

Effects of Pollen Layer Thickness and Botanical Origin on Color and Moisture Content in Dried Bee Pollen

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ABSTRACT

Bee pollen is used as a substitute ingredient, a fortification material, and a dietary supplement. It contains both macro- and micronutrients such as carbohydrates, proteins, lipids, minerals, and vitamins. In addition, bee pollen has a role as an antioxidant and antibacterial. Its chemical and physical characteristics differ by several factors, particularly its botanical origins. Bee pollen is often incorporated into food products in dried form, which offers a longer shelf life, compared to fresh bee pollen. The drying method is a factor that affects the quality of bee pollen. While previous research has focused primarily on drying temperatures, this study differentiates itself by investigating how the interaction between pollen layer thickness and botanical origin affects color and moisture content. This reveals how different floral species respond to physical heat-and-mass limitations during the drying process. The contribution of this research was to investigate the effect of layer thickness during drying in the container and botanical origin, on the color and moisture content of the dried bee pollen. The research experimental design was a completely randomized design (CRD) with two factors: thickness (0 cm; 1 cm; 1.5 cm) and botanical origins. The experiment consisted of 9 treatments, each executed with 3 repetitions. Data were analyzed using the Analysis of Variance (ANOVA); if there was a significant effect, the Duncan Multiple Range Test (DMRT) was conducted with $\alpha=5\%$. The results showed that the interaction between layer thickness and botanical origins of bee pollen significantly affected a* value, and hue angle. On the contrary, the interaction between the two factors had no significant effect on the moisture content, chroma, and b* value. Dried bee pollen had moisture content of just under 10%. Quantitatively, the applied drying conditions successfully reduced the moisture content below 10%, while maintaining a lightness (L^*) of 31–49, an a* value of 4–16, a b* value of 20–30, a hue angle of 58–82 and a chroma 20–30.

KEYWORDS

Color; Drying characteristics; Layer thickness; Trigona bee pollen; Water content

1. INTRODUCTION

Bee pollen is one of the main bee products, alongside honey. It contains protein, fat [1], amino acids [2], [3], antioxidants [4], [5], and minerals [6]. The mineral content of bee pollen includes magnesium, potassium, sodium, and calcium [7], [8]. The concentrations of these compounds vary depending on botanical and geographical origin. This variability influences its biological activities, particularly its antioxidant and antimicrobial properties, which have attracted considerable interest for nutritional and health applications. One method to preserve its nutritional content and extend the shelf life is by storing bee pollen at refrigerator temperature (6 °C) for up to four weeks [9]. An alternative approach is to store bee pollen in a freezer at -18 °C [10]. Freezing is effective in protecting the bioactive compounds from degradation. However, bee pollen stored under frozen conditions is highly susceptible to microbial growth if the temperature is not consistently maintained [11]. The high-water content in bee pollen makes it a suitable place for microbes to grow, especially molds. Therefore, reducing the moisture content through the drying process is essential to ensure safety and prolong shelf life.

Dried bee pollen is considered safer for human consumption due to its lower risk of microbiological contamination [11]. Some drying methods have been used for bee pollen, including solar drying [12], cabin drying [13], hot air drying [14], [15], and microwave drying [16]. Dry bee pollen produced through solar drying does not exhibit damage or swelling in its structure [13]. In addition, the thickness of the bee pollen layer within the drying container may need to be carefully considered, as it significantly influences the quality of dried bee pollen. Previous studies have applied various drying methods with different layer thickness: oven drying (hot air) with a layer of thickness of 0.8 cm [17], solar drying at 1 cm [13], microwave drying at 0.4 cm [15], and drying using hot air and vacuum drying at 0.5 cm [18]. Furthermore, the drying methods used for lotus bee pollen were by both natural sun drying and hot air drying, both with a pollen layer thickness of approximately 5 mm. Sun-dried lotus pollen exhibited a high concentration of alcohol compounds, which may be attributed to fermentation processes during drying [19]. In the case of rape bee pollen, hot-air drying at temperatures ranging from 40 to 70 °C resulted in a progressive increase in browning index and redness parameter (a^*), accompanied by a decline in yellowness (b) and lightness (L^*) values as the drying temperature increased [17].

