CROSS-POLLINATION PRODUCES ARABICA COFFEE BEANS WITH HIGHER CAFFEINE AND LOWER FATTY ACID**PENYERBUKAN SILANG HASILKAN BIJI KOPI ARABIKA TINGGI KAFEIN DAN RENDAH ASAM LEMAK**Dian Anggria Sari¹⁾, Ramadhani Eka Putra^{2)*}

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How to cite:Sari, DA, RE Putra. 2021. Cross-pollination produces Arabica coffee beans with higher caffeine and lower fatty acid. *Journal of Tropical Biology* 9 (3): 185-189.**ABSTRACT**

*Studies showed that cross-pollination also affect fruits and green bean coffee quantity and quality yet study on this subject in Indonesia rarely found. This study focused on assessing the pollination effect of fruit quality and beans in terms of size, weight, physical defect, and content of caffeine and fatty acid. During this study, *Tetragonula laeviceps*, indigenous stingless bees, were applied as a pollination agent to be compared to open pollination. The result showed that open pollination produced bigger beans ($16.73ab \pm 1.57 \times 14.66a \pm 1.48$ mm) with higher caffeine (18.22%) and lower fatty acid (41.86%), while application of *T. laeviceps* produced slightly heavier beans (0.22 gram). Besides producing higher caffeine and lower fatty acid, cross-pollination services mainly required wild pollinators, although *T. laeviceps* could act as a potential replacement or for the inundation of this service.*

Keywords: caffeine, coffee beans, cross-pollination, fatty acid, self-pollination

ABSTRAK

*Sejumlah penelitian menunjukkan bahwa penyerbukan silang juga mempengaruhi kuantitas dan kualitas buah dan biji kopi, dan studi pada subjek ini cukup jarang di temukan di Indonesia. Studi ini menekankan penilaian pada dampak polinasi terhadap kualitas buah dan biji dalam hal ukuran, berat, cacat biji dan kandungan kafein serta asam lemak. Pada penelitian ini, *Tetragonula laeviceps*, salah satu lebah lokal, diaplikasikan sebagai agen penyerbuk untuk dibandingkan dengan penyerbukan terbuka. Pada penelitian ini ditemukan bahwa perlakuan penyerbukan terbuka dihasilkan biji kopi dengan ukuran lebih besar (16.73 ± 1.57 mm) dengan kandungan kafein yang lebih tinggi (18.22%) dan asam lemak yang lebih rendah (41.86%) sementara aplikasi *T. laeviceps* menghasilkan biji lebih berat (0.22 ± 0.03 mm). Selain menghasilkan biji kopi dengan kandungan kafein lebih tinggi dan asam lemak lebih rendah, proses penyerbukan silang secara signifikan membutuhkan penyerbuk liar, walaupun *T. laeviceps* dapat berpotensi untuk menggantikan atau menambah servis ini.*

Kata kunci: kafein, biji kopi, penyerbukan silang, asam lemak, penyerbukan sendiri

INTRODUCTION

As one of the important ecological services in agricultural systems, pollination service in nature is the result of the interaction between plants and animals, especially insects [1, 2]. This Pollination services help to maintain the biodiversity of the plant and productivity of major crops, which is strongly related to human food security [3]. Furtherly, studies showed the importance of the pollinator, mostly insects, especially bees, for crop and economic plant production and economic benefit derived from the products of insect-pollinated plants [4, 5].

Bees had been known as efficient and effective pollinators among insect pollinators [1]. However, many reports stated that the diversity of wild bees is declining, and rise a big concern for pollination service in natural and managed ecosystems, which

relies on the bees species as pollinators [6]. To maintain productivity, many plantations and urban agriculture applied managed insect pollinators to replace and inundate wild pollinators. The most common managed pollinator used in Indonesia is *Apis cerana* L. or Asiatic honeybees. However, most farmers are reluctant to apply this species as pollinator agents due to its aggressive and absconding behavior. Another indigenous bee species, *Tetragonula* sp., Meliponini, the stingless bee, a great diverse and abundant bee species that inhabit the tropical and subtropical area, could be used as a potential pollinator in plantation and agriculture. This species formed perennial colonies, which forage all year and play an important role as pollinators of many plants species [7, 8, 9, 10, 11].

