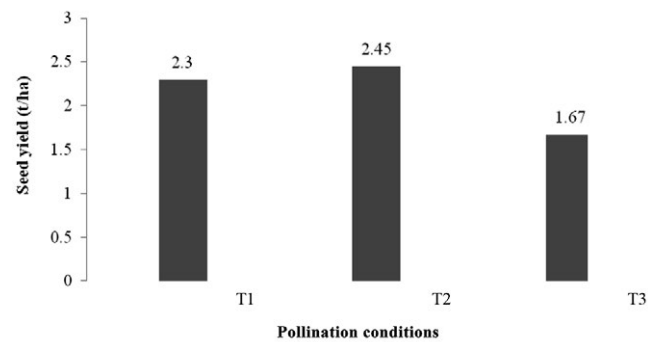


Figure 5. Effect of different pollination condition on the seed yield of mustard (LSD (0.05)=0.14). T₁=Open field (Control), T₂=Netting with honey bee and T₃=Netting without honey bee.

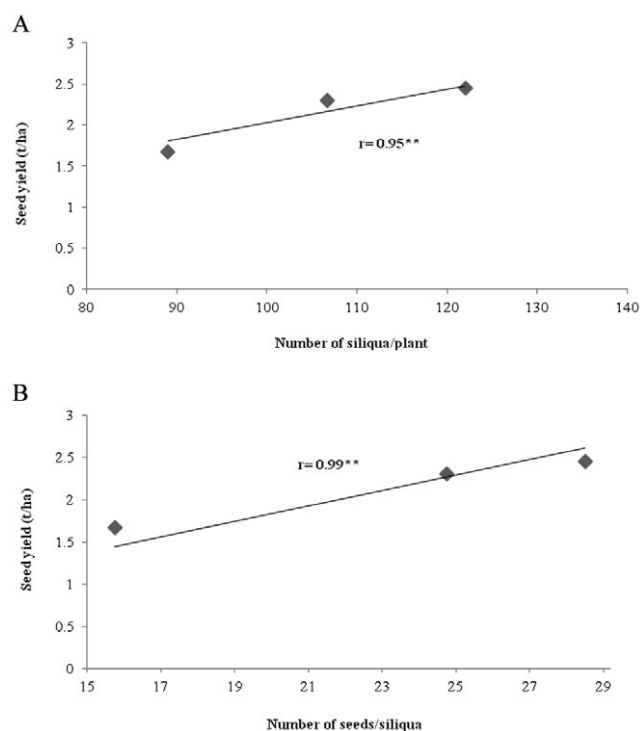


Figure 6. (A) The relationship between number of siliques plant⁻¹ and seed yield (t ha⁻¹) of mustard; (B) Relationship between number of seeds siliqua⁻¹ and seed yield (t ha⁻¹) of mustard.

pollination conditions influenced the different traits of mustard and so, the correlation coefficient was calculated among some yield and yield contributing traits. A linear relation ($r=0.95$) was exhibited between the number of siliques plant⁻¹ and seed yield (t ha⁻¹) of mustard (Figure 6A).

In Figure 6B, a linear relation ($r=0.99$) was found between number of seeds siliqua⁻¹ and seed yield (t ha⁻¹) of mustard. From the correlation study it may be said that, there was about 95 to 99% of seed yield of mustard was dependent on different yield contributing traits. Correlation among various characters indicated that all these characters had significant contribution to seed yield and yield would be increased by improving these yield attributes.

Conclusions

Based on the results of the present study the following conclusion could be drawn that, the honey bee is the most abundant hymenopterans in the mustard field as a pollinator. The growth, yield and yield contributing traits were significantly influenced by different strategies of pollination by the honey bee. Netting with honey bee was the best one for better yield of mustard.

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