Another factor that affects the quality of bee pollen is its botanical origin, which has been shown to influence its protein content [20]. Furthermore, bee pollen from different regions can have different color quality, type of active compounds [21], and quantity of compounds [4], [22], [23]. Several compounds, such as amino acids [24], fatty acid components, carbohydrates, proteins are different in quantity [25], and microbiological contaminants. Such bee pollen can be found in several regions in Turkey [24], India [25]. Other studies also show characteristics regarding colors and components from Fourier Transform Infrared Spectroscopy (FTIR) results from several regions in the Slovak Republic [26]. These different compositions have an impact on the different colors of bee pollen. Bee pollen color is influenced by its carotenoid and natural pigment content. Lutein and zeaxanthin degrade during drying. High temperatures increase the Maillard reaction and the formation of 5-HMF, resulting in a browner color. Botanical origin also influences color, determining the type and amount of pigments, thus varying the color during drying [17]. In addition, color changes in dried bee pollen are influenced by the Maillard reaction and enzymatic browning [27]. Bee pollen dried with hot air drying with a thickness of 4 mm combined with a temperature of 40 °C, produced the lowest total color change (ΔE) value. When the temperature was increased in the layer, there was a decrease in the L^* (brightness) and b^* (yellowness) values, which means the bee pollen became darker due to caramelization due to uneven heat exposure if the layer was too thick [14]. In previous studies, bee pollen from Viracocha, Boyacá, Colombia was dried using both solar and cabin dryers. Solar drying involved lower temperatures than cabin drying, and the resulting bee pollen structure remained unchanged [13]. Therefore, this study used the YSD-UNIB18 dryer to dry bee pollen from Bengkulu, Indonesia. The YSD-UNIB18 dryer is a hybrid dryer that can be used with solar energy, biomass fuels, or a combination of both [28], [29]. This research contributed to investigate the effect of layer thickness and botanical origin on moisture content and color parameters (lightness, a^* value, b^* value, chroma, and hue angle). Moisture content and color need to be tested because moisture content is a key determinant of bee pollen's microbiological stability and shelf life, preventing mold damage during storage. Meanwhile, color serves as a primary visual indicator for consumer acceptance, reflecting the authenticity of its botanical origin and the retention rate of bioactive compounds that are sensitive to thermal degradation.

2. MATERIALS AND METHODS

2.1. Materials

The materials used in this study were bee pollen from 3 regions in Bengkulu: Kepahiang (bordering Central Bengkulu), Kepahiang-Empat Lawang (bordering Empat Lawang), and Bengkulu city. All three types of bee pollen were obtained from Trigona bees. Bee pollen from the Kepahiang, which border Central Bengkulu, is predominantly composed of coffee flower pollen. Meanwhile, bee pollen from Kepahiang near the Empat Lawang region contains a mix of pollen from coffee flowers, durian (*Durio zibethinus*), rubber trees (*Hevea brasiliensis*), and calliandra flowers (*Calliandra* sp.). Bee pollen from Bengkulu originates from *Antigonon leptopus*, santos flowers (*Xanthostemon chrysanthus*), porana flowers (*Porana* sp.), sunflower (*Helianthus annuus* L.), and lavender (*Lavandula angustifolia*). The tools used in this

research were YSD-UNIB18 solar dryer, Linshang LS171 colorimeter, aluminium foil container (top dimensions 7×7 cm; bottom dimensions 5×5 cm; height 2.1 cm), and digital caliper. Pollen layer thickness illustration presented in Figure 1.

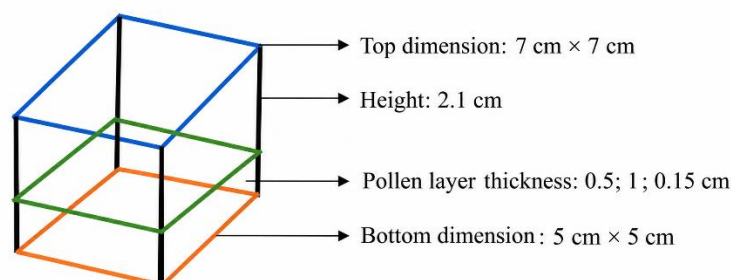


Figure 1. Pollen layer thickness illustration.

