

The Effect of Honeybee (*Apis mellifera*) Pollination in Enhancing Yield of *Nigella sativa* (Darbera Variety) in the High Land of Bale

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Abstract: Pollination is an essential process in the production of seed plants, it results in a genetically diverse production of offspring and its role is not well understood and appreciated in the local farming system. This study was conducted to evaluate the effect of honeybees pollination on Black cumin seed yield and yield related parameters at Sinana Agricultural Research center at on-station. The study had three experiment; these includes plots caged with honeybees (T1), plots caged without honeybees (T2) and open pollinated plots (T3). All collected data were analyzed using One-way-Analysis of Variance (ANOVA). *In the study seven species of insects were identified as Nigella sativa visitors or pollinators with the most frequent visitors were honeybees at 11:30 am time and minimum at 3:00 pm on open pollinated plot.* There were no significant different ($P>0.05$) on Date of blooming, Date of flowering, Flowering period, Primary and secondary branches among the three treatments. Whereas, there were a significant difference ($P<0.05$) on shading time, Number of capsule, Thousand kernel and total seed yield per hector. In general the present study showed that the highest seed yield/hector was achieved from crops caged with honeybees (20.20Qt/ha) and followed by open pollinated crop (17.54Qt/ha). The result also revealed that about 30.84% of seed yield advantage of *Nigella sativa* pollinated by honeybees over control/un pollinated by any insect. From this result it was concluded that visits of honeybees at flowering time of *Nigella sativa* have very helpful in boosting seed yield and yield related components of this crop.

Keywords: Honeybees, Pollination, *Nigella sativa*, Yield

1. Introduction

Black Cumin (*Nigella sativa* L.) is an annual herbaceous plant belonging to the family Ranunculacea [1] It is one of the seed spices grown in Ethiopia having a commercial demand both in the domestic as well as in the international market. It is one of crops chosen in the specialization program to be produced by selected potential areas.

Pollination is an essential process in the production of seed plants, and it results in a genetically diverse production of offspring [2]. Globally, one-third of the total human food supply depends on insect pollination [3-6]. Production of agriculture crops is increasing by 50% through bees pollination [3]. Honey bees (*Apis mellifera*) are economically essential insect pollinators all over the

world [7-12]. They provide ecologically for pollination of natural, wild vegetation plants and agricultural crops; as a result, they play a significant role in the landscape and natural resource preservation [7, 11, 13, 14]. Honey bee plays a major role in agriculture as pollinators, and their contribution to the global economy for food production is estimated between \$ 235 and \$ 285 billion annually [15] and \$0.815 billion in Ethiopia, which is 6.24% of the agricultural GDP [16]. According to Shrestha [17], the impact of honey bee pollination to crop production and quality has been estimated to be more than the value of honey and wax production. The economic benefit of the honey bees pollination service is 4.58 times higher than the

honey production in Ethiopia [16].

Honey bee pollination is as essential for crop production as water and fertilizer but, its role is not well understood and appreciated in the local farming system [18]. Exposing black cumin (*Nigella sativa* L.) to honey bee during flowering time increases the pod and seed productions [19]. Yield instability is a common problem in *Nigella sativa* and little attention has been paid for crop pollination in Ethiopia and the plant was never evaluated for pollination requirement under local conditions. The flowers of *Nigella sativa* are visited by honeybees [20]. Pollination studies on *N. sativa* are very limited. Despite its great importance; little attention has been paid to improve the production, so it remained as minor or underutilized crop. The honeybee (*Apis mellifera*) is of great economic importance in terms of increased yield and quality of commercially grown insect pollinated crops and also in assisting self pollinated crops in the world [21, 22]. So far, there is no detailed information regarding the pollinators or foragers of *Nigella sativa* is available in Ethiopia. Therefore, the aim of this study was to assess the role of honeybees in enhancing the yield and yield related components of *Nigella sativa* in the highland of Bale Zone of Oromia Regional state in Ethiopia.

