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Effect of Honeybee (*Apis mellifera* L.) Pollination on Fruit Yield and Yield Contributing Parameters of Watermelon (*Citrullus lanatus* L.)

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Abstract

Pollination is a critical link in the functioning of ecosystems and it improves the yield of crops. Honeybees are responsible for 70-80% of insect pollination. As a result the need for insect pollination is becoming popular by agricultural community to increase the productivity of crops. Hence, this study was conducted to evaluate the effect of honeybee pollination on fruit yield and yield contributing parameters of watermelon (*Citrullus lanatus* L.). The study was conducted in a randomized complete block design (RCBD) with three treatments and three replications in an experimental plot size of 4m x 6m. Treatments applied were: (i) plots caged with bees (T1), (ii) open plots allowing free visits of bees + other pollinators (T2) and (iii) plots caged without bees (control-T3). The obtained data related to fruit yield and other yield parameters were statistically analyzed using one way analysis of variance (ANOVA). The results revealed that the highest fruit yield was found in fruit crops caged with honeybee (57472 kg/ha) followed by open pollinated fruit crops (38361 kg/ha), whereas fruit crops excluded from insect had the lowest fruit yield (22472 kg/ha). So it concluded that honeybee and insect pollination had a significant effect on fruit yield and quality of watermelon. Therefore, it is recommended to keep sufficient number of honeybee colonies in watermelon fields during the flowering period to increase the pollination efficiency and thereby enhance watermelon fruit productivity.

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Watermelon, Pollinators, Honeybees, Fruit yield

Introduction

Pollination plays an important role in flowering plant reproduction and fruit set for wild plant communities (Corbet *et al*. 1991; Buchmann and Nabhan 1996). Estimates showed that up to 90% of all flowering plant species rely on pollination by insects such as bees (Richards 1986; Buchmann and Nabhan 1996). Of the six known types of pollination agents (insects, birds, wind, gravity, water and mammals) insects are by far the most important in pollination (McGregor, 1976), being responsible for 80-85% of all pollination (Johannsmeier and Mostert, 2001). The biggest groups of insect pollinators are solitary bees, bumblebees and honeybees (Free, 1993), probably due to their large number of hairs and their behavioral patterns such as feeding their young on nectar and pollen (Pitts-Singer, 2008). Among these honeybees are substantially important in world's agricultural economy, in that 35% of the world's food production relies on pollinators, of which the honeybee accounts for 70-80% which is the largest portion (Winfree, 2007). As well as honeybees have a well-developed communication system that enables individual bee to be alerted to the needs of the colony and to the location of suitable food source. The pollinating potential of a single honeybee colony becomes evident when it is recognized that its bees make up to 4 million trips per year and that during each trip an average of about 100 flowers are visited (Free, 1993).

Nowadays the natural habitat is disturbed for many reasons and the vegetation cover is declining worldwide (Kearns *et al*., 1998). Agriculture plays a great role in declining native pollinators through the modification and elimination of pollinator habitats and the use of chemicals (pesticides, fertilizers). Therefore when many hectares are occupied by a single crop and moreover certain localities are selected for growing particular cultivars there may be too few insect pollinators due to the factors mentioned above and it may be necessary to enhance pollinators in that area (Du Toit, 1988).

Honeybee pollinators are required for producing up to 30% of the human food supply directly or indirectly and the farmers rely on managed honeybees throughout the world to provide these services (Greenleaf and Kremen, 2006). The contribution of managed honeybee pollination to crop production and quality has been estimated to be more than the value of honey and wax production (Shrestha, 2004). For all the United States, the annual value of increased agricultural production in yield and quality that is attributed to honeybee pollination varied from US$9.3 billion in 1989 to US$14.6 billion in 2000 (Morse and Calderone, 2000).

