

Pollination and Foraging Potential of European Bumblebee, *Bombus terrestris* (Hymenoptera: Apidae) on Tomato Crop under Greenhouse System

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Abstract.- Commercial production of tomato crop in glasshouse needs proper pollination practices. Bumblebees (for example, *Bombus terrestris*) serve to increase the tomato in quantitative and qualitative way for better economic outcome. The present study was performed in high glasshouse tomato crops with Grandella and Cherry tomato cultivars under controlled environmental conditions. Self, manual and bumblebee pollination methods with different parameters were studied with bumblebee foragers trafficking from hives to flowers for first, fourth and seventh week with foraging time per flower for effective pollination. Effective foraging was observed till 7th week of observation and visitation rate was significantly more in Cherry as compared to Grandella cultivar with more visitation rate per flower. Significant increase in both qualitative as well as quantitative parameters was observed with increased fruit size, roundness and numbers per truss. Bumblebees helped to decrease the need for manual pollination, increased yield and quality of the green house tomato crops. Importance of bumblebees and selection of cultivar for good crop under high tunnel cropping systems is suggested.

Key words: Bumblebee, foraging pattern, pollination, tomato yield, enclosed greenhouse farming

INTRODUCTION

Crop pollination is vital function performed by different vertebrate and invertebrate pollinators to sustain the plant and animal terrestrial ecosystem. About 75% of agricultural crops of the world depend on insect pollination for fruit set and seed development (Klein *et al.*, 2007). World food volume increase 35% by pollination services, supplying essential nutrients for humans (Klein *et al.*, 2007; Gallai *et al.*, 2008). Physical and chemical characters of fruits like roundness, weight, seed number, flesh, acids and vitamin C have positively been affected with adequate pollination (Ikeda and Tadauchi, 1995, Morandin *et al.*, 2001). Agricultural crops vary in their pollination requirements and rely on different insect pollinator species (Morse and Calderone, 2000; Garibaldi *et al.*, 2013). Tomato anthers require vigorous agitation for pollen release, the so called “buzz-pollination” (Buchmann, 1983). Honeybees and

bumblebees are important commercial crop pollinators with high economic returns (Velthuis and van Doorn, 2006).

Bumblebees are more efficient and reliable pollinators of greenhouse crops helping in fruit production of high quality due to their high speed of pollination, buzzing behaviour and efficiency at low temperature and sunlight (Kaftanoglu, 1999; Winston, 2001; Paydas *et al.*, 2000). High cost of labor for manual pollination with vibrators and growth regulators are also used, but considered as less effective than bumblebees. Use of insect pollination within greenhouse, especially bumblebees gave cost effective and attractive substitute of manual pollination (Velthuis and vanDoorn, 2006). An external and internal environmental condition along with foraging efficiency of bumblebees has impact on required pollination of tomato cultivation. Glasshouse cultivation of tomato, pepper and eggplant are under expensive crop cultivation. This requires precise soil, water, nutrients, diseases and pests control and pollination practices to be followed effectively. Absence of wind and plant movements along with pollinators in greenhouse environment affects badly proper tomato pollination (Free, 1970; Banda and

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Paxton, 1991), an important challenge for the producers.

Activity of bumblebees foraging was important to serve as successful pollinators under these greenhouse conditions when compared with honeybees. Bumblebees can fly and pollinate flowers under cool conditions due to their better thermoregulatory abilities (Corbet, 1996). They have better adaptive qualities for pollen and nectar collection near their hives and preference increases in small patches with flower abundance (Sowig, 1989; Sheikh *et al.*, 2014). Different species of bumblebees forage on variety of flowering plants depending upon length of flower corolla, tongue length and amount of nectar produced by flower (Harder, 1982; Heinrich, 1979; Corbet, 1995). *Bombus terrestris* with short tongue forage efficiently on flowers having short corolla tube as in tomato flowers (Velthuis and vanDoorn, 2006).