2. Materials and Methods

The study was conducted at the high land of Bale in Sinana Agricultural Research Centre (SARC) at on-station during the 2017-2019 main cropping season for three consecutive years. It is found at a distance of about 463 km from Addis Ababa in the south-eastern direction, and 33 km from the nearby town, Robe. Geographic location is 07° 07' N latitude and 40° 13' E longitude. The elevation is 2400 meters above sea level. The area is characterized by bimodal rainfall pattern. The amount of rainfall distribution of the last thirty year (from 1990 to 2020), during crop growing seasons, was 905.13 mm. The monthly mean maximum and minimum air temperatures were 20.19°C and 9.58°C, respectively [23].

2.1. Experimental Set up

The experiments were arranged in a randomized complete block design (RCBD) with five replications. For the experiment Darbera variety of Black cumin (*Nigella sativa*) was used and all recommended agronomic practices were also

followed. The plots were kept from any damaging condition throughout the cropping season. The treatments were: plots caged with honeybees (T1)- the plots were covered with an insect proof mesh cage and a honeybee colony with ten frames were placed inside the cage during the flowering peak (50% florets open) time, plots caged pollinator exclusion (T2) -the plots were covered with an insect proof mesh cage before the ray florets started opening and plots kept open to all pollinators (T3) -plots accessible to all flower visitors or left open for natural pollination as control. Insect proof mesh cages (4m x 3m and 2.5m high) were made of wood covered with 20% shade cloth. All insects were removed from all the cages before blooming, to exclude unwanted pollinators. Honeybee colonies used in this experiment received supplementary feeding (dissolved sugar) and water before and after they were placed in the cages. At the time of maturity 10 mature pods were selected randomly from each replication and the number of seeds produced was counted manually. Harvesting was done from each plot after seeds are maturing. The seeds was separated manually from the pods and yield had calculated per plot for all the treatments.

2.2. Flower Visitors Identification

During the whole flowering period, flower visitor identifications were done in each of the plots accessible to all flower visitors, to assess which and how many insect species were visiting the *Nigella sativa* crop; and in the open plots accessible only to honeybees and other visitors to count the number of honeybee pollinators. The number of bees and other pollinators in the open treatment was observed in one m² area for five minutes seven days a week during the whole flowering period to identify other pollinators than honeybees and the data was recorded at 9.30am, 11.30am, 1.30 pm and 3.30pm hours a day. Visiting insects were collected and identified by the entomologist at Sinana Agricultural Research Center.

2.3. Yield Advantage Calculations

The yield advantage/an increase of pollinated crop by insect over control/un pollinated crop in yield and quality of *Nigella sativa* seeds due to managed honeybee pollination was calculated using the formula as follows.

$$\text{Yield increament(\%)} = \frac{(\text{Yield from honeybees pollinated} - \text{Yield from insect excluded})}{\text{Yield from from open pollinated}} \times 100$$

2.4. Data Collection and Measurement

Days to emergence: Days to emergence was determined when about 50% of the plants emerged from the soil after sowing.

Flowering Period: The flowering period was determined by recording the flower starting and ending date of the plants. Ten plants were selected to study the effects of mode of pollination on flowering period of the plant.

Blooming date (50% flowering): The number of days elapsed between date of sowing and date of 50% flowering

was computed and expressed as average number of days to flowering.

Number of primary branches per plant: Number of primary branches per stem was randomly counted from selected ten middle row plants at final harvest.

Number of Secondary branches per plant: Number of secondary branches per stem was randomly counted from selected ten middle row plants at final harvest.

Number of Capsule per plant: On individual plant basis, number of capsule in the tagged plants counted manually. The mean capsule per plant taken for each treatment.

Shading Time: It is days to maturity when about the plants reached 50% physiological maturity and its flowers was totally shades.

Thousand kernel weight (TKW) (g): It was determined based on the weight of 1000 seeds sampled from the grain yields of each plot by counting using an electric seed counter and weighed with an electronic balance.

Seed yield per hectare (kg): Grain yield was determined by harvesting plants from the net middle plot area to avoid border effects. Seeds, which were obtained from the corresponding net plot were cleaned manually and weighed using sensitive balance and recorded as mean values of seed yield per hectare in Quintals.

