Melissopalinological characteristic of stingless bee (Trigona/Tetragonula) honey in Lombok, West Nusa Tenggara

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Abstract. These research objectives were to identify the honey type and the potential plants as food source of stingless bees in Lombok, West Nusa Tenggara. Stingless bees honey samples were collected on September 2016 from four different locations in Lombok which are North Lombok, West Lombok, Central Lombok, and East Lombok. The pollen analysis was performed using Von Der Ohe et al. (2004) methods with modification. The result showed that stingless bees honey samples from four locations did not have predominant pollen which classified as multifloral honey. Meanwhile in the honey samples there identified 27 pollen types belonging to 21 families. The pollen spectrums of honey samples were diverse, with the highest dominancy of Arecaceae (Cocos nucifera) in North and Central Lombok and also Tiliaceae (Type Tiliaceae) in West and East Lombok.

Keywords: Melissopalinology, honey type, pollen type, food source

1. Introduction

Stingless bee is social insect that commonly found in the tropical and subtropical regions, they also adaptable to different type of vegetation in forest, savannas, fields, and mountains [1]. These bees are belonging to the order of Hymenoptera, Meliponini tribes, and Apidae family [1]. One genus of stingless bees which is found in Indonesia especially in Lombok is Trigona (Tetragonula) spp. The activity of certain species of bees from this genus varies considerably from morning to evening. Besides being affected by availability of food, this activity is also affected by other factors, such as temperature, humidity, and light intensity from the site [2, 3].

The diet consumed of Trigona (Tetragonula spp) consisted of nectar, pollen, and resin. Nectar as a source of honey raw material later processed by the bees inside its body as one type of food source that continues to be developed. To keep continuance of honey production, pollen of various flower is needed to be known particularly as the sources of nectar for Trigona sp. Honey which is obtained from a different plant can have a different amount, colour, consistencies, qualities, and taste of nectar [4]. It can be traced through the study of melissopalinology. Vijayakumar and Jeyaraaj [5] present four pollen types from Fabaceae and Arecaceae family with coconut, banana, and sunflower pollen are the most constantly found in Nellithurai village.

Melissopalinology can be defined as an applied subdivision of palynology associated with pollen grains studies inside honey and its application in bee keeping [6]. Previous study suggested that both geographical and botanical origin of particular type of honey can be determined by using pollen
spectrum which reflects the floral situation of certain area. Pollen analysis can be used to determine the quality of honey whether honey is defiled or not [7-9]. To encourage stingless bee honey as a profitable product it is mandatory to know the preferred floral and food source of stingless bee. Thus, this research designed to identify the food source of Trigona/Tetragonula spp and the honey type of stingless bee honey in Lombok for honey production.

2. Experimental method

2.1. Material
Stingless bees honey samples were obtained from beekeepers after flower blooming period at four districts in West Nusa Tenggara, Indonesia. North Lombok (KLU), Central Lombok (LT), East Lombok (LTm) and West Lombok (LB) are chosen as sample locations. Collection of honey samples was held in September 2016. The stingless bees honey was harvested directly from hive. The harvested method was used drainage method.

2.2. Honey preparation
The Melissopalinological analysis was carried out according to the method described by Van Der Ohe [10] with modification. Firstly, 0.5–2.5 g stingless bees honey was dissolved in 20 ml distilled water (20–40°C) and centrifuged for 10 min at 1000g. Then, the supernatant liquid was drawn off. The sediment was dispersed again with 20 ml of distilled water and centrifuged for 5 min at 1000g.

Secondly the pollen sediment was place on a glass slide that have been drawn a square with 20 × 20 mm and spread out. After drying, the sediment was covered with glycerine jelly and cover glass.

2.3. Pollen identification
Pollen identification was conducted using microscopically examination with 400× - 1000× magnification. Then, 500 pollen grains were counted. The identification carried out by comparing the shape and measurement of the pollen with pollen reference. Several publications that used are Shubharani [11], Colinvoux [12], dan Roubik and Moreno [13].

After identification and counting, the pollen type of samples was categorised into Louveaux [7] frequency classes: predominant pollen type (>45%), secondary pollen type (16–45%), important minor pollen (3–15%) and minor pollen (<3%).

3. Result
The result of melissopalinological analysis from four honey samples are showed that there are 27 pollen types belonging to 21 families (table 1). The secondary pollen in KLU and LT is Cocos nucifera with relative frequency (FR) 22.6% and 42% respectively. Meanwhile in LTm is Tiliaceae type (18%) and LB is Tiliaceae and Rubiaceae type with FR 41.2% and 16.2% respectively. This difference indicates that every location has a special vegetation character. The vegetation that commonly found in four districts of Lombok is Capsicum sp, Cassia fistula, Casearia guianensis, Carica papaya dan Moringa oleifera.

