



Research article

Production of propolis and honey from *Tetragonula laeviceps* cultivated in Modular *Tetragonula* HivesMuhammad Yusuf Abduh^{a,b,*}, Abdurrahman Adam^a, Muhammad Fadhlullah^a, Ramadhani Eka Putra^a, Robert Manurung^a^a School of Life Sciences and Technology, Institut Teknologi Bandung, Indonesia^b University Center of Excellence for Nutraceuticals, Bioscience and Biotechnology Research Center, Institut Teknologi Bandung, Indonesia

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ABSTRACT

Propolis and honey produced by stingless bees are regarded as high economic value products due to their bioactive components, which are significantly influenced by conditions at the cultivation location. This study investigated the effect of cultivation location on the amount and quality of propolis and honey produced by *Tetragonula laeviceps* cultivated in Modular *Tetragonula* Hives. Fifteen bee colonies were cultivated for at least three months in coffee plantations at two different locations, namely Cibodas and Cileunyi Wetan, Indonesia. The propolis was harvested from the hives and then evaluated to compare product quality from each location. The average production of propolis in both locations was found to lie in the range of 4.26–4.54 g/colony/month with a flavonoid content of 11.4–14.8 mg/g qE. Meanwhile, the average production of honey in both locations after eight months of cultivation was found to lie in the range of 0.93–1.44 g/colony/month. The vitamin C content of the honey obtained from both locations was 17.2–69.5 mg/100 g with an IC₅₀ of 1188–1341 mg/L, in terms of its ability to inhibit the free radical 2,2-diphenyl-2-picrylhydrazyl. This study shows that cultivation of stingless bees on a coffee plantation in the studied locations has the potential to provide sustainable production of propolis and honey from *T. laeviceps*.

1. Introduction

T. laeviceps is a species of stingless bee commonly found in tropical and sub-tropical areas, including Indonesian forest and settlement areas (Putra et al., 2017; Suriawanto et al., 2017). The hives of *T. laeviceps* may be found in bamboo trees, wooden walls, and iron cavities (Suriawanto et al., 2017). Unlike honey bees such as *Apis mellifera*, stingless bees do not have either a functional sting or a hexagonal nest. Stingless bee nests are typically oval-shaped, and can be categorized into egg pots and food pots (Roubik, 2006). *T. laeviceps* produces less honey than honey bee species (Chanchao, 2013). However, stingless bees produce up to six-times more propolis than honey bees produce, to compensate for their stingless attributes (Kothai and Jayanthi, 2015).

Propolis is the material produced by stingless bees to build the structure of their hives, which protect the bee colonies against macro- and microorganism threats (Kothai and Jayanthi, 2015). Studies have shown that propolis and honey produced by stingless bees have therapeutic effects due to their bioactive components. Therefore, propolis and

honey produced by stingless bees are regarded as high economic value products (Chanchao, 2013). It has been reported in the literature that the antioxidant and phenolic content of the propolis produced by stingless bees is higher than that of the propolis produced by honey bees (Muruke, 2014). In addition, the honey produced by stingless bees also contains higher amounts of phenolic, flavonoid and vitamin C in comparison with the honey produced by honey bees (Muruke, 2014).

The bioactive components in propolis and honey are significantly influenced by the source of resin, nectar and pollen available at the cultivation location (Kasote, 2017). Integrating cultivation of *T. laeviceps* with a local coffee plantation in West Java has development potential because the coffee plantation has pine trees as well as coffee. The pine trees provide a rich source of resin, nectar and pollen, which supports production of high quality propolis and honey (Ramalho et al., 1990). This study focuses on integrating cultivation of *T. laeviceps* with two coffee plantations at Cibodas and Cileunyi Wetan in West Java.

Meliponiculture, as commonly practiced by local farmers in Indonesia, involves cultivating stingless bees using bamboos, which is

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not efficient for the commercial production of propolis and honey from *T. laeviceps* (Contrera et al., 2011). A recent development in the cultivation of *T. laeviceps* is the use of Modular *Tetragonula* Hives (MOTIVEs) equipped with a detachable propolis frame containing small holes that trigger stingless bees to fill in the holes with propolis. Unlike the conventional method, which may damage the bamboos during the harvesting of propolis and honey, MOTIVEs minimize potential disturbance and damage to the bee colonies (Hakim and Abduh, 2019). MOTIVEs also separate propolis on the propolis frame from the bees and from non-propolis products. The use of MOTIVEs for cultivation of *T. laeviceps* may improve the sustainability, production and quality of the propolis compared with conventional cultivation using bamboos. This study investigates the influence of local coffee plantations on the amount of crude propolis and honey produced by *T. laeviceps* cultivated using MOTIVEs, and examines the composition and quality of the propolis extract.