2.5. Data Analysis

All collected data were checked for normality and subjected to analysis of variance using statistical software package (SAS 9.1.3). The data were statistically analyzed using one-way-analysis of variance (ANOVA) and the differences among treatment means were compared using Least Significance Difference (LSD) test at 5% level of significance.

3. Results and Discussions

Pollination is an essential process in the production of *Nigella sativa* seeds, and it results in a genetically diverse production of offspring which results in high and quality seed production. Study was conducted on *Nigella sativa* for three years at Sinana Agricultural Research Center to identify its pollinators and accordingly the number of bees and other pollinators visiting the open experiments were observed and it was found that honeybees were the most frequent visitor with maximum activity at 11:30 am time and minimum activity at 3: 300 pm (Table 1). This is probably due to the bee's activity being limited by environmental factors like daily temperatures. Counts were made on one meter square (1 m²) for 5 minutes, when the flowers were open. The quantity of pollen transferred from anthers to stigmas, visit frequency to flower, pollinator forage pattern during anthesis and floral rewards availability are parameters that can adequately explain the pollination efficiency of floral visitors [24]. It is generally thought that the more visits made, the more efficient is the pollinator, though this also depends on the per visit pollen contribution to the pistil late flower part [24].

Table 1. Black cumin (*Nigella sativa*) visitors number and percent.

No	Insect order	C. Name	Scientific Name	Counted No.	Percentage
1	Hymenoptera	Honeybees	<i>Apis mellifera</i>	35	54.69
		Carpenter bee	<i>Xylocopa</i>	5	7.81
2	Orthoptera	Grass hopper	<i>Orthoptera</i>	6	9.38
		Wasp	<i>Colletes succinctus</i> L	7	10.94
3	Diptera	Dipterafly	<i>Diptera</i>	5	7.81
4	Lepidoptera	Butterfly	<i>Danaus plexippus</i> L	4	6.25
		Spider	<i>Achaearanea tridarium</i>	2	3.13
Total insect count				64	100

The mean data of date of flowering and date blooming (Table 2) showed no significant differences ($P < 0.005$) among the treatment. This might be due to date of flowering and date of blooming are not affected by mode of pollination, however it depends on environmental factors like daily temperate, relative humidity and also on soil type.

In the current study shading time of *Nigella sativa* had significantly affected by pollinators agents. The early shading time were observed in treatment caged with honeybees (114.07days) and followed by open pollination (116 days). This showed that shading time highly depends on pollinating agents.

The flowering period of *Nigella sativa* were significantly affected by mode of pollination. Plots caged without honeybees had the longest flowering period (32.67days), followed by open pollination, while caged with honeybees had the smallest flowering period (26 days). Similarly [25] reported flowering period had affect by mode of pollination and the longest flowering period was observed in Canola crops caged without bees followed by open pollinated crops. This indicated that mode of pollination had great contribution for early maturation of *Nigella sativa* crop.

Table 2. Mean comparison of three years data collected on Date of flowering date of blooming, Shading time and Flowering period.

Treatments (Mean±SE)	DF	DB	ST	FP
Caged with honeybees (T1)	88.07±1.86	111.60±2.44	114.07±1.17 ^b	26.00±2.04 ^b
Caged without honeybees (T2)	88.47±1.58	109.67±2.94	121.13±1.67 ^a	32.67±2.21 ^a
Open Pollination (T3)	87.60±1.94	110.73±2.72	116.00±1.61 ^b	28.40±2.38 ^{ab}
Over all mean	88.04	110.67	117.07	29.02
LSD	NS	NS	6.3155	4.2858
CV	7.53	9.47	4.97	29.53

abc=means with different superscripts within a column are significantly different ($P < 0.05$), NS=none Significant.

Notice: DF=Date of Flowering, DB=Date of Blooming, ST=Shading Time, FP=Flowering period.

