

## THREE BEHAVIORS OF *Apis cerana* (F.) IN RESPONSE TO SUPPLEMENTARY FEED OF PULP AND PASTE OF TONGKA LANGIT BANANA

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### ABSTRACT

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*Apis cerana* (F.) is a honeybee commonly cultivated by beekeepers in Indonesia to produce honey. The use of additional feed acts as a controller of honeybee colonies so that they do not migrate, especially in uncertain weather conditions. Providing additional feed based on tongka langit bananas which have a high  $\beta$ -carotene nutritional content influences the adaptation, eating, and resting behavior of *Apis cerana*. This study describes and analyzes the adaptation, eating, and resting behavior of *Apis cerana* to the provision of additional feed based on tongka langit bananas *Musa troglodytarum* (L). The study was conducted for 10 days with the provision of treatment feeds, namely K + (sugar water), P1 (fresh banana fruit), and P2 (banana paste) on the first day and data collection was carried out the next day with a total of 5 times data collection on the number of individuals. Data on the number of individuals on three behavioral parameters (adaptation, eating, and resting) will be subjected to a two-way analysis of variance and continued with Duncan's test. In the treatments given (K+, P1, and P2) it was found that P2 had the largest number of individuals in adaptation behavior, in eating behaviours it showed that K+ had the highest number of individuals, but it was not significantly different from P2 after analysis so that P2 had good potential as additional feed for *Apis cerana* with high  $\beta$ -carotene content.

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## INTRODUCTION

The honeybee *Apis cerana* Fabricius, 1793 is also known as the Asian honeybee because of its population distribution in the Asian continent. This honeybee has very similar similarities to the European honeybee (*Apis mellifera*), so *Apis cerana* (F.) is also known as the cousin of *Apis mellifera*. The honeybee *Apis cerana* has a vital role in maintaining the environmental ecosystem by contributing to plant pollination. This contribution to plant pollination will directly affect agricultural productivity and environmental sustainability. In addition, the important thing about honeybees is the production of honey as a food that has various nutrients and health benefits. Therefore, an effective maintenance strategy to support the productivity of honeybee colonies is important. The population of *Apis cerana* (F.) honeybees has decreased significantly, especially in Indonesia. Honeybee maintenance is faced with several challenges, including fluctuations in the availability of natural food and rapid environmental changes. Bee colonies as social creatures require adequate nutritional intake to carry out their critical functions in the ecosystem. Providing additional feed is a relevant and potential alternative to improve the condition of honeybee colonies. Tongka langit banana is chosen as an additional feed ingredient because of its abundant availability and its nutritional potential that can support bee productivity. Additional feed based on tongka langit banana is an unusual food for bees. This additional feed will provide a factor in the behavior of bees outside the hive that has a direct effect on worker bees.

The behavior of worker bees in terms of adaptation, feeding, and resting greatly affects the survival and development of the colony. Knowledge about how worker bees respond to additional feed based on sky banana is still very limited. Therefore, this study is important to fill the existing knowledge gap by providing a better understanding of the adaptation, feeding, and resting behavior of *Apis cerana* to the provision of additional feed based on sky banana fruit. In this case, to determine the effect of providing additional feed based on sky banana on the activity behavior of *Apis cerana* needs to be studied. The information generated from this study is expected to provide insight into the patterns of adaptation, feeding, and resting activities of *Apis cerana* in responding to the provision of additional feed based on sky banana. This study used the long fruit type of sky banana as additional feed because the long fruit type has higher  $\beta$ -carotene and has a stable stock compared to other types, namely short fruit bananas. Based on the results of the analysis using the High-Performance Liquid Chromatography (HPLC) method, the  $\beta$ -carotene content in long fruit flesh is 65.33 mg/100g and short fruit 22.31 mg/100g (Letelay et. al., 2020). HPLC is an analytical method that separates a mixture into single components through the interaction of compounds, liquid mobile phases and inert stationary phases. The interaction between the mobile phase and the stationary phase varies and depends on the type of compound being separated. HPLC is very efficient for separating molecules into short compounds. The result of the data analysis system is a chromatogram (Bayne & Carlin, 2010).