2. Materials and methods

2.1. Colonies of *T. laeviceps*

Colonies of *T. laeviceps* cultivated in bamboos were obtained from a stingless bee cultivation center in Cibeusi Village, Subang, West Java, Indonesia.

2.2. Modular *Tetragonula* Hive

The Modular *Tetragonula* Hive used in this study is a wooden box (21 cm × 14 cm x 18 cm) equipped with a cover and a detachable propolis frame. The detachable propolis frame is located between the box and the cover and contains small holes that trigger *T. laeviceps* to cover the holes with propolis (Hakim and Abduh, 2019). The detachable propolis frame

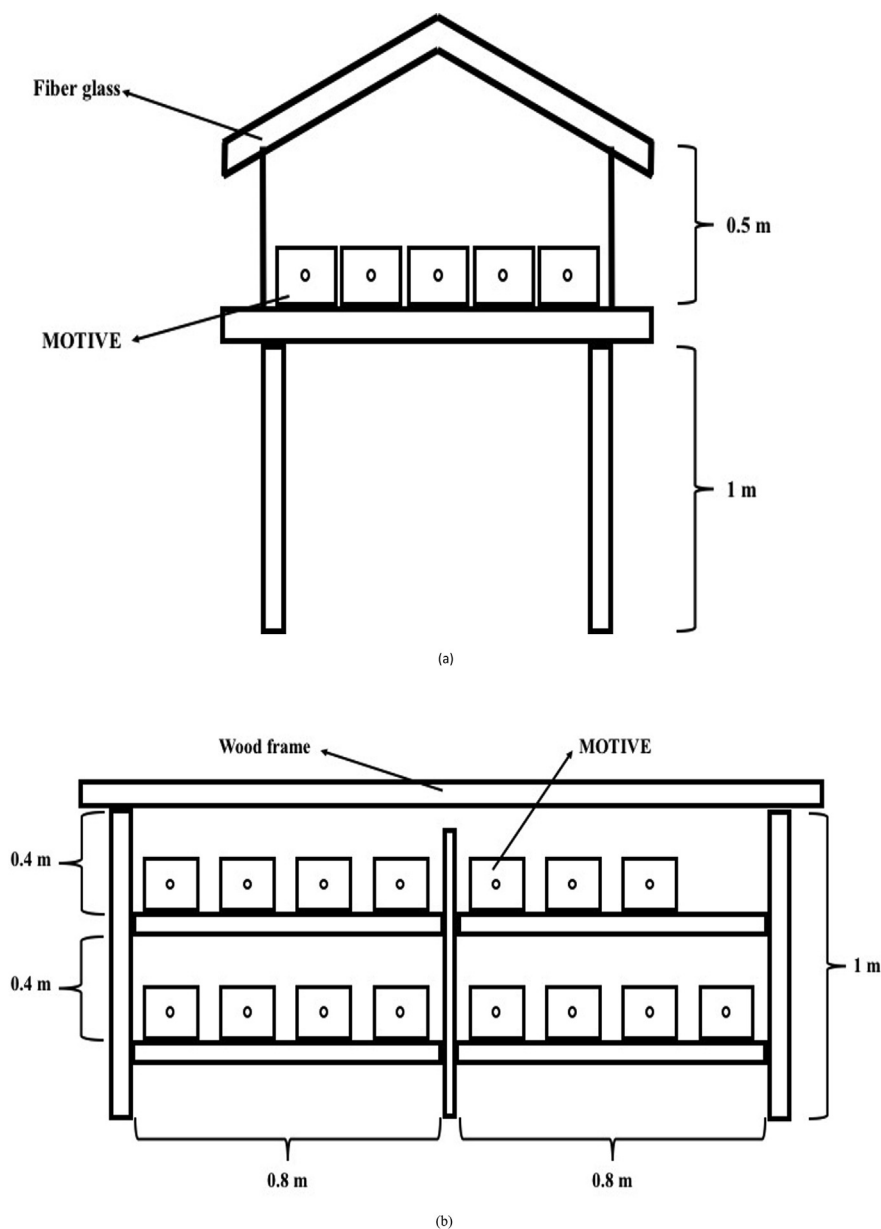
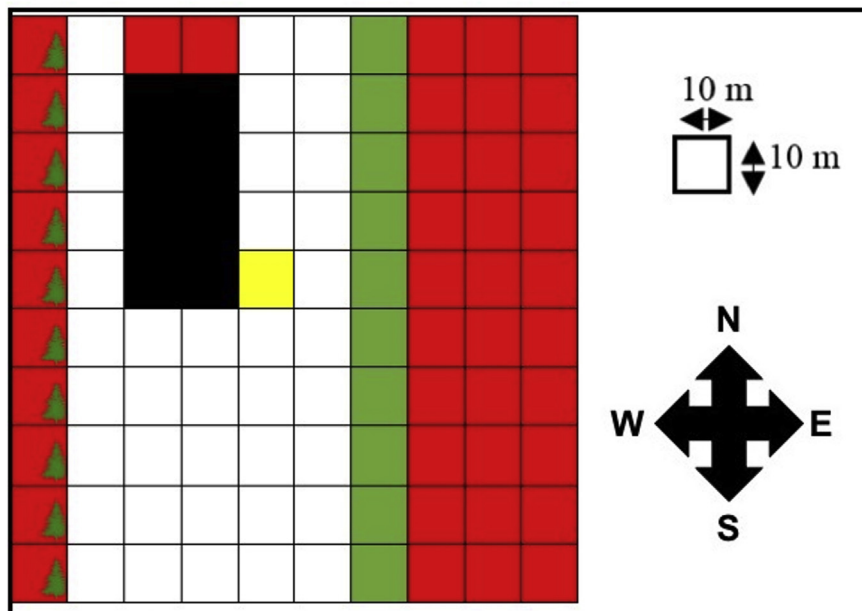
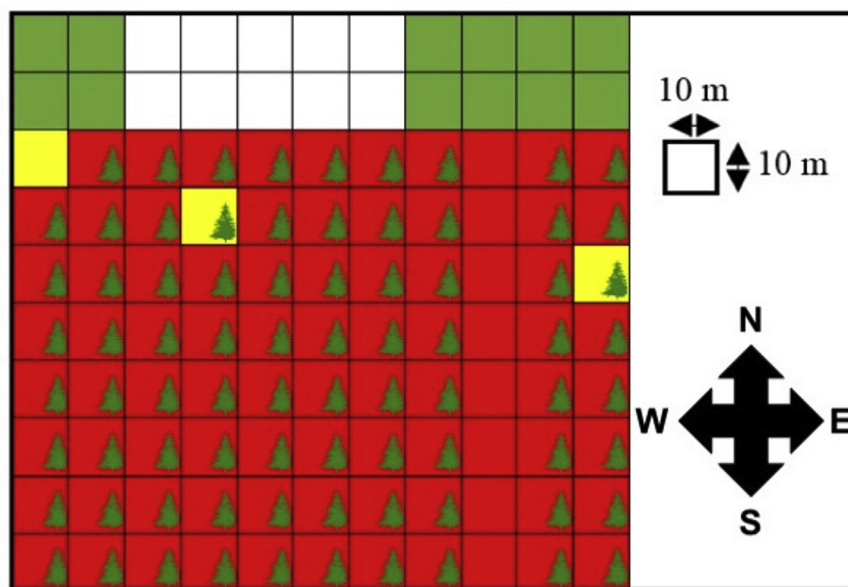