Arabica coffee (*Coffea arabica*) is one of the important Indonesian economic plants and

commodities in the world. Indonesia is the second-largest coffee producer in Asia and Oceania in 2020 [12], which could be due to the number of beans produced from Arabica coffee being less than the other coffee variety [13]. *Coffea arabica* is considered a highland coffee plant and able to produce beans by self-pollination, and the number of fruit sets could increase by improving the frequency of insect pollinator visitation [14, 15].

Klein et al. [14] reported that *Tetragonula* sp., as one of the wild bees visited a coffee flower, yet the application of managed *Tetragonula* sp. as coffee pollinator has not been reported. In this study, the effectiveness of *Tetragonula* sp. application and some concerns regarding its application were observed and discussed.

METHODS

Research area. The research was conducted at the urban coffee plantation in Dusun Wangun, Rancakalong, Sumedang, West Java, $\pm$ 1100-1200 m above the sea level and grown under *Pinus merkusii* forest as shading at 6.82422, 107.80964 ($6^{\circ}49'27.2''S, 107^{\circ}48'34.7''E$).

Pollination experiment. Four pollination experiments, i.e., close pollination, hand pollination, open pollination, and *T. laeviceps* pollination, were conducted to observe the efficiency of *T. laeviceps* as a pollination agent of local Arabica coffee. Eight to ten coffee shrubs were randomly selected as sources of flowers. For each experiment, four different branches and all of the flower buds in the branch (approximately 30-80 flowers), depends on the availability, were bagged with nylon mesh ($\varnothing$ 1 mm). The pollination experiments: (1) closed pollination group, flowers bagged with nylon mesh, to exclude insect pollination; (2) hand pollination group, pollen of same flowers transferred to other stigmas in the same plant with a fine brush then bagged with nylon mesh to imitate self-pollination; (3) open pollination, all bloomed flowers uncovered which allow insect and wind pollination; (4) *T. laeviceps* pollination, bloomed flowers visited by *T. laeviceps* bagged with nylon mesh until fruit produced. The level of pollination success was measured by the proportion of fruit produced from the total observed flowers.

Fruit size measurement and chemical compound analysis. The impact of pollination treatment on the quality of fruit produced was determined by measuring the weight and diameter of ripe fruit using digital scales and digital calipers.

After the measurements, fresh fruit was processed into green beans using the wet processing method no later than 6-8 hours after harvest. The chemical compound analysis in *C. arabica* seeds between pollination treatments was

carried out using *Pyrolysis Gas Chromatography-Mass Spectrometry* (Py-GC-MS) Shimadzu type GCMS-QP2010.

Data analysis. The differences in the quality of the coffee beans (size, weight, and physical defect) were tested by One way ANOVA at a significant value for the test was $P < 0.05$, and LSD was applied as a post hoc test if the significant value was found. All tests were conducted by SPSS 16.0 Statistic Program.

RESULTS AND DISCUSSION

Fruit and green bean coffee size. There was a positive correlation between the size and weight of the beans. The coffee beans produced by closed pollination were slightly shorter than other groups (ANOVA, $P < 0.05$), while other parameters were relatively similar among all groups (Table 1).

Table 1. Fruit and coffee green bean measurements from pollination treatments

	OP	HP	TP	CP
Length (mm)	16.73 ^{ab} ± 1.57	16.62 ^{abc} ± 1.60	16.07 ^b ± 1.53	15.71 ^a ± 1.66
Width (mm)	14.66 ^a ± 1.48	14.34 ^a ± 1.52	14.15 ^a ± 1.80	14.29 ^a ± 1.63
Fruit weight (gr/each)	2.22 ^a ± 0.59	2.23 ^a ± 0.55	2.09 ^a ± 0.61	2.08 ^a $\pm$ 0.59
Seeds without physical defect (%)	83.54 ^a ± 8.48	84.68 ^a ± 8.81	79.83 ^a ± 13.77	85.83 ^a $\pm$ 14.91
Bean weight (gr/seed)	0.18 ^a ± 0.03	0.19 ^a ± 0.06	0.22 ^a ± 0.03	0.19 ^a ± 0.02

OP: Open Pollination, HP: Hand Pollination, TP: *Tetragonula* Pollination, CP: Closed Pollination. The mean value of treatments followed by the same letter is not significantly different ($P > 0.05$), followed with the *Mann-Whitney U* test.

