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Pollination in chili pepper (*Capsicum annuum* L.) by *Trigona laeviceps* and *T. minangkabau* (Hymenoptera, Meliponini)

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Abstract

Two species of stingless bees, *Trigona minangkabau* and *T. laeviceps*, have been known to visit chili pepper plantation in West Sumatera. The objective of research is to study pollination by *T. minangkabau* and *T. laeviceps* in increasing chili pepper yield. It was conducted using Randomized Block Design (RBD) with 3 replicates. Pollination by *T. laeviceps* and *T. minangkabau*, each could increase the following parameters: fruit sets 12.32 and 9.66%, number of seeds 56.36 and 45.91%, number of fruits 29.31 and 25.06, fruit weight per plant 66.46 and 49.75%, and yields ha^{-1} 54.26% and 40.83% if compared to pollination by wind. However, it did not affect length and diameter of fruit. Average flower handling time by *T. minangkabau* was 26.00 ± 4.52 seconds and by *T. laeviceps* was 26.23 ± 4.46 seconds. Time taken to move to other flower by *T. minangkabau* was 7.64 ± 2.7 seconds and by *T. laeviceps* was 7.87 ± 1.88 seconds. Based on all parameters above it could be concluded that both *T. minangkabau* and *T. laeviceps* had the same effectiveness as pollinators on chili pepper plantations.

Keywords: *Trigona minangkabau*, *T. laeviceps*, cross pollination, chili pepper

Introduction

Trigona spp. which are called stingless bees can function as pollinator. There are two species of stingless bees, *Trigona minangkabau* and *T. laeviceps*, found in chili pepper plantations in West Sumatera distributing evenly either in low or high lands. These insects have relatively small size, 3.4 ± 0.2 mm and 4.4 ± 0.2 mm, therefore they are able to obtain nectar from small size flowers^[1]. As a consequence, stingless bees have more various foods than honey bees. Chili pepper flowers are chasmogamy, pollination occurs at the time of opening of the flowers. Thus, in chili pepper there is possibility for cross pollination to occur. This is the reason why the production of chili pepper per plant is higher when planted in group than individually. The increase of yield due to cross pollination was 7.6-36.8%^[2].

Some species of stingless bees that have been used as pollinators to increase production of *Capsicum* are *Melipona favosa*^[3], *Trigona carbonaria*^[4], *Melipona subnitida*^[5], effective to increase production of sweet pepper, but not with *Tetragonisca angustula* which is very small in size. Stingless bee that has been used in research for chili pepper pollination is *Trigona (Tetragonula) minangkabau* and proved to be effective to increase chili pepper production compared to natural pollination or by wind^[6].

The chili pepper flower has small size (10-15 mm), campanulate to rotate flower type with depth to flower base 2 mm^[7]. Having these characters, chili pepper needs small pollinators to visit. In this case it was interesting and important to study the effectivity of pollination done by *Trigona minangkabau* and *T. laeviceps* in increasing yield of chili chili pepper.

Materials and Methods

The study was conducted in rice field in Kelurahan Korong Gadang Kuranji Subdistrict, Padang ($-00^{\circ} 57' \text{ LS } 100^{\circ} 21' \text{ BT}$, altitude 20 m dpl.) from August 2014 – January 2015. A trial was arranged using Randomized Block Design (RBD) with three treatments, pollination by wind, *T. minangkabau* and by *T. laeviceps*. Each treatment was done with three replicates. Data were analyzed by Analysis of Variance (One way ANOVA) and Duncan's New Multiple Range Test (DNMRT) with 5% level of confidence^[8].

Chili pepper plants were planted in 9 plots ($11 \times 1 \text{ m}^2$) with a distance of 0.5 m among them. To avoid pollination by other insects, the plots area was screened using plastic screen with 36 mesh. Three screen houses ($12 \times 4 \times 3 \text{ m}$) were made and each of them contained 3 plots.

Stup of *Trigona* sp. the zise (33 x 25 x 19 cm) with numbers of workers \pm 1500 individuals were placed in screen houses when the plants had produced 25% bloom. Parameters measured were fruit set (percentage of flowers resulted 10 fruits), length and diameter of fruits (cm), fruit weigh (g), number of seeds per fruit, number and weight of fruits per plant, production per ha, and foraging behavior.