Figure 1. Construction of bee hives in Cibodas Village (a) and Cileunyi Wetan Village (b).



(a)



(b)







| Symbol | Description |
|---|-------------------|
|  | Coffee plantation |
|  | Pines |
|  | Bee hive |
|  | Variety of plants |
|  | Empty land |
|  | House |

Figure 2. Layout for the placement of bee hives and surrounding vegetation in Cibodas Village (a) and Cileunyi Wetan Village (b).

may be easily replaced with a new frame after the holes have been filled with propolis.

2.3. Transfer of *T. laeviceps* colonies from bamboos to Modular *Tetragonula* Hives

Thirty colonies of *T. laeviceps* were carefully transferred from bamboo hives to MOTIVEs at a stingless bee cultivation center in Subang. Each MOTIVE contained only one bee colony, comprised of a queen bee, bee brood, propolis and honey pots. Each MOTIVE was weighed before and after the transfer of the bee colonies. The bee colonies were then acclimatized for one week inside the MOTIVE at the original location at Subang. After acclimatization, 15 bee colonies (1–15) were relocated to a coffee plantation in Cibodas, West Bandung Regency. The bee colonies were placed at three nearby sites, with five bee colonies at each site. The other 15 bee colonies (A – O) were relocated to a coffee plantation in Cileunyi Wetan, Bandung Regency, where all the colonies were placed at one site. All colonies were then acclimatized for one week at the new locations. Figure 1 illustrates the construction of the bee hives, while Figure 2 shows the location of the bee colonies and surrounding vegetation in Cibodas and Cileunyi Wetan, respectively.

2.4. Harvesting of propolis and honey produced by *T. laeviceps*

Harvesting of propolis was carried out every two weeks for a cultivation period of three months. The propolis frames containing small holes that had been covered by propolis were removed from the MOTIVE and weighed to calculate the amount of propolis produced by each colony at the different locations. Afterwards, a new propolis frame containing small holes was placed inside each MOTIVE, to be harvested two weeks later. The first harvesting of honey was carried out after a three month cultivation period (August–October 2017). The second harvesting was carried out after an additional five months of cultivation (November 2017–March 2018). The honey produced by *T. laeviceps* bees was harvested by introducing a hole on top of the honey pot followed by suction using a pipette. The empty honey pots were also weighed to determine their weight.