Watermelon (*Citrullus lanatus*) belongs to the family Cucurbitaceae, it also contains a bottle gourd, ridge gourd and snake gourd. Watermelon is a flowering vine-like plant of the family Cucurbitaceae. The fruit of watermelon is composed of 93% water, small amount of minerals, proteins, fats, carbohydrates, lycopenes and vitamins (Namdari, 2011). Its flesh is rich in citrulline; a source of arginine amino acid, which is a substrate for the synthesis of nitric oxide and is associated with cardiovascular and immune roles in humans (Dube *et al*., 2020). Watermelon is a warm seasonal crop, with an optimal crop growth at 38˚C and above in temperature and between 28-32˚C temperature for germination. Its fruits are consumed as dessert and are appreciated for their sweet flesh full of juice. In some parts of Africa, the seeds of watermelon are roasted and eaten (Zoro *et al*., 2003) but are also used to produce oil. Watermelon is an important horticultural crop providing a means of livelihood to most residents in central part of Ethiopia by ensuring food security and creating employment opportunities for the people (Amenti *et al*., 2009). Local demand in the country is on the rise due to population increase especially in the urban areas, where fruits are sold in open air markets and by street vendors located in residential streets in towns and cities. Consequently, both large scale and small-scale water melon farming can be profitable. While high yield is a major goal for watermelon farmers. However, the production in quantity and quality of fruit yield was low in the absence of honeybee pollination.

Watermelon is self-incompatible crops that required honeybees and other insects' for cross pollination. Among insect pollinator agents, honeybees are known to be the most efficient pollinating agents of cucumber for many years (McGregor, 1976). Honeybees are known to increase the yield of Cucurbitaceous crops by 100 to 150% (Mel'nichenk, 1977). According to (Shemetkov, 1957) a cucumber flower should be visited 8 to 10 times for satisfactory fruit set, but the number of seeds and weight of fruit increases by 40-50 honeybee visits. The cucumber plants were caged before bloom to illustrate that female blossoms need to be pollinated by insect to set fruit and placed a strong colony of *Apis mellifera* to supplement a limited supply of wild pollinators (Coleman, 1979). A study was carried out to show that 15 to 20 bee visits were needed to get uniform cucumbers and multiple bee visits increased the average number of seeds which resulted in better and maximum fruit weight. In several regions of the world, watermelon flowers are exclusively pollinated by insects with A. mellifera as the most efficient. In Ethiopia, there is no information concerning the role of honeybees' pollination on watermelon yield and yield contributing parameters. Therefore, this study was initiated to see the role of managed honeybee pollination on fruit yield and yield parameters of watermelon and to identify potential pollinators of watermelon other than honeybees.

Materials and Methods

Description of study area

The study was carried out at Adami Tulu Agricultural Research Centre (ATARC) located in Adami Tulu Jido Kombolcha district, East Shoa Zone of Oromia, Ethiopia. Adami Tulu Agricultural Research Center is situated in mid–rift valley, 167 km south of Addis Ababa on Hawassa road. It lies at latitude of 7°9' N and 38°7' E longitude. It has an altitude of 1650 meters above mean sea level and average annual rainfall is 760.9mm. Rain fall is bimodal and unevenly distributed that extends from February to September with a dry period in May to

June, which separates the preceding short rains from the following long rains. The means maximum and minimum temperatures are 12.6°C and 27°C, respectively. The soil type is fine, sandy loam with sand: silt: clay in the ratio of 34: 38: 18, respectively. The average pH of the area is 7.88 (ATARC, 1998).