Experiments on the use of additional bee feed have been conducted previously in various previous studies, for example, the provision of Coconut Sugar as additional feed (Fauzie, 2017). The use of tempeh as a substitute for pollen, for example, the provision of soybean tempeh pastes as a substitute feed (Noviyanti, 2018). The provision of green bean tempeh as a substitute pollen (Sitorang, 2018). The provision of red bean pastes as a substitute pollen (Pratama, 2018). Research on the behavior of honeybee activity towards the provision of additional feed is still very little found. One study is regarding the effect of providing additional feed on the activity of worker bees carrying nectar and the area of honeycombs in honeybees (*Apis mellifera*) approaching the flowering season (Imaningtyas, Minarti, & Cholis, 2014). Meanwhile, research on the effect of providing additional feed based on tongka langit bananas on the behavior of honeybee activity has never been conducted. This research is expected to formulate alternatives for more productive honeybee maintenance which overall will also support sustainable agriculture. The provision of additional feed based on tongka langit bananas to *Apis cerana* is also expected to increase the productivity of honeybee colonies and produce honey with higher  $\beta$ -carotene content. The use of tongka langit bananas as additional feed will also increase the utilization of tongka langit bananas, which are less popular with the public because there are types of tongka langit bananas that have a bitter taste. Thus, this study has a broad and relevant impact in the context of agricultural sustainability, *Apis cerana* maintenance, and the utilization of tongka langit bananas.

## MATERIALS AND METHOD

### Materials

The equipment used, namely 9 honeybee colony boxes (hives), combs, pieces of ribs, adhesive, stationery, rulers (cm), cameras, measuring cups, transparent plastic feed containers, digital scales with an accuracy of 0.1 g, and masks. Tools for making additional feed, namely knives, tablespoons, stirrers, and large trays, stoves, and

large trays. The materials used in the study, namely nectar, water, Gulaku brand granulated sugar, and ripe tongka langit banana flesh.

## Work Procedures

### Research Preparation

#### a. Preparation of *Apis cerana* (F.) bee colonies

Preparation of honeybee colonies is carried out from the following 11 stages (1) the size of the hive for *Apis cerana* is generally length x width x height as follows: the hive lid is 43 cm x 39 cm x 5 cm. The hive box is 41 cm x 31 cm x 23 cm. The thickness of the board used is 2.0 cm; (2) hive supports to avoid insect attacks such as termites, ants, and other animals, the height of the hive support legs ranges from 40 cm–50 cm from the ground; (3) on the inside of the hive there are 5–9 combs or frames with the length of the upper part with protrusions being 39 cm, the length of the lower part 30 cm, the height 20 cm, the thickness of the hanging wood 2.5 cm, the thickness of the reinforcing wood 1.5 cm, and the width 2.0 cm for making a hexagonal honey bee nest; (4) the distance between one comb and another is around 0.5 cm so that the honey bees can move freely; (5) this study used 9 colony hives that were weighed first; (6) the colonies are active and have good additional feed consumption capacity; (7) the bee colonies are in good physical condition; (8) the initial population of each colony is assumed to be the same; (9) three combs full of bees are assumed to be the same; (10) there are 2 active combs and 7 empty combs and 1 bee food container; (11) giving sample sequence numbers to all samples that are placed randomly and then given sequence numbers. Labeling is intended to facilitate data collection.

#### b. Making feed and treatment

The treatment feed used was (1) sugar solution as K+ (10 g water + 15 g sugar that has been stirred evenly); (2) banana flesh as P1 (25 g fresh ripe tongka langit banana flesh that has been stirred evenly into porridge); and (3) banana paste as P2 (10 g banana flesh and 15 g sugar that has been stirred evenly into porridge). Additional feed was given to each colony as much as 25 g for 10 days with 5 observations, feeding was carried out at 07.00-07.15 WIT. This time was chosen to ensure that the bees were more receptive to changes in the environment, including new food sources, because they had just started their activities. The feed was in a transparent plastic container and placed at the front of the hive box not far from the bee entrance and exit gate. In the feed container, pieces of sticks are provided as a bridge for *Apis cerana*. The research was conducted from April 17, 2023, to April 26, 2023. The observation was located at a honeybee cultivation site in Eri Village, Nusaniwe District, Ambon City, Maluku.