2.5. Extraction of propolis

Propolis that had been harvested from the MOTIVEs was extracted according to the procedures as suggested by Machado et al. (2016). Crude propolis obtained from the propolis frames was cut into 1 cm² with a thickness of <0.5 mm and then placed inside an Erlenmeyer flask. Ethanol (absolute for analysis, Merck, Germany) was added to the flask to extract the propolis, using a crude propolis-to-ethanol ratio of 1:100 on a weight-to-volume (w/v) basis. The crude propolis - ethanol mixture was then stirred using an orbital shaker (FALC F350, Novolab, Belgium) in dark condition at 200 rpm and 25 °C for 36 h. Afterward, the mixture was filtered by a Whatman No. 2 filter paper and dried. The filtrate was kept at 18 °C in a dark vial for further analysis. The insoluble content (in %) that was separated from the crude propolis was calculated according to Eq. (1).

$$\text{Insoluble content (\%)} = [(m_{I,\text{initial}} (\text{g}) - m_{I,\text{final}} (\text{g})) / m_{\text{propolis}} (\text{g})] \times 100\% \quad (1)$$

where $m_{I,\text{final}}$: = dry weight of insoluble and filter paper in grams, $m_{I,\text{initial}}$: = dry weight of filter paper in grams, m_{propolis} : = dry weight of propolis in grams.

2.6. Determination of flavonoid and solubility content in propolis extract

Flavonoid content in the propolis extract was determined according to the procedures as suggested by Machado et al. (2016). Quercetin (Sigma Aldrich, Singapore) was mixed with methanol (Sigma Aldrich, Singapore) to prepare a standard solution with a concentration range of 0–40 mg/L. Each solution (2 mL) was then poured into a falcon tube, mixed with 2% (w/v) aluminum chloride (Sigma Aldrich, Singapore) and

incubated for 30 min. Absorbance of the solution was measured by using a UV-1800 spectrophotometer (Shimadzu, Japan) at a wavelength of 415 nm. The flavonoid content in the propolis extract (qE content_{extract}) in mg/mL was calculated according to Eq. (2).

$$\text{qE content}_{\text{extract}} (\text{mg/mL}) = [(\text{Abs} + 0.0055) \times N] / 7.3 \quad (2)$$

where Abs: = measured absorbance of the sample, N: = dilution factor with a value of 20 for this study.

Soluble content in the propolis extract was determined by heating 0.5 mL of propolis extract solution using a steam cup at 45 °C. The soluble content in g/mL was then calculated by Eq. (3), with the dried filled steam cup ($m_{\text{cup,final}}$) and dry empty steam cup weight ($m_{\text{cup,initial}}$) in grams and propolis extract volume (V_{extract}) in mL.

$$\text{Soluble content (\%)} = [(m_{S,\text{final}} (\text{g}) - m_{S,\text{initial}} (\text{g})) / V_{\text{extract}} (\text{mL})] \times 100\% \quad (3)$$

where $m_{S,\text{final}}$: = final weight of steam cup and sample in grams, $m_{S,\text{initial}}$: = initial weight of steam cup and sample in grams, V_{extract} : = volume of sample in mL.

2.7. Determination of flavonoid content and solubility of crude propolis

The flavonoid content in crude propolis (qE content_{crude propolis}) in mg/g was estimated according to Eq. (4), whereas the solubility of crude propolis in ethanol (%) was estimated using Eq. (5)

$$\text{qE content}_{\text{crude propolis}} (\text{mg/g}) = \text{qE content}_{\text{extract}} (\text{mg/mL}) \times [V_{\text{extract}} (\text{mL}) / m_{\text{propolis}} (\text{g})] \quad (4)$$

where qE content_{extract}: = flavonoid content in the propolis extract in mg/mL, V_{extract} : = volume of propolis extract in mL, m_{propolis} : = weight of crude propolis in g.

$$\text{Solubility}_{\text{crude propolis}} (\%) = [\text{soluble content (\%)} \times V_{\text{extract}} (\text{mL}) / m_{\text{propolis}} (\text{g})] \times 100\% \quad (5)$$

where soluble content: = soluble content in the propolis extract in %, V_{extract} : = volume of sample in mL, m_{propolis} : = initial weight of crude propolis.