2.2. Research Methods

This study used a completely randomized design with a factorial pattern consisting of two treatment factors. The first factor was botanical origin, consisting of 3 origins (Bengkulu, Kepahiang, and Central Bengkulu), and the second factor was pollen layer thickness, consisting of 0.5, 1 cm, and 1.5 cm. All treatment combinations were repeated 3 times (triplicate), resulting in a total of 27 experimental units. Fresh bee pollen from each region was placed into containers. The height or thickness of the bee pollen layer was then adjusted according to the treatment, namely 0.5 [15], [18], 1 [13], [17], and 1.5 cm. The containers were then placed in the rack of the YSD-UNIB18 dryer. Drying was stopped when the moisture content reached less than 10% with the time ranging from 18 to 28 hours. A greater pile thickness of bee pollen results in a longer drying time. Water content was obtained by calculating the weight of the sample per hour and the initial water content of the sample. Recorded drying temperatures varied between 30.7 to 50.7 °C.

2.3. Moisture Content Analysis

The water content test was conducted using thermogravimetry method. A total of 3 g sample was placed in a crucible and dried in an oven at 105 °C for 24 hours. After drying, the crucible was cooled in a desiccator to room temperature before weighing. The moisture content was expressed as wet basis (w.b) and calculated from the difference in sample weight before and after drying, divided by the initial sample weight.

2.4. Color Analysis

The color test used in this study was use colorimetry method [17]. Both fresh and dried bee pollen samples were tested for color in the form of L, a, and b values. These values were obtained from the output of the colorimeter tool. Furthermore, the hue angle and chroma were calculated according to the equation (1) and (2) [30].

$$\text{Hue angle } (^{\circ}) = \tan^{-1} \left(\frac{b}{a} \right) \times 57,3^{\circ} \quad (1)$$

$$\text{Chroma} = \sqrt{b^2 + a^2} \quad (2)$$

2.5. Data Analysis

The collected data were analyzed using analysis of variance (ANOVA). When a significant effect was detected, further comparisons among means were conducted using Duncan's Multiple Range Test (DMRT) at a significance level of $\alpha = 0.05$.

3. RESULTS AND DISCUSSION

3.1. Characteristics of Fresh Bee Pollen

Fresh bee pollen from Bengkulu, Kepahiang-Empat Lawang, and Kepahiang exhibited moisture content above 29%, 30%, and 32%, respectively (Table 1). These values are higher than those reported from Transylvania, which ranged from 17.59% to 29.55%. However, one sample of bee pollen from that area had a moisture content of about 36%. In addition, the lightness value of the three types of bee pollen ranged between 39 and 49, which are lower than the values recorded for fresh Turkish bee pollen with above 52 [18].

The results of this study indicated that fresh bee pollen is darker than Turkish bee pollen. The three types of fresh bee pollen from Bengkulu had a slightly higher value than Turkish bee pollen [11]. The b^* value obtained in this study was below 2, which is lower than the b value of Turkish bee pollen reported by [18], reaching 41.20. This indicates that Bengkulu bee pollen is considerably less yellow in color, compared to fresh Turkey bee pollen.

Table 1. Characteristics of fresh bee pollen.

Bee pollen	Moisture content	Lightness	a^*	b^*	Hue angle ($^\circ$)	Chroma
Kepahiang	32.56 ± 0.75	47.46 ± 0.61	10.95 ± 0.61	24.87 ± 0.42	65.88 ± 0.81	26.78 ± 0.63
Kepahiang-Empat Lawang	30.19 ± 0.71	40.18 ± 1.83	13.07 ± 1.52	25.95 ± 1.21	63.23 ± 3.70	29.09 ± 0.50
Bengkulu	29.93 ± 1.83	39.14 ± 3.11	10.97 ± 8.08	24.23 ± 1.78	66.53 ± 17.23	27.39 ± 2.11

3.2. Moisture Content of Dried Bee Pollen

All three types of bee pollen, when dried with different layer thicknesses, had moisture content below 10% (Table 2). The interaction between layer thickness and botanical origin had no significant effect on moisture content ($p > 0.05$). Bee pollen was dried from 18 to 28 hours, depending on thickness of the layer. The insignificant effect of layer thickness on the final moisture content was primarily due to all treatments being dried to the same target threshold of 10%. A layer thickness of 0.5 cm showed a shorter drying time, approximately 18 hours. Conversely, a layer thickness of 1.5 cm required a longer drying time, approximately 28 hours. Sun-dried lotus bee pollen has the moisture content of 6% after 18 hours of drying [19]. Another study reported that microwave-dried bee pollen had moisture content of approximately 7% [15].