## Methods

Data collection on bee behavior in adaptation, eating, and resting activities was carried out every two days for 5 observations at 09.00-09.15 WIT. The data collected were the number of worker bees around the feed container area and those inside the feed container in each honeybee colony in each hive, totaling 9 boxes. Observations on the 1st day were carried out the day after treatment was given to the bee colony. Observations on the adaptation, eating, and resting behavior of *Apis cerana* honey bees were carried out by (1) assessing adaptation behavior from bees outside the feed container on the 1st day; (2) assessing feeding behavior from bees in the feed container and seen carrying out feeding activities; (3) resting behavior was assessed from bees in the feed container but not carrying out feeding activities and bees outside the container on days 3, 5, 7, and 9. The rest in question is rest that classifies behavior that does not fall into the adaptation and feeding categories. This study used quantitative data analysis techniques.

The parameters used were the adaptation, feeding, and resting behavior of *Apis cerana* honey bees. The three sources of diversity used were feed treatment, day effect, and feed-day interaction. The relative magnitude of the three indicates whether the differences in observations between treatments were real or due to chance. The difference in treatment is said to be real if the treatment diversity is large enough compared to the experimental error. The source of diversity can be said to be real if the F probability is less than 5%.

## RESULTS AND DISCUSSION

### 1. Results of observations behavior *Apis cerana* worker bees in response to the provision of additional feed based on sky banana fruit.

The results of observations behavior *Apis cerana* worker bees for adaptation, eating, and resting behaviors to the provision of additional feed based on tongka langit banana fruit showed that the total average number of individuals per day in each treatment, in resting behavior had the largest number followed by eating behavior, and adaptation behavior had the smallest number of individuals. In treatments (K+, P1, and P2) the number of *Apis cerana* worker bees was the highest in treatment 2 (P2) with the provision of tongka langit banana fruit paste. The results of the observations can be seen in the following table 1.

Table 1. Average number of *Apis cerana* worker bees per day for three types of behavior on food treated with sugar water (K+), fresh fruit flesh (P1), and banana paste (P2).

Behavior	Behavior														
	Adaptation					consume					Rest				
	H1	H3	H5	H7	H9	H1	H3	H5	H7	H9	H1	H3	H5	H7	H9
K+	5.3	0	0	0	0	1.3	8	10	8.7	11.3	4	3.7	6.7	7	26.3
P1	4	0	0	0	0	2	3.3	2	2.3	2.6	6.7	7	7	3.3	7.7
P2	11	0	0	0	0	5	9.3	4	6.3	10.3	27.7	27	6	11.3	18.7

Note: H1 - H9 are observations on days 1, 3, 5, 7, and 9, respectively.

### 2. Adaptation behavior of *Apis cerana* worker bees to additional feed of meat and Tongka Langit banana paste.

The results of observations of treatment on adaptation behavior showed that the adaptation process to feed only occurred on the first day in the three treatments. On the first day of the adaptation process, the highest average number of individuals was treatment 2 (P2) which was 11 individuals, followed by positive control (K+), and treatment 1 (P1) respectively as many as 5 and 4 individuals. The results are presented in Figure 1.