2.8. Analytical methods to determine the physicochemical parameters of honey

The physicochemical parameters of honey produced by *T. laeviceps*, particularly moisture content, reducing sugars, antioxidant activity and vitamin C, were analyzed at Sibaweih Laboratories (Bandung, West Java,

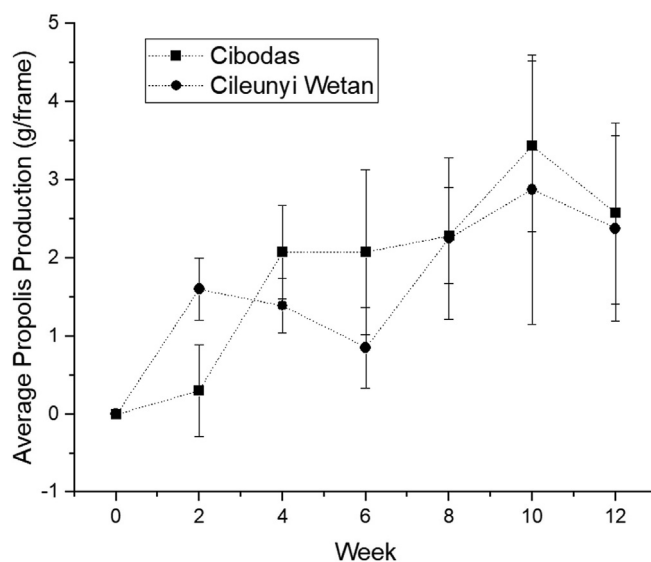


Figure 3. Average productivity of propolis produced by *Tetragonula Laeviceps* cultivated using Modular *Tetragonula* Hives in Cibodas and Cileunyi Wetan.

Indonesia). The reducing sugar content was analyzed according to Luff-Schoorl/SNI 01-2892-1994 method (Badan Standardisasi Nasional, 1994). The antioxidant activity of honey was analyzed by using a 1,1-diphenyl-2-picrylhydrazyl (DPPH) in vitro activity method, and expressed in terms of IC50 of DPPH radical scavenging activity following the procedures suggested by Sadeli (2016). The vitamin C content in the honey was analyzed using a volumetric method as suggested by Silva et al. (2017).

3. Results and discussion

3.1. Production of propolis by *T. laeviceps* cultivated using Modular *Tetragonula* Hives

The average amount of propolis harvested from propolis frames in the MOTIVES at Cibodas and Cileunyi Wetan is shown Figure 3. From the figure, it can be observed that production of propolis tended to increase every week during the three months of cultivation. This indicates that *T. laeviceps* colonies were able to adapt well to the environment and keep producing propolis on the propolis frame. Small holes in the propolis frame stimulated *T. laeviceps* to fill in the holes with propolis within approximately three days to maintain their colonies in a dark condition. The covered propolis frame would also maintain the hive's temperature and protect the hive against external threats, such as microbes, predators and extreme weather (Roubik, 2006; Simone-Finstrom and Spivak, 2010).

The increasing trend in production of propolis, as shown in Figure 3, indicates that the strategy to harvest propolis from the propolis frame every two weeks was able to keep the bees continuously collecting resin to fill in the holes on the propolis frame. According to Nakamura and Seeley (2006), stingless bees, especially the collector bees, can identify the need of their colonies to collect more resin. After propolis was harvested from the frame, penetration of light into the hive triggered the collector bees to collect more resin and fill in the holes with propolis to reduce light exposure to the colony. The increasing trend also suggests that the health of the bee colonies was not affected by the periodic harvesting of propolis (every two weeks) because the *T. laeviceps* had already divided the load to collect resin, pollen and nectar among the collector bees. Hence, periodic harvesting of propolis would not affect the activities of *T. laeviceps* to collect pollen and nectar for the bee colonies (Leonhardt, 2010).

The greatest amount of propolis harvested from the propolis frames was recorded in eek 10, and reached 3.4 ± 1.1 g/frame for bee colonies in

Cibodas and 2.9 ± 1.7 g/frame for bee colonies in Cileunyi Wetan. After eek 4, the average production of propolis in Cibodas was always higher in comparison with Cileunyi Wetan. This indicates that the bee colonies in Cibodas were able to collect more resin to produce more propolis than the colonies in Cileunyi Wetan. The main source of resin for the *T. laeviceps* both in Cibodas and Cileunyi Wetan was from pine trees, whereas the coffee trees provide resin, nectar and pollen for the bees. Other sources of resin, nectar and pollen that were located near the hives are shown in Table 1.

The increased production of propolis after 10 weeks of cultivation indicates an improvement of foraging activity by worker bees that were able to establish a track that could provide more resin with less energy expenditure by leaving a specific chemical trace for other members of the colony (Faheem et al., 2004; Biesmeijer and Slaa, 2004; Simone-Finstrom and Spivak, 2010). In another study by Elitz et al. (2001), it was observed that stingless bees required at least 10 weeks to adapt to a new environment and to collect resources more efficiently. The average production of propolis in Cibodas was higher than in Cileunyi Wetan. This indicates that the environment in Cibodas enabled *T. laeviceps* to collect more resin for producing propolis. Apart from the source of resin surrounding the hives, microclimate conditions also play a crucial role on the daily activities of *T. laeviceps*.

