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The Optimum Storage Conditions on the Quality of the **Stingless bee Honey**

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Abstract. Honey of stingless bee has a higher moisture content than bees. Long-term storage may cause fermentation processes to change the physicochemical properties and taste. Thus, the aim of this research was evaluation of the optimum storage condition on the quality of stingless bee honey. Stingless bee honey (Tetragonular larviceps) was contained in plastic bottle and kept at ambient temperature (30-35°C) and low temperature (4-8°C) for 0-45 days. Before and after storage honey were analyzed the physicochemical properties and sensory investigation for comparison of quality. The results showed that temperatures and storage times have affected on the reducing sugar content, pH, conductivity, color, moisture content with significant difference (p<0.05). While, temperatures and storage times have unaffected on the ^oBrix value (p>0.05). Additionally, the physicochemical properties of honey were according with previously quality report. The sensory investigation indicated that the smell natural flavor, consistency, taste and sourness were not significant difference (p>0.05). In contrast, the color and overall preference were significant difference (p<0.05). For honey quality, Thus, the honey might keep at 4-35°C for \leq 45 day of this study.

1. Introduction

Stingless bee is an insect belonging in Meliponini Tribe, Apidae family. It is without sting and a small insect pollination [1, 2]. The stingless bee honey displayed a variety of biological activities, antimicrobial, anti-cancer, antioxidant, etc. [2, 3]. Nowadays in Thailand, such as Chanthaburi, Chiang Mai, Satun and Chumphon province, there is commercial stingless beekeeping for the sale of honey [4]. The keeping quality of honey for sale is therefore important to consumers that farmers have to consider and it is also related to the value of honey for sale.

In previous reported, the physicochemical properties of various stingless bee strains have been studied. Twenty-eight stingless bee honey samples of 11 stingless bee species from South East Asia (Thailand) were examined physicochemical properties which were an average color (67 \pm 19 mm Pfund), moisture $(31 \pm 5.4 \text{ g}/100 \text{ g})$, ash $(0.531 \pm 0.632 \text{ g}/100 \text{ g})$, electrical conductivity $(1.1 \pm 0.780 \text{ g})$ ms/cm), pH of (3.6 ± 0.198) , total acidity $(164 \pm 162 \text{ meq/kg})$, diastase activity $(1.5 \pm 1.6 \text{ Gothe})$ and hydroxymethylfurfural ($8.7 \pm 12 \text{ mg/kg}$) [5]. Stingless bee honey standard of Malaysian standard

2683:2017 reported quality standard of honey, which indicates the standard range of moisture content with <35 g/100g, ash with 1.0 g/100g and pH with 2.5-3.8 mS/cm [6].

Honey of stingless bee has a higher moisture content than bees. Long-term storage may cause fermentation processes to change the physicochemical properties and quality of honey [7]. Thus, the study of the optimum storage conditions on the quality of the stingless bee honey is of interest. Which may benefit farmers in the distribution of honey.

2. Methodology

2.1 Sample collection and preparation

Honey of stingless bee (*Tetragonular larviceps*) was collected from Pase Yawo sub district, Sai Buri district, Pattani province, Thailand in January 2019. Sample was filtrated and contained into 100 mL transparent plastic bottle (Polyethylene). They were kept at ambient (30-35°C) and low (4-8 °C) temperatures for 0-45 days. Before and after storage honey were analyzed the physicochemical properties and sensory investigation for comparison of quality.

2.2 Physicochemical properties evaluation

2.2.1 Moisture content The empty dish and lid were dried with hot air oven at 105°C for 3 h and transferred to desiccator to cool. The 2-3 g of honey (W1) was place in drying dish. Take it in the hot air oven at 105°C for 3 h. After drying, it was transferred to desiccator to cool. Then, it was weighed (W2) and calculated the moisture content [8]. The moisture content was calculated with the formula (1)

% Moisture content =
$$[(W1-W2) \times 100]/(W1)$$
 (1)

2.2.2 °Brix value °Brix value was determined using ATC portable refractometer. The 1-2 drop of honey place on the measurement prism. Ensure that the honey spreads across the entire surface of the prism. Close the prism cover and holding the instrument under a light source. Looking the °Brix value through the eyepiece. Each sample was measured in in triplicate.