Experimental Layout

The experiment was arranged in a randomized complete block design (RCBD) with three treatments and three replications each. The plot size (4 x 6 m^2) consists of two rows with row to row distance of 2m and plant to plant distance of 1m. Treatments applied were: (i) plots caged with bees (T1), (ii) open plots allowing free visits of bees + other pollinators (T2) and (iii) plots caged without bees (control - T3). Proper site selection was made to minimize risk of pests and disease occurrence, to reduce fertility difference of the experimental plots, flooding and water logging conditions. Land was prepared by digging the ground and smoothing. Good looking seeds with uniform size were planted. Variety of Charleston Gray 33 watermelon seeds were planted under rain fed condition with supplemental irrigation in months with inadequate rains during the production period with recommended agronomic package practices. Five seeds were planted per hole and then thinned to one seeding per hole after two-true leaves to minimize the risk of having vacant hill. The crop condition was observed daily, intensity of flowering was recorded. Honeybee colonies used in this experiment received supplementary feeding (dissolved sugar) and water before and after they were placed in the cages.

Data collection

Flower visitation surveys

Flower visitor surveys were done for four consecutive for five consecutive days to assess which and how many insect species were visiting the watermelon plants. Watermelon plants that have approximately above 40 flowers were selected and number of honeybees visiting at these flowers was recorded for 15 minutes at 1 h interval surveys from 6.00 to 18.00 h (7– 8 h, 9 –10 h, 11–12 h, 13 – 14 h, 15– 16 h, 17- 18h). In a slow walk along all labeled flowers of treatment, the identity of all insects that visited watermelon flowers was recorded. All insects encountered on flowers were recorded and the cumulated results expressed in number of visits to determine the relative frequency of honeybee.

Thousand seed weight (test weight)

The dependence of watermelon crop on honeybee pollination (i.e., the contribution of managed honeybee pollination to the crop yield) was measured. This was done by comparing the seed yield from watermelon accessible to all flower visitors, the seed yield from watermelon not accessible to any insects (self pollinated) and the seed yield from watermelon accessible only to honeybees. This can be expressed formally as:

$$\text{yield increment (\%)} = \frac{(\text{yield from honeybee pollinated} - \text{yield from insects excluded})}{\text{Yield from open pollinated}} (100)$$

Germination percentage

Observations were also taken on germination percentage of the seeds of all three treatments. For germination potential test 100 seeds of watermelon were sprinkled on a 10 cm diameter petri plate which was covered with moist filter paper. Moisture was maintained by spraying water. The filter paper was removed after the germination was over and the number of seeds germinated out of hundred was counted. The experiment was replicated three times. A germination success study was conducted by considering the principle of maximum percentage germination, following the necessary steps used by the International Rules for Seed Testing (ISTA, 2009). Finally germination percentage of seeds was determined by using the following formula:

$$Germination\ Percentage = (\frac{n}{N}) \times 100\%$$

where:

n = Total number of germinated seeds;

N = Total number of seeds in the sample (Labouriau and Agudo, 1987).

Number of seeds per fruit

This observation was made by selecting fifteen fruits at random, from each replication of treatment during harvesting. The seeds in each fruit were counted and mean seeds per fruit were calculated.

Fruit setting percentage

The percentage of fruit set was calculated as follows:

$$\frac{\text{Number of watermelon produced}}{\text{Number of female flowers}} \times 100$$

Number of fruit per plant

In each plot ten plants were selected randomly and number of fruits in these plants was counted. Average number of fruits per plant was calculated.

Weight (kg), circumference of fruit, and length of fruit (cm)

Randomly selected five fruits in each replication were harvested, weighed and their circumference/length was measured individually and then average fruit weight/ circumference/length was calculated.

Yield (kg)

During the whole fruit harvesting period yield was recorded on five randomly selected plants and calculated the average yield per plant. Later on, yield per hectare was calculated by multiplying the yield of one plant with the number of plants in one hectare.

Statistical analysis

The obtained data related to fruit yield and yield parameters were statistically analyzed using one way analysis of variance (ANOVA) and least significant difference (LSD) was calculated to identify the significant differences among the treatments means.