In Table 2, it can be observed that Cileunyi Wetan has an altitude of 720 m above the sea level, and had an annual temperature range of 19–29 °C during 2017, whereas Cibodas has an altitude of 1260 m above the sea level and a temperature range of 19–22 °C. Rainfall in Cibodas during 2017 was also higher than in Cileunyi Wetan. The stingless bee was more active in producing propolis during the rainy season to protect their hives from the heavy rain and to keep their hive warm (Krell, 1996). The microclimate conditions and the availability of resin sources that were closer to the hives promoted greater production of propolis in Cibodas than in Cileunyi Wetan.

In addition to the propolis harvested from the propolis frames, propolis that made up honey pots in the hives was also harvested to determine the total production of propolis. This was then compared with the literature, as shown in Table 3. The total production of propolis in both Cibodas and Cileunyi Wetan lies in the range of 3.09–5.42 g/colony/month as reported by Salatnaya (2012) for cultivation of *T. laeviceps* in Bogor (Indonesia), but is lower when compared with the production of propolis (9.50–15.40 g/colony/month) reported by Agussalim and

Table 1. List of plants surrounding the cultivation of *Tetragonula laeviceps* using Modular *Tetragonula* Hives in Cibodas and Cileunyi Wetan.

| Cileunyi Wetan | | Cibodas | | | | | |
|----------------|--------------|----------------|--------------|----------------|--------------|----------------|--------------|
| | | Location 1 | | Location 2 | | Location 3 | |
| Plant | Distance (m) | Plants | Distance (m) | Plant | Distance (m) | Plant | Distance (m) |
| Arabica coffee | 0.5–1 | Chili | 2.8 | Arabica coffee | 0.8 | Banana | 1 |
| Chili | 2 | Pine | 2.5 | Pine | 1.3 | Arabica coffee | 1.5 |
| Snake fruit | 2 | Banana | 2.5 | Banana | 2.4 | Pine | 3 |
| Rose | 3.2 | Arabica coffee | 5.1 | Eggplant | 3.8 | Cassava | 4 |
| Papaya | 3.45 | Bamboo | 6.7 | Calliandra | 75 | Taro | 7.8 |
| Orange | 4 | Calliandra | 100 | | | Calliandra | 50 |
| Jackfruit | 4.45 | | | | | | |
| Rosemallow | 6.2 | | | | | | |
| Banana | 9.25 | | | | | | |
| Avocado | 10–15 | | | | | | |
| Water apple | 10–15 | | | | | | |
| Suren | 10–15 | | | | | | |
| Djenkol | 10–15 | | | | | | |
| Passion fruit | 10–15 | | | | | | |
| Mango | 10–15 | | | | | | |
| Coconut | 16 | | | | | | |
| Bamboo | 19.5 | | | | | | |
| Pine | >80 | | | | | | |

Table 2. Microclimate conditions in Cibodas and Cileunyi Wetan.

| Parameters | Cileunyi Wetan* | Cibodas** |
|------------------------------|-----------------|-------------|
| Temperature (°C) | 19–29 | 19–22 |
| Humidity (%) | 61.90–84.77 | 61.90–84.77 |
| Rainfall (mm/month) | 3.64 | 14.79 |
| Altitude (m above sea level) | 720 | 1,260 |

*Badan Pusat Statistik Kabupaten Bandung (2017); **Badan Pusat Statistik Kabupaten Bandung Barat (2017).

Erwan (2015) for cultivation of *T. laeviceps* in Lombok (Indonesia). The differences may be caused by different cultivation locations and durations, which in turn affect the availability of resin sources for the *T. laeviceps* to produce propolis.

3.2. Characteristic of propolis produced by *T. laeviceps*

The harvested propolis in Cileunyi Wetan has two different colors, notably red and yellow, whereas the harvested propolis in Cibodas is black, red and yellow. This indicates the different resin sources collected by *T. laeviceps* to fill in the holes of propolis frames in the MOTIVES with propolis. The color change of propolis during the cultivation period could be caused by the change in chemical composition in the propolis since *T. laeviceps* tend to maximize efficiency by collecting resinous materials from a specific location (Bankova et al., 2000; Elitz et al., 2001; Faheem et al., 2004). However, the identification of plant resources responsible for producing specific colors has not been determined in this study.

Characteristics of the crude propolis harvested from both Cibodas and Cileunyi Wetan, particularly flavonoid content, solubility in ethanol and insolubility content of the propolis, are shown in Table 4. As shown in the table, the average flavonoid content of crude propolis obtained from Cibodas (14.8 ± 6.2 mg/g qE) is higher than that from Cileunyi Wetan (11.4 ± 4.4 mg/g qE). These values are higher than the flavonoid content of propolis reported by Salatnaya (2012) of 2.9 mg/g qE, but lower than the values reported by Tagliacollo and Orsi (2011) and Bridi et al. (2015), which were 23.3 ± 6.0 mg/g qE and 78.0 ± 1.0 mg/g qE, respectively. Nevertheless, the flavonoid content of crude propolis that was determined in this study meets the standard flavonoid content of crude propolis in Brazil, namely 5 mg/g qE (Tagliacollo and Orsi, 2011).