Table 2. Moisture content of dried bee pollen.

Treatment	Moisture content (%)
K1A1	9.13 ± 0.67
K1A2	8.27 ± 0.55
K1A3	8.43 ± 0.81
K2A1	9.24 ± 0.03
K2A2	9.13 ± 1.11
K2A3	9.05 ± 1.17
K3A1	9.87 ± 0.06
K3A2	8.78 ± 1.01
K3A3	8.98 ± 0.38

Note: K1 (thickness 0.5 cm); K2 (thickness 1 cm); K3 (thickness 1.5 cm); A1 (Kepahiang); A2 (Kepahiang-Empat Lawang); and A3 (Bengkulu).

3.3. Lightness of Dried Bee Pollen

Lightness indicates the brightness of a sample. The interaction between layer thickness and botanical origin of bee pollen was not significant ($p > 0.05$). However, the layer thickness during the drying process had a significant effect on lightness ($p < 0.05$). Figure 2 shows that the lightness value of Kepahiang bee

pollen demonstrated the highest value compared to bee pollen from Bengkulu and Kepahiang-Empat Lawang. The bee pollen samples, predominantly derived from coffee blooms (Kepahiang-Bengkulu Tengah, and Kepahiang-Empat Lawang), had lower lightness values than coffee flower (robusta) bee pollen (62.37) from Daklak province, Vietnam [31].

The result of this study showed that Kepahiang bee pollen exhibited higher lightness than Bengkulu and Kepahiang-Empat Lawang bee pollen, with a lightness value around 51. Similarly, the lightness value of fresh Turkish bee pollen was around 52 [18]. Several factors influence the lightness value of bee pollen, including the drying process; for instance, the equipment and temperature used during drying can decrease the lightness [18]. Additionally, the layer thickness may affect the lightness value due to variations in bee pollen temperature between thicknesses of 0.5; 1; and 1.5 cm.

The lightness value observed in this study averaged below 48 (Figure 2). Bee pollen with a lightness value around 48 is categorized as dark-colored. Plant species known to contribute to darker hues include berberis, lupin, red deadnettle, blackthorn, pear/crab apple, white horse chestnut, fake acacia, rose bay willow herb, sycamore, red/white clover, gooseberry, and red horse chestnut [23]. Conversely, a lightness value of approximately 61.69 indicated a pale hue.

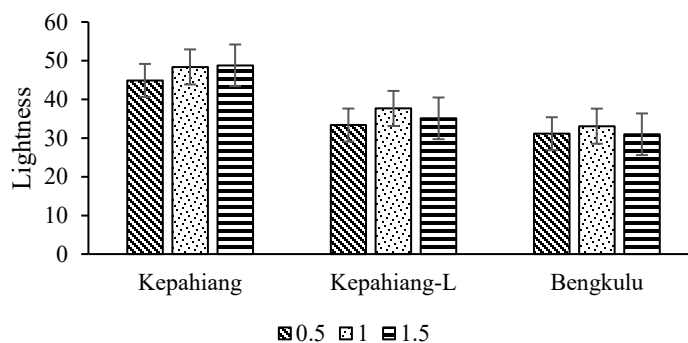


Figure 2. Lightness value of dried bee pollen.

3.4. a^* value of Dried Bee Pollen

The interaction between layer thickness and bee pollen origin had a significant effect on a^* values ($p < 0.05$). The a -values of fresh and dried bee pollen from Bengkulu and Kepahiang-Empat Lawang were similar, ranging from 9-12 and 8-11, respectively (Figure 3). Comparable a^* values were also reported for Turkish bee pollen in previous research [18].

