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# Comparison of Royal Jelly Production among Cross Breed of Honey Bee in Period of Nectar Flow and Non-Nectar Flow

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## Abstract

Selection, using the cross breeds, has been made on the basis of the royal jelly productivity which is measured by the yield of royal jelly and per one queen cup as well as the larval acceptance in the period of nectar flow and non-nectar flow. Among them, cross breed B yielded 16.82g of royal jelly equivalent to increase of 46% and 29%, as compared to those of the control and cross breed A in the period of nectar flow, while yielded 19.37g of royal jelly corresponding to increase of 95% and 89%, as compared with the control and cross A in the period of non-nectar flow. The larval acceptance of the control was highest by 67.6% in the period of nectar flow, while that of cross B was highest by 86.0% in the period of non-nectar flow. The cross B yielded 0.34g of royal jelly per queen cup in the period of nectar flow and 0.39g in the period of non-nectar flow, which was increased by 61% and 50%, respectively as compared with the control. In conclusion, cross B yielded 18.16g of the total royal jelly and selected as the superior one in terms of the total royal jelly productivity.

Key words: Honey bee, *Apis mellifera*, Royal jelly, Yields, Cross breed

## INTRODUCTION

Honey bees are collecting water, nectar, pollen, and propolis from the nature, and produce mainly honey, and collected pollen, royal jelly, beeswax, and bee venom as well. Among them royal jelly is an ivory white colored liquid secreted from hypopharyngeal glands and mandibular of a 5~15 day-olds nurse bee for nutrition of the queen larvae and younger larvae in colony.

In as much as the official statistics of national royal jelly production has not been made in Korea yet. China is the world largest royal jelly producing country of which total production of royal jelly reaches to 3,500 tons that accounts for over 85% of the world total production (Cao *et al*., 2016; Zhang *et al*., 2017). China has already developed production techniques for royal jelly as well as established its mechanization and automation (Chen, 2002; Liu *et al*., 2011).

On the other hand, the college of animal science of Zhejiang University in China developed a high royal jelly-producing lineage of honeybees named as Zhenongda no.1ngda no.1 using a local honey bee race specified for the high yield of royal jelly since 1960 in Zhejiang province area (Chen *et al*., 2002; Cao *et al*., 2016; Zhang *et al*., 2017). Since then, studies on behavioral genetics and morphological genetics in relation to the yield of royal jelly have been conducted (Li *et al*., 2003; Su *et al*., 2003). Sahinler and Sahinler (2005) insisted that Carniolan and

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Mugla bees were found to be more suitable for royal jelly production than the Caucasian bee. Mouro and Toledo (2004) presented that the Carniolan had a higher production of royal jelly compared to the Africanized honeybees.

In Korea, a few studies in regard to production method and breeding for royal jelly have done. Lee *et al*. (2007) suggested that grafting larvae and arrangement of comb in super of hive would be efficient for royal jelly production. Kim *et al*. (2017) attempted to evaluate inbred lines related to royal jelly production and to analyze on the racial differences in regard to the major jelly protein. In Korea, breeding program of honey bees are about to set the stage, and selection of behavioral traits is mainly based on foraging behaviors of honey bees. This situation of breed programs in Korea gave rise to the suggestion to establish a breeding apiary on island areas as there is high density of honey bee populations in inland area (Lee *et al*., 2014; Lee *et al*., 2013). The objective of this study is to select a cross breed for the high yield of royal jelly during period of nectar flow and non-nectar flow.

## MATERIALS AND METHODS

### Experimental colony

The cross breed used for the experiment were bred at the isolated apiary of Department of Agricultural Biology, National Institute of Agricultural Sciences, Rural Development Administration, Rep. of Korea. The control colony was purchased from a local beekeeper producing royal jelly. All the queen bees of the experimental colonies mated between May to July, 2016 and observed 5 colonies per experimental groups.

### Colony management

The experimental colonies were managed in the double hive colony separated into queenright in chamber of hive and queenless in super of hive by a queen excluder. The experimental colonies in the period of nectar flow were comprised of eight combs in the chamber of hive and six combs in super of hive. Each colony was equalized based on uniformed developmental stages and equal numbers of bees in May 23, 2017. During the period of nectar flow, a frame of queen cups was inserted between combs of early pupa stages and positioned at the 2nd and 3rd from the left of the super of hive.

During the period of non-nectar flow, the experimental colonies were set with seven combs in the chamber of hive and five combs in super of hive. They were at the uniform growth status and of equal number of bees in August 5, 2017. The frame of royal jelly placed in the same way as in the period of nectar flow. When the early pupa combs were not enough, storage combs of honey were added.

### Observation of productivity

The frame of queen cups consisted of two movable bars which were available 33 of queen cups per bar, 66 queen cups in total. All the frames were plastic and were exclusively used for royal jelly production. They were purchased from a local beekeeping market. One day-old larvae from a single hive colony were collected and grafted to the queen cups. To minimize the error grafting from grafting larvae, grafting on the experimental colonies was done in turns one after another repeatedly.

After grafting larvae, the frame of queen cup was introduced to the super of hive and the yield of royal jelly production was measured after 72 hours. Observation was done 10 times from May 26, 2017 to June 22, 2017, and no supply of sugar was applied in the period of nectar flow. In the period non-nectar flow, 11 time observation was made from August 8, 2017 to September 7, 2017, and 600 ml of sugar was provided in every two days. 30ml of sugar syrup was supplied to the frame in a zigzag to enhance the acceptance of grafting larvae just after the frame of queen cup were put to the colonies.