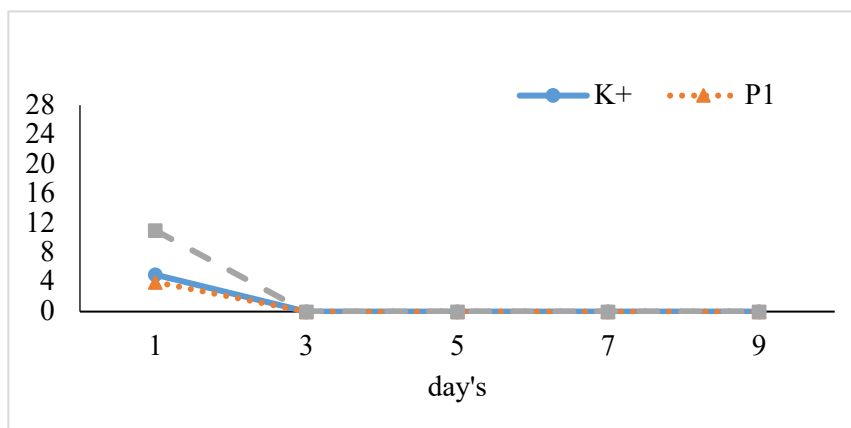


Figure 1. Number of worker bees per day in each treatment (K+, P1, and P2) for adaptation behavior.

The results of the analysis of variance on adaptation behavior indicate an influence on the interaction of treatment and day which shows a probability value F countless than  $\alpha = 0.05$ . The results of this analysis also show that there are differences in each level of diversity sources for treatment and day. However, the analysis of interaction and treatment has the highest priority, which is then continued with Duncan test.

Table 2. Results of two-way analysis of variance for adaptation data.

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Treatment	2	16.58	8.29	4.24	0.0239
Days	4	330.76	82.69	42.28	0.0001
treatment*days	8	66.31	8.289	4.24	0.0017
Error	30	58.67	1.96		
Corrected Total	44	472.31			

The results of the Duncan test showed that treatment 2 (P2) had the highest number of individuals on the first day, as many as 11 individuals who carried out adaptation activities, which were significantly different from the positive control and treatment 1 (P1), in this case different superscript letters indicate inequality. If comparing the number of individuals on the first day between K + and P1, it was found that K + and P1 were not different. Observations on days 3 to 9, no individual bees were found to have adapted.

Table 3. Duncan test adaptation data

Treatment	H1	H3	H5	H7	H9
K+	5.3 ± 2.5 <sup>b</sup>	0 <sup>c</sup>	0 <sup>c</sup>	0 <sup>c</sup>	0 <sup>c</sup>
P1	4 ± 2 <sup>b</sup>	0 <sup>c</sup>	0 <sup>c</sup>	0 <sup>c</sup>	0 <sup>c</sup>
P2	11 ± 4.4 <sup>a</sup>	0 <sup>c</sup>	0 <sup>c</sup>	0 <sup>c</sup>	0 <sup>c</sup>

Note: Mean values followed by the same letter indicate insignificant values, at  $\alpha = 0.05$ .

### 3. Feeding behavior of Apis cerana worker bees on supplementary feeds of meat and Tongka Langit banana paste.

The results of observations of treatment on eating activity showed that the positive control had the highest number of individuals on the 9th day, namely 12 individuals, followed by the 7th day, the 5th day and the 3rd day respectively as many as 9 individuals, 10 individuals and 8 individuals, then on the first day as many as 1 individual. Treatment 1 had a relatively static number of individuals, namely on the 3rd and 9th days had the same number of individuals, namely 3 individuals followed by the 1st day, the 5th day and the 7th day with several individuals as many as 2 individuals. Then treatment 2 had the highest number of individuals on the 9th day and the 3rd day as many as 10 and 9 individuals, then on the 7th day, the 1st day, and the 5th day as many as 6 individuals, 5 individuals and 4 individuals.