Results and Discussions

Flower visitation

Various insects belonging to different orders visited watermelon flowers during its flowering time. These include honeybee, butterflies, flies, wasps, moths, Bumblebee and Xylocopa. Amongst the 821 insect visitors recorded on watermelon flowers during the study period, honeybee is the major pollinator of watermelon with 548 visit (67%) in the open pollination on the field, while moth was the least abundant visitors in the open pollination (table 1).

Fruit setting percentage

Significant variation was observed in number of fruit set per plant between fruit crops caged with honeybee and caged without pollinators (table 2). Among the three treatments, fruit crops caged with honeybee had the highest number of fruit per plant 12.5/plant followed by open pollination 7.4/plant, whereas fruit crops caged without any pollinator had the least number of fruit per plant 4.3/plant (Table 2).

Number seeds per fruit

Significant variation was observed in number of seeds/fruit among the treatments (table 3). Among of the three treatments, plots caged with honeybees had the highest number of seeds/fruit 588 seeds followed by open pollinated 526 seeds, whereas the plots caged without pollinators had the least number of seeds/fruit 430 seeds (Table3). Prakash *et al.,* (2004) found that the number of seeds per fruit and average fruit weight in bee pollinated plants might be attributed to the sufficient number of pollen grains received by the flowers which were best provided by honeybees in caged conditions as compared to caged without bees and open pollination. This also might be due to the adequate pollination done by honeybee inside the cage whereas this study obtained the lowest value in yield in case of without honeybee pollination. Closely related results were reported by Meisels (1997) in a greenhouse experiment involving sweet paper and using bees as pollinators in which the number of seeds per fruits were increased as compared with the control.

Fruit diameter

Superior fruit diameter 27.9 cm was recorded in honeybee pollinated treatment followed by open pollinated 27.3 cm and caged without any pollinator 22.9 cm (Table 2). Statistical analysis of data shows significant differences between honeybees pollinated and open pollinated treatments but no statistical significance difference between pollination caged with bees and open. Hossanian *et al.,* (2018) also stated that lower fruit diameter was found in pollination caged without honeybee. It was concluded that honeybees (*A. millifera* L.) pollination was correlated positively with fruit diameter of watermelon.

Fruit yield /kg/ha

Significant variation was observed among the treatments regarding fruit yield/ha (table 2). Results obtained show that the highest fruit yield /ha was found in fruit crops caged with honeybee 57472 kg/ha followed by open pollinated plants 38361 kg/ha, while fruit crops excluded from insect had the lowest yield 22472 kg/ha (Table

2).Honeybee pollination increases the yield of watermelon fruit by 91.2% compared to the yield in the absence of honeybees. This indicated how much honeybees are contributing to seed yield increment of this valuable crop. The higher yield of fruit crops caged with honeybees might be due to the higher pollination efficiency of honeybee inside the cage. The fruit yield difference between the treatments indicated that the crop requires insect pollination particularly honeybees for fruit production. Delaplane and Mayer (2014) also stated that, the number of pollen grains deposited on the stigma by pollinators is directly related to seed formation, which often determines fruit size; provide greater fruit yields and treatments without honey bee colony that provided lower water melon, pumpkin and cucumber seed numbers, fruit yields and weights. It has been identified that pollination should be realized by honeybees in 90% in order to harvest more and higher quality fruits in *citrullus lanatus* (Ozbek, 2008). Velthuis *et al*., (2002) also conducted study it has been found that the honeybees provide a significant increase in *citrullus lanatus* pollination compared to wind and small insects pollination and the highest yield has been obtained from the field allowed free for honeybee entrance.

Weight of fruit (kg)

Fruits from honeybee pollinated of watermelon were bigger and weighed more compared to those from control watermelon. The average watermelon fruit weight in honeybee pollinated plots was the highest 7.5 kg followed by open pollinated 6.0 kg, whereas the least 4.37 kg was recorded for plot received the caged without any pollinator (Table 2).