The number of primary and secondary branches were not significantly different ($p > 0.05$) among treatments of *Nigella sativa*. This may probably because of the primary and

secondary branches were not affected by mode of pollination, but it is affected by environmental factors and soil type.

Significant different ($P < 0.05$) was observed in capsule

setting among treatments (Table 3). Plots caged with honeybees had the highest number of capsule setting per plant (18.8), while plots caged without honeybees had the lowest number of capsule setting per plant (13.87). Similarly,

in Sunflower crops caged with honeybees increased significantly the percentage of seed setting, number of filling seed per head compared with crops caged without honeybees [26].

Table 3. Mean comparison of three years data collected on Primary branch, secondary branch and Number of Capsule.

Treatments	PB	SB	NCP
Caged with honeybees (T1)	4.07±0.41	11.07±0.44	18.80±1.21 ^a
Caged without honeybees (T2)	4.33±0.33	9.80±0.58	13.87±1.07 ^b
Open Pollination (t3)	4.40±0.34	11.00±0.47	16.73±1.33 ^{ab}
Over all Mean	4.27±0.20	10.62±0.30	16.47±0.75
LSD	NS	NS	3.4434
CV (%)	32.72	18.22	28.37

abc=means with different superscripts within a column are significantly different (P<0.05), NS=none Significant.

Notice: PB=Primary branches, SB=secondary branches and NCP=Number of capsule.

The present result revealed that there was a significant different (P<0.001) among treatments regarding thousand seed weight (TKW). Plots caged with honeybees the highest TKW (0.00451kg), whereas plots caged without honeybees had the lowest TKW (0.00257). Munawaret *al.*, [26] found similar result from Pakistan on the Black cumin crop caged with honeybees.

Mode of pollination had significant effect on the yields per hector. From the current study, the total yield of plots under different treatments were compared and significant (P<0.001)

differences were found. The yield from all treatments were differ and the highest yield per hector was observed in treatments plots caged with honeybees (20.20 Qt/ha) and followed by open pollinated crop (17.54 Qt/ha). The lowest yield per hector was gained from caged without honeybees (14.79 Qt/ha) (Table 4). The higher yield of crops caged with honeybees might be because of the higher pollination efficiency of honeybees inside the cage. These results are in general agreement with previous result of [27] on *Guizotia abyssinica* at Tigris Region of Ethiopia.

Table 4. Mean comparison of three years data collected on Thousand Kernel weight and Total Seed Yield.

Treatments	TKW (Kg)	TSY (Qt/ha)
Caged with honeybees (T1)	0.0045±0.000373 ^a	20.20±0.42 ^a
Caged without honeybees (T2)	0.0026±0.000192 ^b	14.79±0.27 ^c
Open Pollination (T3)	0.0033±0.000268 ^b	17.54±0.28 ^b
Over all mean	0.0035±0.000202	17.51±0.38
LSD	0.0008	0.94
CV%	32.09	7.29

abc=means with different superscripts within a column are significantly different (P<0.001), NS=none Significant.

Notice: TKW=thousand Kernel weight, and TSY=Total seed yields.

In the present investigation, the result revealed that plots caged with honeybees had yield advantage of 30.84% in Black Cumin crop over control / caged without honeybees. The result in line with [28], who reported that insect pollination enhanced average crop yield between 18 and 71% depending on the crop. This might be because of black cumin pollination was highly affected by mode of insect pollination.

4. Conclusion

The importance of managing pollination to obtain higher yields still has been overlooked in Ethiopia. The present study revealed that honeybees and other pollinators had a significant effect on seed yield and yield related components of *Nigella sativa*. The highest seed yield/hector was achieved from crops caged with honeybees (20.20Qt/ha) and followed by open pollinated crop (17.54Qt/ha). The result revealed that plots caged with honeybees had yield advantage of 30.84% over control / caged without honeybees and it indicated honeybees were important pollinators of black cumin. Therefore, we recommended that keeping enough number of honeybees colony near the fields of *Nigella sativa* during flowering period

will enhance production and productivity of *Nigella sativa* and other similar cross pollinated spices crop.

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