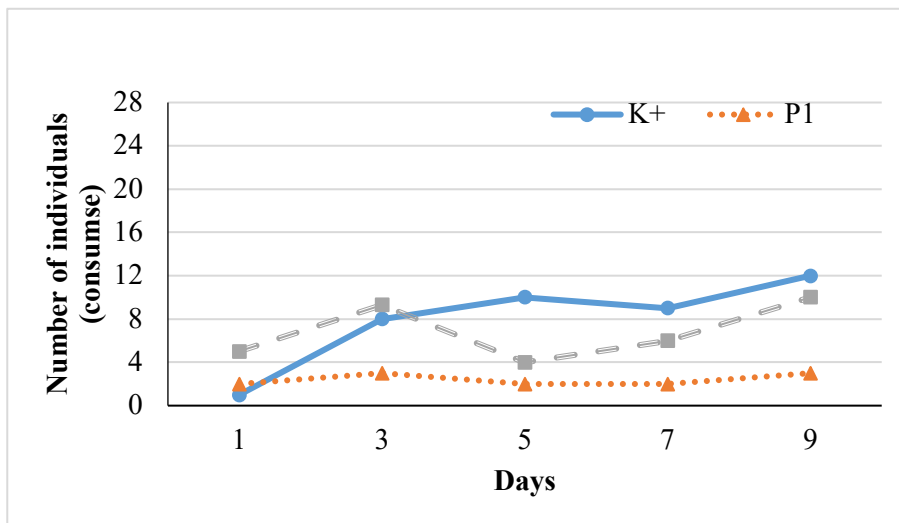


Figure 2. Number of worker bees per day in each treatment (K+, P1, and P2) for feeding behavior.

The results of the analysis of the variety of eating activities showed that there was an influence on the interaction between treatment and day, based on the calculated F probability value being smaller than  $\alpha = 0.05$ .

Table 4. Results of two-way analysis of variance for eat data

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Treatment	2	252.31	126.16	31.02	0.0001
days	4	142.89	35.72	8.78	0.0001
treatment*days	8	130.58	16.32	4.01	0.0024
Error	30	122.00	4.07		
Corrected Total	44	647.78			

Further tests conducted showed that on the 9th day, the number of K+ and P2 individuals was the highest value and was considered no different. In other parts, the number of individuals eating at P1 generally showed a very low value. The results of further tests for eating behavior data can be seen in the following Table

Table 5. Duncan's further test results for eating data

Treatment	H1	H3	H5	H7	H9
K+	1.3 ± 0.6 <sup>c</sup>	8 ± 4 <sup>abc</sup>	10 ± 1 <sup>ab</sup>	8.7 ± 4.7 <sup>ab</sup>	11.3 ± 2.1 <sup>a</sup>
P1	2 ± 1 <sup>c</sup>	3.3 ± 0.6 <sup>dc</sup>	2 ± 1 <sup>c</sup>	2.3 ± 0.6 <sup>c</sup>	2.7 ± 0.6 <sup>dc</sup>
P2	5 ± 1 <sup>cdc</sup>	9.3 ± 1.5 <sup>ab</sup>	4 ± 1 <sup>dc</sup>	6.3 ± 2.3 <sup>bcd</sup>	10.3 ± 2.1 <sup>a</sup>

Note. Mean values followed by the same letter indicate insignificant values, at  $\alpha = 0.05$ .

#### 4. Resting behavior of Apis cerana worker bees on additional feed of meat and tongka Langit banana paste.

The results of observations of treatment of resting behavior showed that the positive control had the highest number of individuals on the 9th day, namely 26 individuals, followed by the 7th day, and the 5th day as many as 7 individuals then on the 1st day and the 3rd day as many as 4 individuals. Treatment 1 had the largest number of individuals in this behavior which was on the 9th day, namely 8 individuals, then on the 1st day, the 3rd day and the 5th day which were relatively 6 individuals and on the 7th day as many as 3 individuals. Treatment 2 had the highest number of individuals on the 1st day and the 3rd day, namely 28 individuals and 27 individuals followed on the 9th day and the 7th day as many as 16 individuals and 11 individuals and on the 5th day as many as 6 individuals. The results of the observations can be seen in the following Figure