The photographic plate of honey samples indicated that the honey samples from Lombok have high diversity (figure 1 to 4). This figure confirmation the explanation of the dominating pollen that found in honey samples in each location. Based on literature study the stingless bee food source can be grouping as nectar, pollen, and resin sources (table 2).

All of identified plants in stingless bee honey are consider as pollen source. The vegetation that be nectar sources of Trigona/Tetragonula are Cassia fistula, Dalbergia sp., Mangifera indica, Cocos nucifera, Capsicum sp, Cucumis sativus, Citrus aurantifolia, Helianthus annuus L, Sizygium aqueum, and Carica papaya. While the resin sources are Mangifera indica, Euphorbia pulcherrima, and Manilkara sp.

According to the domination of the plant families, the honey samples in Lombok North (KLU) and Lombok Central (LT) are dominated by Arecales family (Cocos nucifera and Arenga pinnata) with
25% and 45% respectively. *Malvaceae* and *Muntingiaceae* are followed in KLU and *Curcubitaceae* in Lombok Central. Meanwhile honey sample from Lombok East and Lombok West are dominated by *Tiliaceae* with value 18% and 41% respectively. Followed by *Fabaceae* and *Caricaceae* in Lombok East and followed by *Rubiaceae* and *Caricaceae* in Lombok West.

### Table 1. Pollen type in stingless bee’s honey samples.

| Sample | Predominat pollen (>45%) | Secondary pollen (16-45%) | Important minor pollen (3-15%) | Minor pollen (<3%) |
|--------|---------------------------|---------------------------|-------------------------------|-------------------|
| KLU    | -                         | *Cocos nucifera*          | *Mangifera indica,*           | *Cassia fistula,*  |
|        |                           |                           | *Muntingia calabura,*         | *Anacardium*      |
|        |                           |                           | *Citrus aurantifolia,*        | *occidentale,*    |
|        |                           |                           | *Helianthus annuus L,*        | *Capsicum* sp.    |
|        |                           |                           | *Moringa oleifera,*           | *Gnetum gnemon,*  |
|        |                           |                           | *Sisygium aqueum,*            | *Turnera Subulata*|
|        |                           |                           | *Theobroma cacao L,*          | *Casearia guianensis.*|
|        |                           |                           | *Carica papaya*               |                   |
| LT     | -                         | *Cocos nucifera*          | *Arenga pinnata,*             | *Capsicum sp,*    |
|        |                           |                           | *Cucumis sativus,*            | *Amaranthus sp*   |
|        |                           |                           | *Momordica charantia,*        |                   |
|        |                           |                           | *Euphorbia pulcherrima,*      |                   |
|        |                           |                           | *Moringa oleifera,*           |                   |
|        |                           |                           | *Carica papaya,*              |                   |
| LTm    | -                         | Type *Tiliaceae*          | *Lablab sp,*                  | *Cassia fistula,* |
|        |                           |                           | *Capsicum sp,*                | *Dalbergia sp.*   |
|        |                           |                           | *Moringa oleifera,*           | *Theobroma cacao L,|
|        |                           |                           | *Carica papaya,*              | *Casearia guianensis,*|
|        |                           |                           | -                             | *Cyperus sp.*     |
| LB     | -                         | Type *Tiliaceae*, Type    | *Cassia fistula,*             | *Lablab sp,*      |
|        |                           | *Rubiaceae.*              | *Capsicum sp.*,               | *Arenga pinnata,* |
|        |                           |                           | *Gnetum gnemon,*              | *Physalis* angulata,*|
|        |                           |                           | *Amaranthus sp,*              | *Citrus aurantifolia,*|
|        |                           |                           | *Casearia guianensis,*        |                   |
|        |                           |                           | *Carica papaya,*              |                   |

**Figure 1.** Multifloral honey of *Trigona/Tetragonula* (KLU) showing pollen grain of *Cocos nucifera* with 100x magnification.

**Figure 2.** Multifloral honey of *Trigona/Tetragonula* (LT) showing pollen grain of *Cocos nucifera* with 100x magnification.
Figure 3. Multifloral honey of *Trigona/Tetragonula* (LTm) showing pollen grain of *Tiliaceae* type with 400x magnification.

Figure 4. Multifloral honey of *Trigona/Tetragonula* (LB) showing pollen grain of *Tiliaceae* and *Rubiaceae* type with 400x magnification.