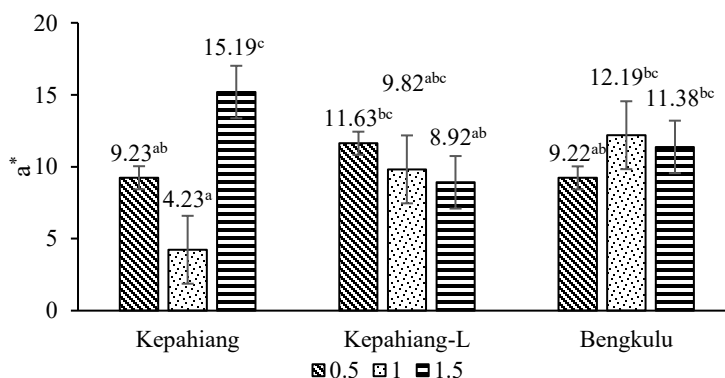


Figure 3. a^* value of dried bee pollen.

The a^* values show red or green. The color of bee pollen is influenced by the type of flowers consumed by bees [21]. There are yellow, green, red-brown, brown, brownish yellow, and red-brown [26].

The bee pollen from Daklak Province is exclusively derived from coffee blooms (mono floral) [31]. In contrast, bee pollen from Kepahiang consists of pollen gathered from various floral sources (multifloral), including coffee flowers.

3.5. b^* value

The b^* value of bee pollen was tested on both fresh (early) and dried (final) states. The interaction between layer thickness and botanical origin was not statistically significant ($p>0.05$). Figure 4 shows that the b^* values of Kepahiang, Bengkulu and Kepahiang-Empat Lawang were at a layer thickness of 0.5-1 cm, ranging from 20 to 30. Bee pollen sourced from coffee flowers exhibited a slightly higher b^* value (35.04) compared to Kepahiang bee pollen, which predominantly comes from coffee flowers [31]. Both types of bee pollen were dried using solar exposure and have the moisture content below 10%. In this study, the b^* values in all bee pollen results showed lower values, compared to Turkey bee pollen, which ranged from 33 to 45. The b^* value showed blue and yellow axis. When comparing the results of this study with the other [18], which used Turkish bee pollen with a more intense yellow hue. The difference in b values is likely caused by differences in the composition of the flower's pollen, which ultimately results in differences in the pigments that affect color.

Carotenoids are one of the pigments that influence the yellow color of bee pollen. Carotenoids, such as lutein and zeaxanthin, are degraded during drying [17]. Zeaxanthin is a rare carotenoid in foods, unlike lutein and beta-carotene, which are commonly found in several foods. Nevertheless, this type of carotenoid is contained in bee pollen [32]. Furthermore, the Maillard reaction also contributes to the formation of dark brown color. This reaction occurs due to the presence of proteins and reducing sugars that are exposed to heat during drying [17]. Bee pollen contains protein (10-40%), carbohydrates (13-55%), which contain reducing sugars [33].

Bee pollen from Bengkulu exhibited lower b^* values at each layer thickness during the drying process in comparison to other bee pollens (Figure 4). Another source of bee pollen from Bengkulu was sunflowers. Sunflowers serve as a nutritional resource for bees with the pollen containing approximately 15% protein [34], [35]. The protein content has been suggested to influence the color of dried bee pollen [36]; however, the color alteration remains minimal due to the drying process being conducted at a relatively low temperature.

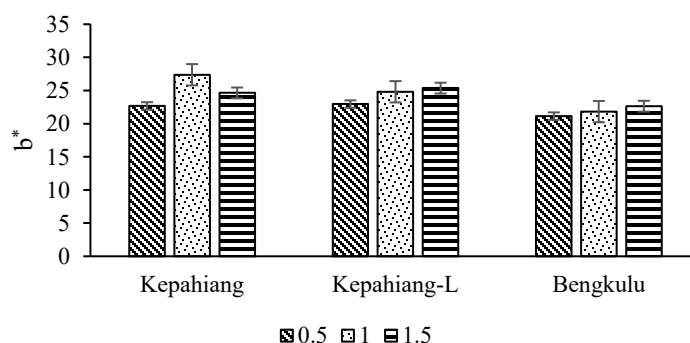


Figure 4. b^* value dried bee pollen.