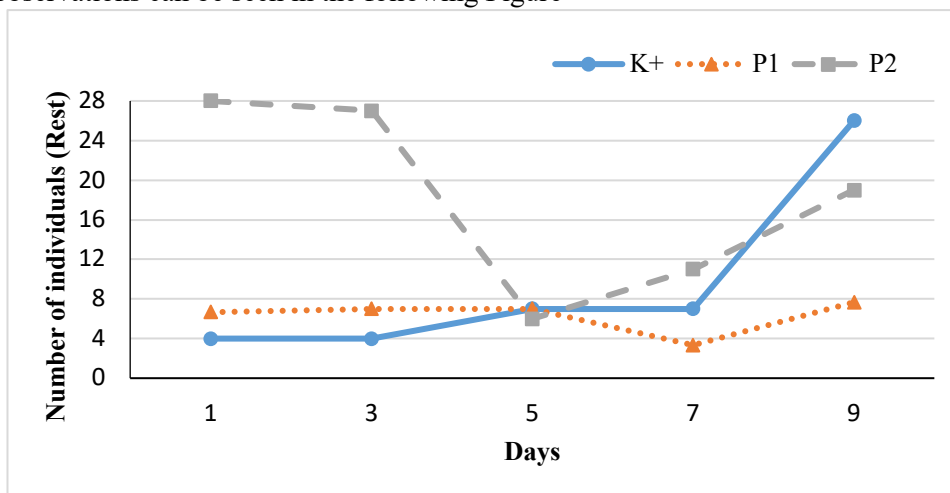


Figure 3. Graph of the average number of worker bees per day in each treatment

The results of the analysis of variance of rest activities show that there is an influence on the interaction between treatment and day, namely the calculated F probability value is smaller than  $\alpha = 0.05$ . The results of the analysis are shown in the following table.



Table 6. Results of two-way analysis of variance for rest data.

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Treatment	2	1117.20	558.60	33.52	0.0001
Days	4	738.22	184.56	11.07	0.0001
treatment*days	8	1472.58	184.07	11.04	0.0001
Error	30	500.00	16.67		
Corrected Total	44	3828.00			

Interaction and treatment analysis has the highest priority, followed by Duncan's test.

Table 7. Duncan's test results for rest data

Treatment	H1	H3	H5	H7	H9
K+	4 ± 0 <sup>cd</sup>	3.7 ± 2.1 <sup>cd</sup>	6.7 ± 4.7 <sup>cd</sup>	7 ± 5.7 <sup>cd</sup>	26.3 ± 0.6 <sup>a</sup>
P1	6.7 ± 2.1 <sup>cd</sup>	7 ± 1 <sup>cd</sup>	7 ± 5 <sup>cd</sup>	3.3 ± 1.5 <sup>d</sup>	7.7 ± 2.5 <sup>cd</sup>
P2	27.7 ± 2.1 <sup>a</sup>	27 ± 11.5 <sup>a</sup>	6 ± 3.6 <sup>cd</sup>	11.3 ± 0.6 <sup>c</sup>	18.7 ± 1.5 <sup>b</sup>

Note. Mean values followed by the same letter indicate insignificant values, at  $\alpha = 0.05$ .

Duncan's test on rest activity showed that P2 on days 1 and 3 had the highest number of individuals, then on day 5, the three treatments K+, P1, and P2 had the same number of individuals. Then the number of individuals on K+ day 9 also had high results.

The results of the study showed that there was a significant effect between P1 and P2 with the addition of sugar on eating behavior. The number of individuals eating at P1 without the addition of sugar had a very small number of individuals compared to P2 with the addition of sugar. This is directly proportional to K + which is a sugar solution. The level of sweetness in the additional feed can be concluded to affect the eating behavior of *Apis cerana* honey bees (F.). This is supported by research by Oktavia (2017) which states that the higher the sugar content of a flower's nectar, the more often bees visit the flower and bees tend not to like nectar with a sugar content below 4%. This means that the higher the sugar content in the additional feed, the more often bees visit the feed, including consuming the feed.

The highest pattern of the number of individuals performing eating behavior was in the positive control (sugar solution) in accordance with its role as a comparative treatment which is expected to produce a positive effect and also has the highest sugar content. The sugar solution as a control does have a high number of individuals compared to P2 but P2 (banana paste) has an advantage in terms of nutritional content. The use of positive control if set aside will get the best treatment, namely in P2 (banana paste). In this feeding behavior, the bees have gone through an adaptation process to the additional feed given. After identifying additional feed as a food source, the bees will use the proboscis. The proboscis is used to absorb banana juice, which is the first step in consuming additional feed. Bees use the proboscis similar to the way they consume nectar. In soft and ripe fruit flesh feed, the bees will break it using their mandibles. In banana paste, after digesting the remaining liquid, the bees use their mandibles to chew and consume the thickened paste. When making observations, it was noted that a number of *Apis cerana* honey bees used their leg pouches to carry previously digested banana paste. When bees consumed banana paste that had a thicker texture and was not in the form of a liquid, some of the *Apis cerana* bees (F.) were seen using their hind legs to accommodate a small amount of digested banana paste. *Apis cerana* bees (F.) were clearly seen using their hind legs to tuck some of the remaining banana paste into their leg pouches. The movements appear careful and coordinated, with the proboscis remaining attached to the banana paste while the hind legs are used to store the remaining paste.