20. ults

Fruit set, number of seeds, length and diameter of fruit

Trigona minangkabau and *T. leaviceps* as pollinators increased fruit sets significantly in which percentages of fruit set resulted from pollination by *T. minangkabau* and by *T. leaviceps* were 78.84% and 81.50%. These numbers were significantly different from the one pollinated by wind, 69.18%. *T. minangkabau* and *T. leaviceps* could increase fruit sets 9.66% and 12.32%.

Trigona minangkabau and *T. leaviceps* also increase number of seeds. Chili pepper plants pollinated by *T. minangkabau* and *T. leaviceps* produced 64.20 and 68.80 seeds, which were significantly different from the one pollinated by wind, 44.00 seeds. In this case, *T. minangkabau* and *T. leaviceps* could increase number of seeds 45.91% and 56.36%. However, pollination by *T. minangkabau* and *T. leaviceps* did not affect length and diameter of fruits. The length of fruits ranged 18.75 -21.06 cm and diameter ranged 1.03-1.07 cm (Table 1).

Table 1: Percentage of fruit sets, number seeds, length, and diameter of fruits of chili pepper pollinated by *Trigona* sp and wind

Pollinator	Fruit set (%)	Number seeds	Fruit length (cm)	Fruit Diameter (cm)
<i>Trigona laeviceps</i>	81.50 ^a	68.80 ^a	20.47 ^a	1.07 ^a
<i>T. minangkabau</i>	78.84 ^a	64.20 ^a	21.06 ^a	1.05 ^a
Wind	69.18 ^b	44.00 ^b	18.75 ^a	1.03 ^a

Numbers followed by the same letters were not significantly different according to DNMRT with 5% level of confidence

4

Number and weight of fruits per plant, and yield

Since *T. minangkabau* and *T. leaviceps* could increase percentage of fruit 4s and number of seeds, they automatically increased number and weight of fruits per plant. As a result, they increased the yield per ha (Table 2)

Table 2: Number and weight of fruits per plant and yield of chili pepper ha⁻¹ pollinated by *T. leaviceps*, *T. minangkabau* and wind

Pollinator	Fruit Number per plant	Fruit Weight per plant (g)	Yield per ha (kg)
<i>T. leaviceps</i>	82.93 ^a	562.00 ^a	8186.0 ^a
<i>T. minangkabau</i>	80.20 ^a	505.67 ^a	7473.7 ^a
Wind	64.13 ^b	337.67 ^b	5306.7 ^b

Numbers followed by the same letters were not significantly different according to DNMRT with 5% level of confidence

21

The increase in number of fruits per plant by *T. minangkabau* and *T. leaviceps* as pollinator was significantly different from the one done by wind. Average number of fruits per plant pollinated by *T. minangkabau* and *T. leaviceps* were 80.20 and 82.93 individuals, while the one by wind was 64.13 individuals. The increases in fruit numbers by *T. minangkabau* and *T. leaviceps* were 25.06% and 29.31% compared to the one by wind.

Because of increasing number of fruits per plant 25.06 - 29.31%, and number of seeds 45.91 - 56.36%, there should be an increase in fruit weight per plant which was 505.67 g using

T. minangkabau and 562.00 g using *T. leaviceps*. That numbers were significantly different from the one pollinated by wind, 337.67 g. Percentage of fruit weight increase per plant was 49.75% by *T. minangkabau* and 66.46% by *T. leaviceps*. By the increase of fruit weight per plant it would automatically increase yield per ha which were 7473.71 kg by *T. minangkabau* and 8186.00 kg by *T. leaviceps* different significantly from the yield pollinated by wind, 5306.73 kg. As a result, the increases of yield per ha were 40.83% by *T. minangkabau* and 54.26% by *T. leaviceps*.

Food Foraging Behavior

It was found that flower handling time by *T. minangkabau* was 16.81-33.72 seconds with average 26.23 \pm 4.46 seconds and by *T. leaviceps* was 17.28 - 35.71 seconds with average 26.00 \pm 4.52 seconds. Time taken to move from one to another flower for *T. minangkabau* was 5.04-14.42 seconds with average 7.64 \pm 2.7 seconds and for *T. leaviceps* was 5.48-11.28 seconds with average 7.87 \pm 1.88 seconds. Both parameters did not show significant different between the two stingless bees (Table 3)

Table 3: Flower handling time and time taken to move from one to another flower by two species of stingless bees

Foraging behavior	Stingless bees spesies	
	<i>T. minangkabau</i>	<i>T. leaviceps</i>
Flower handling time (second)	16.81-33.72 (26.23 \pm 4.46)	17.28-35.71 (26.00 \pm 4.52)
Time taken to move from one to another flower (second)	5.04-14.42 (7.64 \pm 2.7)	5.48-11.28 (7.87 \pm 1.88)

Before alighting on flower, *T. leaviceps* showed movement like Z letter, while *T. minangkabau* did not show specific pattern.