Bumblebees have been regularly used for crop pollination under such glasshouse intensive cropping systems for high yield crops throughout the year. Different bumblebee species including *B. terrestris*, *B. impatiens*, *B. ignitus* and *B. ephippiatus* are in year round rearing and then wide use to meet the pollination needs (Vergara and Buendia, 2012). More than a million bumblebee hives are used for the purpose to meet pollination needs for these crops (Velthuis and van Doorn, 2006). To increase fruit set and yield in tomatoes, an important vegetable grown worldwide, and other solanaceous crops, various techniques like plant growth regulators, manual vibrators, honeybees and bumblebees pollination service have been used (Paydas *et al.*, 2000; Sun *et al.*, 2006). Parameters like high yield, more number of seeds, high specific gravity and better fruit firmness resulted from bumble bee pollinated tomatoes compared to plant vibration and plant growth regulators (Dogterom *et al.*, 1998; Morandin *et al.*, 2001; Al-Attal *et al.*, 2003). Floral scent as attractant of pollinators to tomato flowers varies in different tomato cultivars, which could affect bumble bee foraging (Kunze and Gumbert, 2001; Laloi and Pham-Delegue, 2004; Dobson, 2005). Indigenous bumblebee species, *B. haemorrhoidalis* Smith has been identified and its rearing and breeding as indigenous species is underway to avoid problems carried by the invasive

species (Sheikh *et al.*, 2014). We planned to determine the foraging behavior and effectiveness of *B. terrestris* foraging workers for flower visitation time and frequency at different day times with pollination role of *B. terrestris* in comparison with other methods for greenhouse tomato for quantitative and qualitative production, based on seeds per fruit, weight size and shape.

MATERIALS AND METHODS

The present study was conducted in a greenhouse hydroponics farm (19,800m²) in Kalyam, Pakistan. The greenhouses were rectangular in shape made with glass-aluminum structure. Daily temperature and relative humidity were maintained between 18-27°C and 75-85%, respectively with cooling pad wall and exhaust fans. Density of tomato plants was 2.5 plants per square meter. Two cultivars named as Grandella and Cherry tomato were grown in pots of Rockwool which were of 12 cm² and placed in coco peat slab of 100 cm long and 23 cm wide placed 40 cm above the ground level. These plants were maintained according to standard commercial practices in two separate greenhouses with first flowering truss appeared from early March and keep on flowering till October.

Eight bumblebee boxes of *Bombus terrestris* were imported from Koppert Biological Systems, Netherlands to meet the pollination requirement of foraging bumblebees. Each bumblebee hive was placed at equal distance from each other for maximum flower visitation by these foraging bumblebees. In order to evaluate the pollination efficiency of bumblebee foragers, time spent per flower was measured in seconds by using stopwatch, which started with the bumblebee landing on flower until it worked on it and left for another one. This bumblebee trafficking was monitored for five minutes for four colonies in each greenhouse for these two cultivars. During this period, incoming and outgoing bees were recorded at three different timings: 8-9 am, 11-12 pm and 5-6 pm of the day during first, fourth and seventh week of arrival of bumble bee colonies and data were recorded. Number of visits per flower on both cultivars by bumblebee foragers was observed for their preference for visit to collect pollens. Bee

trafficking was also compared statistically for three observation times and means were compared at 5% probability using DMR test. Visitation rate of bumblebee foragers were presented graphically and value of correlation was estimated using MS Excel software.

Manual pollination was done daily for a week with a vibrator (manual pollinator). Six trusses in six different plants of Grandella and Cherry tomatoes were manually pollinated with vibrator and covered with muslin cloth bags of 34cm long and 24cm wide in order to avoid bumble bee interaction with those trusses. Similar number of trusses was bagged with muslin cloth bags and no bumblebees nor manual pollination was practiced and another six trusses were pollinated by bumble bees. Fruits were picked for the first week bumblebee visitation on maturity for both cultivars, respectively. Diameter and height of tomatoes were measured in millimeter by using Vernier calipers and weight of tomatoes was taken in grams by using electric balance. Number of seeds of each tomato was counted and roundness of tomato fruits was taken (Dogterom *et al.*, 1998). Number of fruits per truss, seed count, fruit weight and roundness were compared statistically using ANOVA performed using MSTATC software (MSTAT-C, 1983) and means were compared using LSD test at 5% probability.

RESULTS

Bumblebee foragers were observed for first, fourth and seventh weeks for three times of the day i.e., 8-9 am, 11-12 pm and 5-6 pm after arrival from the Supplier company with eight weeks suggested effective foraging life. The results for foraging bumblebees statistically differ in their foraging rate for the first week with maximum number of bees (9.0) going out from their bee hives for pollination at first observation time followed by 7.25 at midday and 1.0 at evening time. There existed non-significant difference in bees' number leaving for pollination during fourth observation week during morning and midday times, but lowest and significantly different at evening time. There was minimum number of bumblebee foragers left for pollination in morning time of the seventh week

with no visitation observed during midday and evening time. Overall foraging rate was more in morning and midday time as compared to that at evening time of observation (Table I).