### Analysis of statistics

Statistical analysis of the data was employed with Duncanrs. Observation was done 10 times fro SAS Version 9.3 (SAS Institute, 2010).

**Table 1.** Comparison of yield of royal jelly among cross breed in the period of nectar flow

| Cross breed | Acceptance percentage of grafting larva (%) | Average yield (g) | Yield per one queen cup (g) |
|---|---|---|---|
| A | 56.58a±12.97 | 13.01b±2.71 | 0.35b±0.02 |
| B | 63.73a±10.37 | 16.82a±2.47 | 0.39a±0.05 |
| Control | 67.60a±7.94 | 11.50c±1.58 | 0.26c±0.04 |

*Different letter indicates significant mean differences (P<0.05).

**Table 2.** Comparison of yield of royal jelly among cross breed in the period of non-nectar flow

| Cross breed | Acceptance percentage of grafting larva (%) | Average yield (g) | Yield per one queen cup (g) |
|---|---|---|---|
| A | 57.58c±10.34 | 10.21b±2.42 | 0.28b±0.07 |
| B | 86.01a±5.00 | 19.37a±3.23 | 0.34a±0.07 |
| Control | 72.53b±9.81 | 9.93c±1.67 | 0.21c±0.04 |

*Different letter indicates significant mean differences (P<0.05).

**Table 3.** Comparison of the yield of royal jelly among cross breed in the period of total observation

| Cross breed | Acceptance percentage of grafting larva (%) | Average yield (g) | Yield per one queen cup (g) |
|---|---|---|---|
| A | 57.10c±11.38 | 11.54b±2.88 | 0.32b±0.06 |
| B | 75.40a±13.81 | 18.16a±3.11 | 0.37a±0.06 |
| Control | 70.18b±9.10 | 10.68c±1.78 | 0.23c±0.05 |

*Different letter indicates significant mean differences (P<0.05).

## RESULTS

### Yield of royal jelly in the period of nectar flow

The yield of royal jelly of the cross breeds in the period of nectar flow were measured. It showed that acceptance percentage of the control was 67.60%, highest among the experimental colonies, of cross breed A was 56.58% and of cross breed B was 63.78%, as it is shown in Table 1. On the other hand, average yield of cross breed B was 16.82g and highest among the experimental colonies, while cross breed A, 13.01g and control, 11.50g. These figures indicated that the average yield of cross breed B is higher by 46.3% and 29.3 %, as compared to those of cross breed A and control, respectively.

In the total comparison of nectar flow, acceptance percentage of cross breed B is lower by 3.87% than that of control but there was no statistical difference. Yield per one queen cup of cross breed B, however, was 0.39g equivalent to 0.13g higher than control. The average yield of Cross breed B was 16.82g, that is 3.81g and 5.32g more than that of the cross breed A and of the control.

### Yield of royal jelly in the period of non-nectar flow

In regard to acceptance percentage of grafting larvae in the period of non-nectar flow cross breed B was highest by 86.01%, followed by 72.53% for the control and 57.58% for cross breed A, as shown in the Table 2. The average of yield of cross breed B was 19.37g, cross breed A, 10.21g and control, 9.93g. Cross breed B was highest among them. Cross breed B was also highest in the royal jelly yield per one queen cup, which showed 0.34 g, while the control was lowest with 0.21g of yield per one queen cup and was middle with acceptance percentage among three groups.

Yield per one queen cup showed 0.28g for cross breed A, 0.34g for cross breed B, and 0.21g for control, and it indicated that cross breed B was 50.0% and 11.4% higher

percentage than control and cross breed A. In the total comparison of acceptance percentage in the period of non-nectar flow cross breed B was 13.48%, higher than the control, and of yield per one queen cup cross breed B was 0.34g, 0.13g more than the control. Also, average yield of cross breed B was highest with 19.37g, while control and cross breed A were 9.93g and 10.21g, respectively. They were below the average yield.

### Total Yield

As shown in Table 3, acceptance percentage of grafting larva of cross breed B was 75.4%, of average royal jelly yield, 18.16g and 0.37g, of royal jelly yield per one queen cup. This indicates that cross B is the best in the period of nectar flow as well as in the period of non-nectar flow.

## DISCUSSION

The study of royal jelly has been largely focused on the development of production techniques and breeding of high yielding varieties. Recently, Kim *et al*. (2017) reported that there was a racial difference in the yield ability of royal jelly but there was no statistical difference. This report, however, revealed that there was significant differences in the royal jelly yield of cross breeds such as acceptance of grafting larvae, average yield of royal jelly, and yield per one queen cup between the period of nectar flow and in the period of non-nectar flow. Overall, it was observed somewhat low results in acceptance percentage of grafting larvae. Cross B showed 18.16g of average yield of royal jelly which was higher than the control by 70%, 0.37g of yield per one queen cup, higher than the control by 60.9%. These figures suggested there was a possible successful selection of high yielding cross breeds.

Li *et al*. (2003) also described that genetic factors were probably involved in the difference between total yield and yield per one queen cup was due to the genetic factor. Chen *et al*. (2002) succeeded in the establishment of the exclusive line for royal jelly through generation to generation selection over 30 years in the isolated area of China. In this regards, further studies on this lineage are needed. This is obviously involved in interaction of genetic and environmental factors. We have required for the future to compare and analyze genetic characteristics of superior cross breed that has outstanding yield.

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