2.2.3 pH value Honey with concentration 10% (w/v) in milli Q water was measured pH value using pH meter (Mettler Toledo). Each sample was measured in in triplicate [9].

2.2.4 *Electrical conductivity value* The 20% (w/v) of honey in milli Q water was analyzed conductivity value using conductometer (Senion 378, HACH). Each sample was measured in in triplicate and expressed in microsiemens/centimeter [5].

2.2.5 Color value The 10 g of honey was dissolved in 20 mL of distilled water. The honey solution was measured absorbance with a UV-Vis spectrophotometer (Jasco V-730) at wave length $\lambda = 635$ nm. Each sample was determined in in triplicate [10].

2.2.6 Reducing sugar content The 1 mL of Honey with concentration 1 mg/mL in distilled water was mixed with 1 mL DNS solution. The solution was boiled for 10 minutes and then cooled rapidly for 5 minutes. After that, 10 ml of distilled water was added. It was measured absorbance with a UV-Vis spectrophotometer (Jasco V-730) at wave length $\lambda = 540$ nm. Each sample was determined in in triplicate. The absorbance was compared with glucose standard curve to calculate the sugar concentration [11].

2.3 Sensory investigation

The 50 persons of untrained panel were evaluated the sensory investigation. They were asked to rate the acceptability of honey. Different attributes viz. color, taste, smell natural flavor, consistency,

sourness and overall preference were rated on the basis of 9-point hedonic scale ranging from 1 to 9 [12].

2.4 Statistics analysis

Analysis of variance ANOVA was performed. For all analysis, p<0.05 were considered significant difference.

3. Results and Discussion

3.1 Influence of storage condition on physicochemical properties

The results showed that the storage times and temperatures have affected on the physicochemical properties of honey (shown in Table 1). The moisture content and conductivity are increased with the storage period, which according to previously studied that moisture was positively correlated with conductivity [13]. In the same way, the values of them at ambient temperature are more than at low temperature which was consistent with the results of Bai et al. [14]. Whereas, color is increased with 7-15 days but it is decreased with 30-45 days, which may relate with precipitate of honey and honey composition [15]. While, "Brix and pH are decreased, which may relate with moisture content and/or chemical composition changing of honey. For the reducing sugar content, the amount of reducing sugar decreased after 15 days, but increased when it was 30-45 days. Prolonged storage of honey test is related to its chemical composition [14, 17]. Likewise, electrical conductivity and pH depend on the amount of minerals, organic acids and proteins in honey.

Generally, the standard values for the conductivity and pH of the honey are at 0.08 mS/cm and 3.71-3.82, respectively. As for sugar and sweetness are the main components of honey, standardized, the total fructose and glucose content generally does not exceed 85 g/100 g, while sweetness ranges from 64.5-75.8 °Brix [14, 18]. For stingless bee honey, it has a higher moisture content than normal bees and the values are in the range of 25-29%. However, the data of this study indicated that physicochemical properties of before and after storage honey were according with previously quality report.