Table I.- Interaction between different observation weeks and times for bumblebee (*Bombus terrestris*) foraging for greenhouse tomato pollination

Weeks	Time of observation (five minutes per bee hive)		
	0800-0900	1100-1200	1700-1800
1	9.00 a	7.25 b	1.00 d
4	2.75 c	3.25 c	1.13 d
7	0.38 d	0.00 d	0.00 d

Means sharing a similar letter in rows are non-significant at probability less than 0.05 using LSD test.

On Grandella cultivar, bumblebees spent 1-4 seconds per flower with mean value of 2.2 ± 0.8 seconds and 1.1-2.2 seconds with mean value of 1.4 ± 0.3 on Cherry flowers, showing more variation in time spent per flowers on Grandella than cherry flowers. Flowers visitation was comparatively more frequent on Cherry flowers than that of Grandella, respectively 10.6 ± 3.5 and 22.6 ± 3.2 flowers per minute. Visitation marks on Grandella and cherry flower pistils showed more visits per flower on Cherry flowers (2.7 ± 0.9) than that of Grandella flowers (1.2 ± 0.4) (Table II).

Comparison of pollination methods performed on Grandella and Cherry showed variation in quantitative as well as qualitative parameters. In Grandella, maximum fruit weight (104 g/fruit) was recorded with bumblebee foragers followed by manual pollination (74.4 g/fruit). The least fruit weight was obtained by self pollination (20.6 g/fruit), which was five times less than bumblebee visited treatment and near to four times less than manual pollination method. Bumblebee pollination fruits bear more seeds (126 seeds/fruit) than manually pollinated with 102 seeds/fruit. Least number of seeds was formed in self-pollinated flowers (9.63 seeds/fruit). However, bigger values for maximum fruit height, diameter and roundness were found in bumblebee pollinated treatment, followed by manual and self pollination, but not much significant. Bumblebee pollinated Cherry

Table II.- Comparative foraging efficacy of *Bombus terrestris* foraging bumblebees on Grandella and Cherry tomato cultivars under greenhouse cultivation system.

	Grandella tomato cultivar				Cherry tomato cultivar			
	Time spent (s) /flower	Flowers visited / minute	Visitation marks / flower	Flowers / truss	Time spent (s) /flower	Flowers visited / minute	Visitation marks / flower	Flowers / truss
Minimum	1.0	5.0	1.0	5.0	1.1	18.0	1.0	14.0
Maximum	4.0	18.0	2.0	9.0	2.2	28.0	4.0	25.0
Mean	2.2±0.8	10.6±3.5	1.2±0.4	6.9±1.3	1.4±0.3	22.6±3.2	2.7±0.9	18.8±2.7

Table III.- Qualitative and quantitative comparison of bumblebee pollination, manual pollination and self pollination of tomato cultivars Grandella and Cherry under greenhouse farming system.

Parameters	Grandella tomato			Cherry tomato		
	Bumblebee	Manual	Self	Bumblebee	Manual	Self
Weight (g)	104 a	74.4 b	20.6 c	8.5 a	6.2 b	1.9 c
Seed (count)	126.5 a	102.4b	9.63 c	78.8 a	33.8 b	6.2 c
Height (mm)	53.3 a	48.3 b	27.9 c	22.5 a	18.3 b	12.9 c
Max. diameter (mm)	56.3 a	54.0 a	30.3 b	23.1 a	18.4 b	12.0 c
Roundness	1.2	2.2	3.4	1.8	6.0	5.3

Means in the row followed by different letters are significantly different at the *P*, 0.05 level as determined by ANOVA by the least significance difference test for two cultivars separately.

flowers yielded higher fruit weight (8.5 g/fruit) followed by manually and self-pollinated flowers (6.2, 1.9 g/fruit), respectively. More seeds were formed in bumblebee pollinated fruits as compared to manually and self pollinated fruits (Table III).

