Profiling pH and Moisture Content of Stingless Bee Honey in Closed and Opened Cerumen Honey Pots

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Abstract. The aim of this study is to profile the pH and moisture content of stingless bee honey in closed and opened cerumen honey pots from a single hive of stingless bee. The evaluation was conducted in February and September 2020. Portable pH meter and refractometer were used to measure on-field data of honey pH and moisture content. On-field data measurement revealed that the average pH for closed pots was 3.43 while 3.62 for opened pots. Honey measured in September was found to have higher moisture content with mean value of 32\% compared to honey measured in February with 25\%. High moisture content is a worrisome problem because it will accelerate quality deterioration of stingless bee honey which mostly contributed by fermentation process. Low pH value also influences the consumer preference as the taste of honey will be more sour. So, this study suggests that wetter season in September could give rise to the moisture content of honey due to higher humidity of surrounding contrarily from dry season in February and could affect the quality of stingless bee honey.

1. Introduction
Stingless bee lives in permanent colony consisting queen and foragers who are responsible for pollen and nectar collection \cite{1}. Stingless bee constructs their nest by incorporating their beeswax with plant resins which originated from various botanical sources \cite{2}. Likewise, both honey bee and stingless bee transform the nectar collected into honey which mainly containing mixture of carbohydrates such as glucose and fructose \cite{3}. In addition, honey also consists of enzymes, amino acids, organic acids, minerals and vitamin \cite{4}. Stingless bee honey is commonly known as ‘Kelulut’ in Malaysia and contrarily with honey bee, it has higher water content which also influences various properties of honey. Water is the second biggest component in honey composition and is usually influenced by the botanical origin of the nectar, the handling during the harvesting of the honey and the climatic conditions \cite{5}.
Moreover, type of soil, physiological state of colonies and maturation of honey also influence the composition of honey.

Nowadays, stingless bee honey is gaining more attention worldwide as it is revealed to have therapeutic properties which potentially can help in wound healing, eyes diseases and also can pose as anti-inflammatory agent [6]. The significant different is mainly contributed by the smaller body size of stingless bee which helping them to collect nectar from various nectar sources including small flowers that cannot be penetrated by honey bee. However, the consequences of having smaller body is lesser production of honey annually and making the market price per kg is higher. The stingless bees will transform the nectar collected into honey by using their specific substances before letting it matures during storage inside the cerumen pots [7].

The nest of stingless bee commonly consisting closed pots and opened cerumen pots whereas most of them are used to store honey and some of them also used as pollen and nectar pots as pollen and nectars are stored in separate pots [8]. The stingless bee will fill up each of the open pots until it full before sealing the pots. The open pots normally containing newly collected pollen and the honey is freshly produced by the bees compared to honey inside closed cerumen pots. Interestingly, the difference in properties of honey for both closed and open pots have not yet been widely reported. Hence, in this study, we wanted to know how the close and open cerumen pots itself contribute to the distinctive properties in between stingless bee honey besides one more factor which is the climatic condition.

Malaysia is located in a tropical climate zone whereas rainfall is expected regularly every year. Specifically, Johor has tropical rainforest climate with monsoons rains from November to February from the South China Sea. The mean annual rainfall is 1778 mm with average temperatures ranging between 25.5°C and 27.8°C [9]. Honey is reported to have high hygroscopic properties whereas it is dependent on environmental factor especially during harvesting and storage [10]. To the best to our knowledge, the report on the effect of seasonal changes especially the rainfall on quality of stingless bee honey is still scarce. Hence, the aim of this study is to observe whether the properties of honey in two different months with different rainfall accumulation averages will affect the moisture content and pH of stingless bee honey whereas the measurement was performed directly on field using portable pH meter and refractometer.

2. Materials and methods

2.1. Sample origin
Sample of stingless bee honey from the hive in Universiti Teknologi Malaysia, Skudai, Johor Bahru, Johor, Malaysia (N 1 33.506, E 103 38.302). The evaluation was performed in February and September where February representing dry season with 139.8mm of precipitation and September representing wet season with 190.8 mm of precipitation in Johor Bahru.

2.2. Species
The stingless bee species involved in this study was Heterotrigona itama and only one single hive was employed for data collection. Heterotrigona itama is the most common species to be reared in Malaysia. The hive of the stingless bee was made up of a chopped trunk equipped with a wooden roof box for honey collection and protection from predators.

2.3. Data collection
pH of honey was measured directly on-field using a portable pH meter. The two points calibration using two standard buffer solutions (pH: 7.01:4.01) was performed beforehand prior to pH reading of sample. The probe of pH meter was immersed directly into the selected pots in the hive of stingless bee and was stirred until the glass hour symbol stop blinking. Then, the reading was recorded.

Moisture content (MC) of honey was analyzed using portable digital refractometer. First of all, the digital refractometer was calibrated using a drop of distilled water. Then, three drops of honey were
dropped onto the prism of refractometer. The reading of the refractive index was recorded as R1. MC was calculated using the equation below:

\[ \text{Moisture content (\%)} = \left[ -0.2681 \times \log (R1 - 1) \right] / 0.002243 \]

3. Results and discussion

3.1. pH

For pH, range of data obtained was from 3.24 to 3.78 with the average of 3.73 for closed pots and 3.62 for opened pots as shown in Figure 1. There were no significant different resulted in between close and open pots for pH value measured during February. Similarly, the same outcome resulted for pH measured during September despite of the elevated MC.

pH is the most consistent parameter which displaying the least variability [11]. pH value is commonly influenced by geographical and botanical origin of honey including the type of nectar used during honey production by bees [10]. Besides, the acidity of honey is also related to honey maturation caused by fermentation process as it is expected to increase by days of storage inside the pots [12]. The acidity of honey is also influenced by the activity of glucose-oxidase enzyme which also contributed by bacterial activity during honey maturation [13].

3.2. Moisture content (%)

As presented in Figure 2, MC recorded for open pots were slightly higher compared to close pots. During a nectar flow, collected food is discharged in open pots by the bees before subsequently further dehydrated to lower the MC [14].

![Figure 1](image1.png)  ![Figure 2](image2.png)

**Figure 1.** Graph of pH comparison between stingless bee honey harvested from closed and opened pots  
**Figure 2.** Graph of moisture content comparison between stingless bee honey harvested from closed and opened pots

Therefore, open pots will usually have higher MC’s than the closed pots. However, MC of honey recorded in September showed significantly higher with mean value of 31.88% compared to honey in February with 25.47%. This is because September had higher humidity caused by high rainfall accumulation in contrast to February which explained the elevated MC recorded [15]. Moreover, honey has high hygroscopic characteristics whereas it will easily absorb the humidity from surrounding.
4. Conclusion
In conclusion, there was no significant different of pH between close and open cerumen pots. Different months of measurement which were February and September also did not contribute any inconsistency of pH data collected. However, this study suggests that wetter season in September could give rise to the moisture content of honey due to higher humidity of surrounding contrarily from dry season in February.

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