A higher flavonoid content in the crude propolis from Cibodas as compared to that from Cileunyi Wetan could possibly be due to the presence of closer resin sources, as detailed in Table 4. This may facilitate *T. laeviceps* in collecting resin sources that can provide the composition of propolis necessary to identify their colonies as well as to protect their colonies from predators (Leonhardt, 2010; Kothai and Jayanthi, 2015). Solubility in ethanol indicates the extractive substances in the crude propolis, while the insolubility content is inferred from the mixture of resin, wax and other insolubilities in ethanol. According to Santos et al. (2003), the percentage of insolubility may reach up to 49.3% of the crude propolis.

3.3. Production of honey by *T. laeviceps* cultivated using Modular Tetragonula Hives

Table 5 shows the amount of honey produced by *T. laeviceps* cultivated using MOTIVES in Cileunyi Wetan and Cibodas. The average amounts of honey for a cultivation period of eight months in Cibodas and

Cileunyi Wetan were 1.44 and 0.93 g/colony/month, respectively. For the cultivation period of August–October 2017, the average amount of honey produced in Cileunyi Wetan (1.13 g/colony/month) was higher than in Cibodas (0.13 g/colony/month). These results are in line with findings by Salatnaya (2012) showing that the amount of honey produced by *T. laeviceps* lies in the range of 0.1–0.8 g/colony/month. These values are lower than the amount of honey produced by *Apis* sp. (Silva et al., 2017). This may be because *T. laeviceps* are smaller in size compared to *Apis* sp and therefore have a shorter flight distance that limits the amount of nectar and pollen that could be collected to produce honey (Faheem et al., 2004).

During the cultivation period of November 2017–March 2018, the average production of honey in Cileunyi Wetan slightly decreased to 0.8 g/colony/month, whereas the average production of honey per month in Cibodas dramatically increased to 2.22 g/month/colony. The change in the production of honey may be caused by the transition phases between the rainy season (August–October 2017) and dry season (November 2017–March 2018, as suggested by Elitz et al. (2001)). The availability of nectar and pollen in both locations during the cultivation period may also cause the difference in the measured amount of honey produced by *T. laeviceps*.

3.4. Characteristics of honey produced by *T. laeviceps* cultivated using Modular Tetragonula Hives

Characteristics of the honey produced by *T. laeviceps* in Cibodas and Cileunyi Wetan are shown in Table 6 and compared with the physico-chemical characteristics of honey produced by *T. laeviceps* in its original location (Subang). The color of the honey varies from clear yellow to cloudy brown depending on cultivation location. The honey tastes sour, as commonly reported in the literature, due to the mixture of honey and pollen and to fermentation during storage inside the honey pots (Deliza and Vit, 2013; Chan et al., 2017). Fermentation occurs because of the high moisture content of honey in the honey pots (Table 6), because *T. laeviceps* requires more time to accomplish the capping process of the honey pots. In the case of honey bees, fermentation of honey occurs due to product decomposition, whereas the fermentation that occurs in the honey pots of *T. laeviceps* is natural and valued by consumers, since it produces a unique sour taste.

Unlike the taste of honey from Cileunyi Wetan and Subang, the honey obtained from Cibodas had a sweet-sour taste. This may be due to fewer pollen sources near the cultivation area in Cibodas. Consequently, less pollen was mixed with the nectar, causing the honey to taste less sour. The sweet-sour taste of honey in Cibodas could also indicate that the honey may not have been stored for a long time. Normally honey that has been stored in a honey pot for a longer period taste sour (Menezes et al., 2013; Kedzierska-Matysek et al., 2016).

During the production of honey by *T. laeviceps* in the MOTIVES, the honey underwent physical change (moisture reduction), biological change (fermentation due to microorganisms inside the bees' bellies and hives) and chemical change (enzymatic degradation of sucrose to glucose and fructose), as highlighted by Menezes et al. (2013). From Table 6, it can be observed that the reducing sugar content of honey from Cileunyi Wetan is higher than that from Cibodas. The most likely explanation is the presence of plants in Cileunyi Wetan that produce more nectar and pollen as compared with the cultivation area in Cibodas and Subang.