Table 2. Nectar, pollen and resin foraging type from identified plants.

| Family          | Pollen type          | Local name    | Food type |
|-----------------|----------------------|---------------|-----------|
| Nectar^1        | Pollen^2             | Resin         |
| Fabaceae        | *Cassia fistula*     | Tengguli      | V         |
|                 | *Lablab sp*          | Komak         | V         |
|                 | *Dalbergia sp.*      | Sonokeling    | v         |
|                 | *Anacardium occidentale* | Jambu mete   | V         |
| Anacardiaceae   | *Mangifera indica*  | Mangga        | v         |
|                 | *Arenga pinnata*     | Aren           | V         |
|                 | *Cocos nucifera*     | Kelapa        | v         |
| Areceae         | *Capsicum sp*        | Cabe           | v         |
| Solanaceae      | *Physalis angulata*  | Ciplukan      | V         |
|                 | *Cucumis sativus*    | Mentimun      | v         |
| Cucurbitaceae   | *Momordica charantia*| Paria         | V         |
| Euphorbiaceae   | *Euphorbia pulcherrima* | Kastuba         | v         |
|                 | *Muntingia calabura* | Kersen        | V         |
| Rutaceae        | *Citrus aurantifolia*| Jeruk nipsis  | v         |
| Gnetaceae       | *Gnetum gnemon*      | Melinjo       | V         |
| Asteraceae      | *Helianthus annuus L*| Bunga matahari| v         |
| Amaranthaceae   | *Amaranthus sp*      | Bayam         | V         |
| Moringaceae     | *Moringa oleifera*   | Kelor         | V         |
| Myrtaceae       | *Sizygium aqueum*    | Jambu air     | V         |
| Malvaceae       | *Theobroma cacao L*  | Kakao         | V         |
| Passifloraceae  | *Turnera Subulata*   | Bunga pukul 9 | V         |
| Salicaceae      | *Casearia guianensis*| Casearia r    | V         |
| Tiliaceae       | *Type Tiliaceae*     | Waru          | V         |
| Caricaceae      | *Carica papaya*      | Pepaya        | V         |
| Rubiaceae       | *Type Rubiaceae*     | Kopi-kopian   | V         |
| Cyperaceae      | *Cyperus sp.*        | Rumput teki   | V         |
| Sapotaceae      | *Manilkara sp.*      | Sawo          | v         |

4. Discussion
Melissopalino logical analysis can show the dominating pollen in honey samples [14], which is the honey samples from Lombok are no predominant pollen. Therefore, the stingless bees honey type is multifloral honey [15, 16]. Multifloral honey has higher diversity of pollen than unifloral honey [17].
Pollen richness of the honey depends on the various factors such as climatic condition, topography and demography largely define the vegetation and floral of the sampling location [18]. North Lombok and West Lombok are dense settlement with higher pollen type, which is 15 and 10 type respectively. Meanwhile Central Lombok and East Lombok are located near protected forest with lower pollen type that is 9 and 8 type respectively.

People in settlement usually grow horticulture crop for their daily need and combine with stingless beekeeping in their farm. Therefore, the pollen richness in North Lombok and West Lombok are higher than other sampling location. Capsicum sp, Carica papaya, Cocos nucifera and Moringa oleifera are the common plants in honey sample of Lombok. This vegetation is a crop that consumed by people of Lombok. Therefore, these crops typically are vastly visited by stingless bee. East Lombok and Central Lombok are sparsely settlement with large farm. People in this area have not utilized their land optimally, thus most part of the farm overgrown with grass. Accordingly, the pollen grain in honey samples of this areas sampling consist of grass and shrub pollen (wild plant pollen). Combination farm as agriculture and beekeeping are common in this area. However due to land use is not optimal the pollen diversity in this area lower in varies.

This study also revealed that Cocos nucifera, Tiliaceae and Rubiaceae is an important plant for stingless bee diet that classified as secondary pollen in Lombok. Bees belonging to Trigona and Apis are good pollinator for coconut trees [19]. The vegetation that identified from honey sample can considered as food source of the stingless bee (Trigona/Tetragonula). This knowledge of the potential plant can enrich the stingless bee food sources and enhances the meliponiculture application. According to table 2, the 27 plant species are a complete diet for stingless bee, which is consist of nectar, pollen and resin source for the stingless beekeeping (meliponiculture) practice. Thirty seven percent of identified plants are nectariferous plants that effectively contribute to honey production.

The melissoplinological characteristics of honey permit us to determine the botanical and geographical origin [20] of honey from Lombok. Furthermore, the knowledge of relationship between the stingless bee and the plant species broaden.

5. Conclusion
The stingless bee honeys from Lombok are multifloral honey due to there is no dominating pollen. The most common pollen types from honey samples in four districts of Lombok are Cocos nucifera, Tiliaceae, and Rubiaceae type as secondary pollen. The plant species that have been identified can be considered as potential food sources of stingless bee (Trigona/Tetragonula).

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