3.6. Hue angle

The hue angle of bee pollen was tested on both in its fresh bee pollen (initial) and after drying (final). The interaction between layer thickness and botanical origin showed a significant effect on hue angle ($p<0.05$). The hue angle was influenced by the presence of pigments such as carotenoids and phenolic compounds, which were responsible for its characteristic colors ranging from orange to red [37]. Furthermore, the botanical origin of bee pollen significantly affected the hue angle due to the varying

pigment composition of different plant species. This variation can be used to classify and differentiate pollen samples based on their source [37].

According to Figure 5, the mean hue angle of Kepahiang bee pollen was higher than that of Bengkulu and Kepahiang-Empat Lawang bee pollen. The hue angle of the three types of bee pollen ranged from 60 to 81°. This falls within the broader hue range of 54 to 90°, which corresponds to yellow-red hues [25]. In addition, the hue of dry bee pollen from Tirli, Grosseto, Italy was around 88° [39].

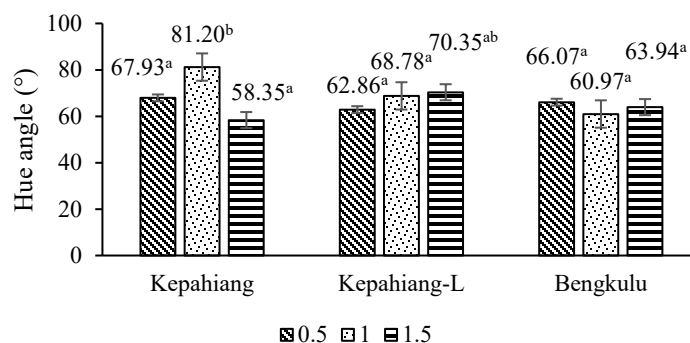


Figure 5. Hue angle of bee pollen after drying.

3.7. Chroma

Chroma represents the color intensity or degree of saturation, excluding brightness [40]. A higher chroma value indicates a greater color intensity in the sample. As shown in Figure 6, chroma values range from 23–31. The chroma values in Kepahiang and Kepahiang-Empat Lawang bee pollen were nearly identical. However, the chroma value of dried Bengkulu bee pollen was lower, meaning that reduced color intensity compared to bee pollen from other regions. Neither drying layer thickness nor botanical origin significantly affected chroma, indicating that color saturation remained relatively stable under the applied drying conditions. These findings suggested that the natural pigments of bee pollen, particularly carotenoids, were largely preserved during drying, while oxidative reactions and browning remained minimal. Similar color stability was observed during drying of rape bee pollen [17].

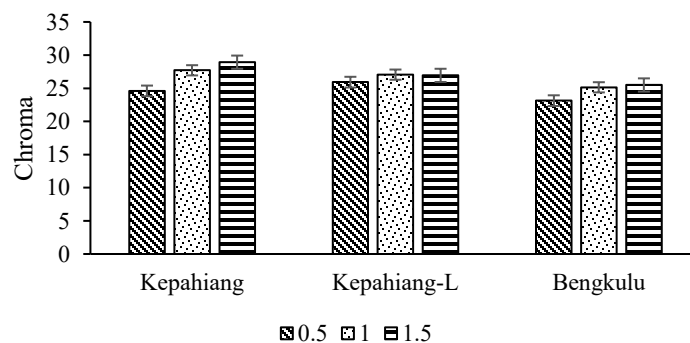


Figure 6. Chroma bee pollen after drying.

4. CONCLUSION

The findings demonstrate that both layer thickness and the botanical origin of bee pollen influence important color characteristics, particularly hue angle and a value, indicating that these factors should be considered when optimizing drying conditions and maintaining product quality. In contrast, the absence of interaction effects on lightness, b value, chroma, and moisture content suggests that these quality attributes are relatively stable across the combinations of the two factors evaluated. These results provide useful

information for improving bee pollen processing and quality standardization. Future studies should investigate a wider range of drying conditions, additional botanical origins, and the effects of processing on the nutritional, bioactive, and sensory properties of bee pollen to support its broader industrial application.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

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