Table 3. Productivity of propolis produced by *Tetragonula laeviceps* at different cultivation locations.

| Cultivation location | Propolis productivity from propolis frame (g/colony/month) | Propolis productivity from honey pots (g/colony/month) | Total propolis productivity (g/colony/month) |
|------------------------------------|--|--|--|
| Cileunyi Wetan | 3.80 ± 0.82 | 0.46 | 4.26 ± 0.82 |
| Cibodas | 4.24 ± 1.13 | 0.30 | 4.54 ± 1.13 |
| Bogor (Salatnaya, 2012) | - | - | 3.09–5.42 |
| Lombok (Agussalim and Erwan, 2015) | - | - | 9.50–15.40 |

Table 4. Flavonoid content, solubility and insolubility of crude propolis from different cultivation locations.

| Cultivation location & literature | Flavonoid content (mg/g QE) | Solubility in ethanol (%) | Insolubility (%) |
|-----------------------------------|-----------------------------|---------------------------|------------------|
| Cileunyi Wetan | 11.4 ± 4.4 | 40.8 ± 14.3 | 23.7 ± 5.7 |
| Cibodas | 14.8 ± 6.2 | 69.2 ± 13.3 | 22.9 ± 6.9 |
| Bogor (Salatnaya, 2012) | 2.9 | | |
| Chile (Bridi et al., 2015) | 78.0 ± 1.0 | - | - |
| Brazil (Santos et al., 2003) | 13.7 ± 0.1 | - | - |

Table 5. Productivity of honey produced by *Tetragonula laeviceps* at different cultivation locations.

| Cultivation location | August–October 2017 (g/colony) | November–March 2018 (g/colony) |
|----------------------|--------------------------------|--------------------------------|
| Cileunyi Wetan | 3.4 ± 0.17 | 4.0 ± 0.18 |
| Cibodas | 0.4 ± 0.07 | 11.1 ± 0.35 |

Table 6. Physicochemical characteristics of honey produced by produced by *Tetragonula laeviceps* at different cultivation locations.

| Parameters | Cileunyi Wetan | Cibodas | Subang | Reference* |
|-----------------------------|----------------|-----------------------|--------------|-----------------|
| Color | Cloudy yellow | Clear yellow | Cloudy brown | Amber chocolate |
| Taste | Sour | Sweet (slightly sour) | Sour | - |
| Vitamin C (mg/100 g) | 17.2 | 69.5 | 24.1 | 56–67 |
| Reducing sugars (%) | 77.4 | 73.9 | 44.8 | 55–86 |
| Antioxidant activity (mg/L) | 1341 | 1188 | 1359 | 1370–53800 |
| Moisture content (%) | 14.20 | 20 | 13 | 25.02 |
| Density (g/ml) | 1.29 ± 0.03 | 1.37 ± 0.01 | - | |

* Muruks (2014); Rao et al. (2016); Chan et al. (2017); Silva et al. (2017).

The harvested honey from Cibodas has a slightly higher antioxidant activity (1188 mg/L) in comparison with the honey from Cileunyi Wetan (1341 mg/L) and Subang (1359 mg/L), as determined using a DPPH *in vitro* activity method (IC₅₀). These values are at the high end of the antioxidant activity (1370–53800 mg/L) reported by Silva et al. (2017). The antioxidant activity indicates bioactivity of honey caused by the presence, among others, of vitamin C as well as phenolic and flavonoid compounds in the honey (Vit et al., 2013; Muruks, 2014). The data in Table 6 shows that the vitamin C content of honey from Cibodas is higher than in Cileunyi Wetan and Subang. According to a study by Menezes et al. (2013), fermentative micro-organism in the honey may increase bioactive compounds in the honey and consequently increase the antioxidant activity of the honey.

4. Conclusions

The amount and quality of propolis and honey produced by *T. laeviceps* cultivated using MOTIVES are significantly influenced by cultivation location. *T. laeviceps* that were cultivated in Cibodas had greater production of propolis (4.54 g/colony/month) and honey (1.44 g/colony/month) than *T. laeviceps* that were cultivated in Cileunyi Wetan. The propolis from Cibodas contains a relatively high amount of flavonoid (14.8 mg/g QE), whereas the honey contains 69.6 mg/100 g of vitamin C and an IC₅₀ of 1188 ppm. On the other hand, *T. laeviceps* that were cultivated in Cileunyi Wetan had lower production of propolis (4.26 g/colony/month) and honey (0.93 g/colony/month) as compared with *T. laeviceps* that were cultivated in Cibodas. The propolis from Cibodas contains a flavonoid content of 11.4 mg/g QE whereas the honey contains 17.2 mg/100 g of vitamin C and an IC₅₀ of 1342 ppm.

Declarations

Author contribution statement

Muhammad Yusuf Abduh: Conceived and designed the experiments; Analyzed and interpreted the data; Contributed reagents, materials, analysis tools or data; Wrote the paper.

Abdurrahman Adam: Performed the experiments; Analyzed and interpreted the data.

Muhammad Fadhullah: Analyzed and interpreted the data; Wrote the paper.

Ramadhani Eka Putra, Robert Manurung: Contributed reagents, materials, analysis tools or data.

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Competing interest statement

The authors declare no conflict of interest.

Additional information

No additional information is available for this paper.

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