Thousand seeds weight (gm)

The treatment caged with honeybee produced significantly and statistically superior test weight (3.65 g/1000 seeds followed by open pollinated 3.5g/1000 seeds and caged without honeybee and any pollinator 2.17g/1000 seeds (table 3). An increment of 34% in 1000 seeds weight of watermelon was obtained by placing honeybee (*Apis mellifera*) colonies in watermelon fields in comparison to that without any pollinator (Table 3). S.Alan, Bradley and Taylor (2015) also reported that water melon (*citrullus lanatus)* fruit weights increased when honeybee colonies were included. Natural pollination was insufficient to stimulate maximum fruit size development and seed weight per fruit. Although watermelon (*Citrullus lanatus*) fruit set will occur with natural pollinators, addition of honeybee colonies will ensure the presence of pollinators to maximize fruit size.

Germination percentage

Insect pollination had immense impact on seed germination. Significantly higher per cent germination 28.36% was recorded in honeybee pollinated treatment as against 21.57% and 13.60% per cent in the open pollinated and caged plot without bees, respectively (Table 3). The increase in germination rate in the open plots is a result of a superior pollinating efficiency of honeybees. Similarly Yücel and Duman (2005) reported that the germination rate was greater on average by 12% in onion with honeybee activity.

Table.1 Insect visitors on watermelon crop during the flowering period

Insect order	Common name	Total number	Percent (%)
Hymenoptera	Honeybee	548	67
	Bumblebee	157	19
Orthopter	Xylocopa	61	7.4
Dipter	Fly	24	2.9
Lepidopter	Butterfly	13	1.6
	Moth	7	0.9
Orthopter	Wasp	11	1.3

Table.2 Effect of bee pollination on fruit yield and yield contributing parameters of watermelon

Treatments	Yield (kg/ha)	No. of fruit/plant	Fruit wt (kg)	Fruit diameter (cm)
Caged with honeybees	57472±6.41[a]	12.5±0.78[a]	7.50±0.50[a]	27.9±0.36[a]
Open pollinated	38361±4.83[c]	7.4±1.20[c]	6.0±0.50[c]	27.3±0.45[a]
Caged without honeybee	22472±2.22[b]	4.3±0.99[b]	4.37±0.24[b]	22.9±0.61[b]
LSD (5%)	38.4	1.7	0.68	1.2
CV (%)	10.24	7.36	3.21	6.08

Means followed by different letters within a column are significantly different at the 5% level of probability using Tukey Student Test (USD).

Table.3 Effect of Honeybee Pollination on Yield & Yield attributing parameter of watermelon fruits

Treatments	Total no. Seeds/fruit	1000 seeds wt.(gm)	Germination (%)	Marketable yield (kg/ha)
Caged with honeybee	588±28.7[a]	3.65±0.25[a]	28.36±3.2[a]	48524±2.85[a]
Open pollinated	526±15.5[c]	3.50±0.10[a]	21.57±6.5[c]	31467±1.2[b]
Caged without honeybee	430±32.9[b]	2.17±0.00[b]	13.60±2.4[b]	13248±2.52[c]
LSD (5%)	18.34	1.32	3.58	17.04
CV (%)	3.12	13.02	3.66	4.84

Marketable fruit yield/kg/ha

The mean yield of marketable fruits matured per plot was statistically significant (P<0.05).The average marketable fruit yield is 48524 kg/ha, if caged with honeybees and 31467 kg/ha for plant left open to all insect pollinators. Honeybee pollination increases the yield of marketable watermelon fruit yield by 36.4% (Table 3).

Conclusions and Recommendation

The study revealed that honeybee and other insect pollinators had significant effect on fruit yield of watermelon. The highest fruit yield was found in plants caged with honeybee (57472 kg/ha), whereas plants excluded from insect had the lowest yield (22472 kg/ha). Therefore, it is recommended that moving honeybee colonies to watermelon production areas during the flowering period is essential for maximum fruit production and quality increment.

Conflict of interest

The authors have not declared any conflict of interest regarding this publication

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