DISCUSSION

Bumblebee foragers preferred to visit the flowers during morning and midday than evening time with more time spent, flowers visitation and visitation marks per flowers on Cherry flowers than Grandella. Their foraging for pollination during morning time has also been observed in hot pepper but differed not only for another *Bombus* species, *B. impatiens* but also for the same species received from different bumblebee breeders and suppliers with higher activity during midday (Meisels and Chiasson, 1997; Kwon and Saeed, 2003; Roman and Szczêsna, 2008). In our study, *B. terrestris* foraging workers actively pollinated at both times of morning and midday as they observed. Such variation might exist due to the use of different *Bombus* species as well as weather conditions maintained inside

greenhouse during hot months (Heinrich, 1975). Present studies were conducted during May to July hot months in the study area and variation in greenhouse might have influenced on foraging preference of bumblebees. Foraging flights are regulated by the combined effect of light and temperature, however, non-significant differences existed for different species to these parameters affecting their foraging on high-density forage plants (Lundberg, 1980; Roman and Szczêsna, 2008). Such variation might be due to field condition observations under subalpine to alpine region with lower temperature and sunlight than tropical region. However, relatively higher temperature were required for the foraging worker bumblebees than males and queens. Higher foraging rates might be due to easy temperature adjustment around 25°C than other day times which makes them to easily achieve desired temperature necessary for their muscle to fly (Heinrich, 1975). Similarly low intensity of light in morning time may also have influenced on their activity pattern (Kwon and Saeed, 2003). Other factors for later day lower foraging activity may include high temperature,

amount of brood, workers strength per colony and colony completion of food needs (Kwon and Saeed, 2003). Too high or too low temperature and light intensity influenced and retarded foraging activity in greenhouse tomato crop (Roman and Szczêsna, 2008). Bumblebee foragers even do not visit flowers of different varieties randomly but depends on density of available of resources to forage (Lefebvre and Pierre, 2006). We observed relatively greater number of flowers per truss on Cherry than Grandella, which might influence in foraging for energetic gains more easily available in Cherry cultivar flowers than that of Grandella (Heinrich, 1972). More number of bumblebee visitations per flowers in present study was adequate for pollination of both cultivars more prominently in Cherry (Morandin *et al.*, 2001).

Their high speed of pollination, buzzing behaviour and efficiency at low temperature and sunlight make them reliable pollinators of greenhouse crops helping in fruit production (Kaftanoglu, 1999; Winston, 2001; Paydas *et al.*, 2000) with managed cost alternatives to manual pollination and growth regulators (Velthuis and Van Doorn, 2006). However, floral scent varies in different tomato cultivars, which could affect bumble bee foraging (Kunze and Gumbert, 2001; Laloi and Pham-Delegue, 2004; Dobson, 2005). Forage on variety of flowering plants depends upon the length of flower corolla, tongue length and amount of nectar produced by flower (Harder, 1982; Heinrich, 1979; Corbet, 1995) highlighting short-tongued *B. terrestris* on tomato flowers (Velthuis and van Doorn, 2006).

Qualitative and quantitative comparison of bumblebee, manual and self-pollination also strengthen their utility for pollination of enclosed farming crops like tomato. Fruit weight, seed count and better fruit formation were significantly more in bumblebee pollination treatment as compared to manual and self-pollination. Pollination services either used by manual pollination with the help of vibrators or bee-pollination improved fruits yield and their chemical characters (Ikeda and Tadauchi, 1995; Morandin *et al.*, 2001). However, bee-pollination is considered as easier and better option to increase the fruit set and yield than other pollination methods (Paydas *et al.*, 2000; Sun *et al.*,

2006). Bumblebee pollination services not only helped to increase the crop yield but also decreased manual pollination practice supporting the finding of the present study (Dogterom *et al.*, 1998; Morandin *et al.*, 2001; Al-Attal *et al.*, 2003). Absence of wind and plant movements along with pollinators in greenhouse environment badly affect proper tomato pollination, which highlights the important role of bumblebees in crop pollination for better economic returns (Free, 1970; Banda and Paxton, 1991).

Number of bumblebee hives, their active duration and preference for a specific cultivar may serve as important factors to maintain their optimum activity. Present studies stress the use of bumblebee hives as monthly incorporation rather than required numbers after every two months. The presence of overlapping in foraging bumblebees better pollinate crop than that of their introduction after every two months to support the previously placed bumblebee hives. Selection of high yielding cultivars can help to increase the economic returns when these foraging bees can result in 100% crop pollination. We suggest the use of bumblebees as crop pollinators for better and improved tomato crop under tunnel farming and greenhouse enclosed farming in Pakistan. More foraging of Bumblebees in tomato cultivars need to be adjusted on spatial flower distribution, which needs to be considered to maximize their foraging and ultimately their crop yields.

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Conflict of interest declaration

There exists no conflict of interest among authors.

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