The pattern of resting behavior on the fifth day that has the same value or position of the number of individuals is interesting to investigate further. This could indicate a stable pattern of resting behavior after several days of additional feeding, or there are other factors that influence resting behavior on the fifth day that need to be explored further. Find out the pattern of resting behavior in more depth, it is necessary to conduct further research involving further monitoring of the dynamics of the worker bee population and environmental factors that influence the behavior of *Apis cerana* (F.). A more controlled experimental approach can help reveal the mechanisms underlying the pattern of worker bee behavior in response to environmental changes in this case additional feed. During the resting period, *Apis cerana* honey bees tend to do a number of different activities from when actively foraging or collecting other resources. Worker bees use the resting phase to restore energy after spending time collecting and processing additional feed. At the time of observation, the resting bees had their wings stored tightly along the body because they were temporarily not using their wings, so the wings did not appear to be fully expanded. The antennae of these worker bees looked more relaxed and inactive in detecting the surrounding environment. However, there were some bees with active antennae that might be used to

communicate with other bees in the nest. The legs located near the head are sometimes used to clean the antennae or other body parts. During the resting phase, bees experience decreased activity and energy needs, which are reflected in body posture and limb activity. This study only observed the number of worker bees at a certain time, thus providing a limited picture of the adaptation, feeding, and resting behavior of *Apis cerana* (F.) honey bees. A deeper understanding of rest time, feeding duration, and social interactions between bees has not been completed in this study. This study also used sugar as an ingredient in the treatment feed, so an analysis of the sugar content in the honey composition needs to be carried out to further understand the effect of adding sugar on the composition of honey produced from this study, as mentioned in the study Sugar Profile and Rheological Behavior of Four Different Indian Honey Varieties (Kamboj, et. al., 2020).

## AUTHORS CONTRIBUTION

Hartanto, M designed and conducted the study, analyzed and interpreted the data, and wrote a draft of the manuscript. Lamerkabel, J.S.A, designed the research, analyzed and interpreted the data. Riupassa, P.A, reviewed the draft manuscript, and supervised the entire process.

## CONCLUSION

This study shows that there is an effect of providing additional feed based on tongka langit banana fruit on honey bees *Apis cerana* (F.). *Apis cerana* (F.) showed the highest adaptation to banana paste (P2) compared to sugar water (K+) and fresh fruit flesh (P1). *Apis cerana* (F.) prefers banana paste (P2) because of its sweet taste. The number of bees resting was highest on banana paste (P2), followed by sugar water (K+) and fresh fruit flesh (P1). Fresh fruit flesh (P1) was less preferred because of its low sweetness.

The study showed that P2 (banana paste) gave the best results in terms of adaptation, eating, and resting of *Apis cerana* (F.). P2 (tongka langit banana paste) and K+ (sugar water) showed a final pattern of feeding behavior that was not significantly different. In addition, banana paste (P2) has advantages over sugar water (K+) in terms of nutrition, especially higher  $\beta$ -carotene content. This makes Tongka Langit Banana Paste a potential alternative as additional feed for honey bees. This study also shows that *Apis cerana* (F.) honey bees have good adaptability, this can be seen in the second treatment (P2) in adaptation behavior and eating behavior has a high number of worker bees, meaning that even though P2 has a high adaptation pattern because its familiarity level is lower, it does not affect the eating behavior pattern. P2 still has a high number of worker bees in the eating behavior pattern.

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