A study by Karanja et al. [16] showed that coffee bean weight and coffee cup quality were affected by pollination types (in open and hand pollination), which means pollination not only increases crop yields but also improves the taste and coffee aroma. This study also supported the importance of non-managed social bees as pollinators of coffee, as shown by bigger fruit produced through open pollination [17]. However, in terms of economic, the application of *T. laeviceps* as a pollination agent improved the weight of the beans (part of coffee with the highest economic value) although not significant than beans produced by open and hand pollination. This result indicated the high potency of *T. laeviceps* to be applied as a potential pollinator agent and inundate the pollinator population at the coffee plantation.

Chemical content of green bean Arabica coffee. Caffeine, palmitic acid, and linoleic acid were the most dominant chemical constituent in all treatments (Table 2). The caffeine concentration was different in each treatment, in which the highest concentration was founded in green beans produced by open pollination treatment, and the lowest concentration was produced by closed pollination treatment. Furtherly, the highest concentrations of palmitic acid and linoleic acid were recorded from beans produced by closed pollination.

Table 2. Chemical constituent in green bean Arabica coffee from different pollination treatment

Chemical content (%)	CP	TP	HP	OP
Caffeine, C ₈ H ₁₀ N ₄ O ₂	12.92	14.42	11.09	18.22
Hexadecanoic acid - Palmitic acid, C ₁₆ H ₃₂ O ₂	27.69	21.49	26.27	26.89
9,12-Octadecadienoic acid - Linoleic acid, C ₁₈ H ₃₂ O ₂	22.27	14.39	15.81	14.97

OP: Open Pollination, HP: Hand Pollination, TP: *Tetragonula* Pollination, CP: Closed Pollination

Caffeine, tannin, fixed oil, carbohydrates, and proteins are commonly known as the main chemical constituent in coffee [18]. Caffeine is the most widely consumed group of alkaloids in the world and is mostly found in coffee beans. Caffeine in nectar could increase the pollination success by increasing the visitation rates of bees as the main pollinator of coffee [19, 20]. Increasing visitation may improve the number of pollen deposited in stigma and produced high-quality beans susceptible to herbivore attack. The increasing caffeine content in the bean may be related to the protection mechanism of the bean. Caffeine is part of the defense of coffee beans as caffeine has been recognized as an antifungal, certain phytotoxin, and chemosterilant against several types of insects [21, 22, 23].

Most of the fatty acid contained in green coffee bean is saturated fatty acids; palmitic and stearic acids, and unsaturated acids; like oleic and linoleic acids. Palmitic acids and linoleic acids were triglyceride fatty acids commonly founded on green coffee beans and roasted bean oil. Linoleic acid content was commonly found for 40-45% and palmitic acids for 25-35% from total fatty acids in green bean and roasted coffee bean [24, 25, 26]. Fatty acids are important for human health since they have roles as bio-regulators of many cellular metabolic processes [27]. Linoleic acid is an essential fatty acid required by humans from external sources, which could contribute to

decreasing the ratio of low-density lipoproteins to high-density lipoproteins and participates in the synthesis of prostaglandins and other biological processes related to cell regeneration and lack of these fats can lead to skin disorders [28, 29]. Saturated fatty acids, including stearic and palmitic acid, are potential to determine the quality of specialty coffees, and unsaturated fatty acids, such as oleic, linoleic, and linolenic acid, can be associated with fragrance, less intense acidity, and flavor in coffees [30].

CONCLUSION

In conclusion, cross-pollination improves the physical quality of Arabica coffee and changes the chemical constitution of beans produced with higher caffeine and lower fatty acid. *Tetragonula laeviceps* has the potency to be utilized as a replacement or inundation for pollination services provided by wild pollinators.

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