Discussion

Pollination is a sexual reproduction process which result in fruits forming and inside fruits there are seeds. Inside ovule there is ovulum. The perfect pollination mainly supported by insect pollinators would increase yield and in turn increase number of seeds. Generally, pollination supported by eusocial bees would contribute to increase in yield, both qualitatively and quantitatively^[9, 10].

Effectiveness of pollination supported by *Trigona minangkabau* and *T. leaviceps* could increase yield of chili pepper by increasing 9.66% fruit set, 45.91% seed numbers by *T. minangkabau* and 12.32% fruit set and 56.36% seed numbers by *T. laeviceps*. Increasing fruit set and seed numbers automatically would increase fruit numbers, 25.06% by *T. minangkabau* and 29.31% by *T. laeviceps* (Table 2.).

Many other researchers have found the same result from pollinating by insect pollinators. Santos *et al.* (2004) compared effectivity of pollination on tomatoes by *Melipona quadrifasciata* and *Apis mellifera*. The result showed that the size of tomatoes was bigger, higher weight, and more seeds when pollinated by *M. quadrifasciata* compared to the one pollinated by *A. mellifera*^[11]. Cruz *et al.*, (2004) also reported the increase in number of fruits 13, 51%, number of seeds 85.85% and fruit weight 29.69% on sweet pepper because of pollinating by *Melipona subnitida* compared to pollinating by wind^[5]. Jarlan *et al.* (1997) reported the increase of yield of *Capsicum* up to 19.3% due to pollinating by stingless bees^[12]. Zwon and Saeed (2003) found the increase in fruit weight 27.2% and number of seeds of 47.8% of *Capsicum* due to pollinating by bees^[13].

Pollination on sweet pepper supported by various species of stingless bees showed that not all species were successful as

pollinators. It depended on size of stingless bees. *Melipona favosa*, *Trigona carbonaria*, and *Melipona subnitida* having bigger size were effective to increase yield of sweet pepper, but *Tetragonisca angustula* having smaller size could not make [5, 4, 14]. Howe [16] for *T. minangkabau* and *T. leviceps* with body size 3.4 ± 0.2 mm and 4.4 ± 0.2 mm and included subgenus *Tetragonula* showed the same effectivity as pollinators.

Besides species of pollinator, architecture of flowers like size, the site of reproduction organ, accessibility to nectar, and flower structure, could affect interaction between plant and its pollinator. If there is an incomplete factor like small size of flower, thus a big size of insect is not appropriate to be pollinator [15, 16].

Sakagami *et al.* (1985); Osawa and Tsubaki, (2003) described that having smaller size (2-14 mm) enabled the stingless bees to access various flowers with a very narrow opening that could not be accessed by other bees. Therefore, for chili pepper the appropriate pollinator is *Trigona* spp [17, 18]. Suryani (1999) reported that using *Trigona minangkabau* as pollinator on chili pepper plant could effectively increase fruit set (65%) compared to pollinating by other insects (59%) and wind (17%) [6].

Frequency and duration of visit by pollinators to nectariferous depend on nectar production level [19]. Nectar consists of 25-75% sugar with various numbers of amino acids and lipids. Generally, biotic pollinators need physical contact with reproductive organ of flower for pollinating to occur [20]. Due to their small body size of *Trigona minangkabau* and *T. leviceps*, when obtaining nectar their heads could enter up to the base of flower and their legs contact with pollen so that pollination could occur. Besides that, in foraging behavior, stingless bees tends to show flower constancy [21]. In this case there would be repeated visit to the same flower. Thus, *Trigona minangkabau* and *T. leviceps* were found to be effective as pollinator for chili pepper plants.

Conclusion

Pollination on chili pepper plants supported by *Trigona minangkabau* and *T. leviceps* was effective in increasing fruit set and seed numbers. By increasing the fruit set and seed numbers it would automatically increase fruit numbers per plant and weight and in turn would increase yield ha^{-1} . *Trigona* spp. as pollinator did not affect length and diameter of fruit. Flower handling time and time taken to move from one to another flower between *T. minangkabau* and *T. leviceps* were not significantly different. *T. leviceps* made Z letter movement before alighting while *T. minangkabau* did not show specific pattern before alighting on flower.

Acknowledgement [13]

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