Days	Temp.**	% Moisture	°Brix	Color	Conductivity	pН	% Reducing
		content		(Abs ^{635nm})	(µS/cm)		sugar content
0*	-	25.40±0.10 ^a	73.33±1.00 ^d	0.32±0.01 ^a	1041.67 ± 2.08^{a}	3.82 ± 0.00^{f}	68.12±3.23 ^b
7	А	28.50 ± 0.10^{d}	69.50±1.00 ^{b,c}	0.47 ± 0.00^{e}	1062.00 ± 2.65^{e}	3.71±0.01 ^a	67.08 ± 0.09^{b}
	L	27.20±0.10 ^b	71.00±1.00 ^c	$0.39{\pm}0.00^{d}$	1045.00 ± 5.20^{b}	3.71±0.01 ^a	75.52±0.38 ^d
15	А	29.90±1.00 ^f	68.00±1.00 ^a	$0.52{\pm}0.00^{g}$	1062.00 ± 2.65^{e}	3.71±0.01 ^a	55.71±1.01 ^a
	L	28.50±0.50 ^d	69.00±1.00 ^a	0.48 ± 0.00^{f}	1045.00±5.20 ^b	3.76±0.01°	68.76±1.28 ^b
30	А	28.10±0.10 ^c	70.00±1.00 ^c	0.48 ± 0.02^{f}	1063.00 ± 1.73^{e}	3.73±0.01 ^b	72.84±0.67 ^c
	L	27.70±0.10 ^{b,c}	70.50±1.00 ^c	0.38±0.00 ^c	$1051.00\pm 2.00^{\circ}$	3.73±0.01 ^b	74.76±0.66 ^{c,d}
45	А	29.00±1.00 ^e	69.00±1.00 ^{a,b}	0.47±0.01 ^e	1064.00 ± 2.00^{e}	3.77±0.01 ^d	71.94±0.85 ^c
	L	28.10±0.10 ^c	70.00±1.00 ^c	0.37 ± 0.02^{b}	1057.00 ± 3.46^{d}	3.80±0.01 ^e	73.34±1.08 ^c

Table 1. Physicochemical properties of each storage honey

* = raw honey

** = A: ambient temp.; L: low temp.

^{a-g} = Different letters in the same column indicate significant statistical difference (p<0.05)

3.2 Influence of storage condition on sensory investigation

The result of sensory investigation displayed that the smell natural flavor, taste, consistency and sourness were not significant difference (p>0.05). While, color and overall preference were significant difference (p>0.05) as shown in Table 2. It may be related to the physicochemical properties, pH, ^oBrix and reducing sugar content, that result in the taste no different. Therefore, influence of storage condition might not on sensory investigation.

Days	Temp.**	Color	Smell natural	Taste ^{ns}	Consistency ^{ns}	Sourness ^{ns}	Overall
			flavor ^{ns}				preference
0^*	-	7.77±0.63 ^a	6.73±1.72	7.03±1.03	6.43±1.85	6.13±1.83	7.24±0.95 ^a
7	А	7.40±1.22 ^{a,b}	6.37±1.73	6.67±1.18	6.73±1.44	6.23±1.65	7.10±1.03 ^{a,b}
	L	7.47±1.07 ^{a,b}	6.23±1.87	6.67±1.49	6.63±1.38	6.57±1.48	7.27±1.20 ^a
15	А	6.93±1.20 ^b	6.47±1.41	6.77±1.07	6.50±1.11	6.20±1.35	7.00±0.95 ^{a,b,c}
	L	7.37±1.10 ^{a,b}	6.40±1.57	6.67±0.88	6.73±1.01	6.57±1.04	6.54±1.07 ^{b,c}
30	А	7.20±0.85 ^{a,b}	6.70±0.79	7.00±0.98	6.97±1.03	6.63±1.13	7.20±0.96 ^a
	L	7.47±1.07 ^{a,b}	6.77±1.36	6.63±0.67	6.70±1.02	6.50±0.94	6.90±0.92 ^{a,b,c}
45	А	7.40±1.22 ^{a,b}	6.73±1.20	6.80±0.96	6.63±1.38	6.57±1.22	7.28±0.88 ^a
	L	7.47±1.07 ^{a,b}	6.70±1.34	6.70±0.88	6.57±1.19	6.43±0.94	6.77±0.90 ^{a,b,c}

Table 2. Sensory investigation of each storage honey

* = raw honey

** = A: ambient temp.; L: low temp.

 a^{a-g} = Different letters in the same column indicate significant statistical difference (p<0.05)

4. Conclusion

The aim of this research was evaluation of the optimum storage condition on the quality of stingless bee honey. The results indicated that the stingless bee honey was contained into 100 mL transparent plastic bottle (Polyethylene) can kept at temperature with 4-35 °C for 0-45 days. The quality of honey is also still up to the standard according to previously reported although moisture content was a range of 25.40-29.90%. This research may be useful to entrepreneurs and farmers who will be able to use it in the storage of stingless bee honey for sale.

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