APICULTURE

# Relation between Honey Production, Defensiveness, and Diameter of Brooding Cells of *Apis mellifera* L., in the Ecuadoran Highlands

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## ABSTRACT

**Objective:** The purpose of this research was to analyze the relationship between defensiveness, diameter of worker brooding cells, and bee honey production (*Apis mellifera*) in the Ecuadoran Highlands.

**Methods:** A number of 75 bee colonies were evaluated in March-April and May-July (production stage), and September, in altitudes of 2 600 and 3 274 m above sea level. Each colony's defensiveness was determined using the pin method; cell diameter was measured in ten worker brooding cells in the hive, and honey production was determined by weight difference.

**Results:** A mean value of 14 stingers/min was observed; the maximum and minimum values of this trait were 47 and 4 stingers/min. The diameter of the 10 cells was estimated to have a mean of 5.28 cm, with maximum and minimum values, respectively. A significant correlation (-0.358******) was identified, which meant that the colonies with the highest defensiveness had a lower cell diameter. Production showed a mean of 25.08 kg of honey/colony, with no relation to the variables studied.

**Conclusions:** No relationship was observed between production and the other variables studied, thus allowing the selection of less defensive colonies and greater cell diameter, without affecting honey production.

**Key words:** *Apis mellifera, defensiveness, cell diameter, honey production*

## INTRODUCTION

Bee keeping is an important part of food safety (Sánchez *et al.*, 2013); it is also essential for the economy, due to the value generated by the products in the hives (Valdés, 2013), and their key role as pollinators of staple foods in human nutrition.

Defensiveness of colony bees is an important trait to manage apiaries. A high defensive behavior is not favorable for keepers, but colonies with low defensiveness may be easy preys of natural enemies, such as wasps, birds, or mammals (Uzunov *et al.*, 2014). It involves a complicated sequence of actions by bees, and it can be expressed through a variety of intensities, from aggressive to nonresistance (Kastberger *et al.*, 2009). This type of behavior evolved in both open-nesting honeybees (Kastberger *et al.*, 2008) and those living in holes (Ruttner, 1988). African bees and their hybrids are generally known to express high defensiveness (Tibatá *et al.*, 2018), which limits their value as honey producers. In that sense, the expression of that behavior is used to recognize the presence of Africanization, though it is not defining.

Of all the services bees offer to humans, honey production is highly important for apiculture, therefore, it is included in several selection and breeding programs. However, their high variability due to environmental, management, sanitary, and genetic factors make colony selection difficult. Therefore, it is important to study the relations between defensiveness, worker brooding cells, and honey production, in order to choose high producing bees with low defensiveness (author), which has not been studied in the Ecuadoran Highlands. The aim of this research was to analyze for the first time the relationship between defensiveness, diameter of worker's brooding cells, and bee honey production (*Apis mellifera*) in the Ecuadoran Highlands.

## MATERIALS AND METHODS

The research was done between March and September (2017). Overall, 15 apiaries (15 combs/apiary) comprising 75 Africanized hives were studied in the provinces of Tungurahua and Chimborazo (Table 1).

**Table 1. Geographical location of the apiaries**

| Apiary | Location | Latitude | Longitude | Height |
|---|---|---|---|---|
| A1 | Tungurahua | 1°16'06"S | 78°34'50"W | 2 607 |
| A2 | Tungurahua | 1°22'09"S | 78°36'19"W | 2 879 |
| A3 | Tungurahua | 1°18'16"S | 78°39'16"W | 2 936 |
| A4 | Tungurahua | 1°19'02"S | 78°39'16"W | 3 047 |
| A5 | Tungurahua | 1°35'17,37"S | 78°46'05,25"W | 3 279 |
| A6 | Tungurahua | 1°33'11,2"S | 78°42'32,4"W | 3 168 |
| A7 | Chimborazo | 1°41'45,57"S | 78°45'16,46"W | 2 939 |
| A8 | Chimborazo | 1°39'26,17"S | 78°34'38,49"W | 2 727 |
| A9 | Chimborazo | 1°42'46,63"S | 78°39'50,33"W | 2 967 |
| A10 | Chimborazo | 1°35'11"S | 78°45'09"W | 3 205 |
| A11 | Chimborazo | 1°35'18"S | 78°46'03"W | 3 262 |
| A12 | Chimborazo | 1°41'34"S | 78°40'11"W | 2 834 |
| A13 | Chimborazo | 1°35'46,75"S | 78°39'51,45"W | 2 870 |
| A14 | Chimborazo | 1°43'46,5"S | 78°36'47,6"W | 2 616 |
| A15 | Chimborazo | 1°46'40,91"S | 78°35'10,99"W | 2 863 |

Samples of defensiveness and the diameter of brooding cells were collected three times in the March-April period (before production), May-July (during production), and September (after production); the productive yields were achieved at the end of the production stage.

The colony inclusion and exclusion criteria used were,

Inclusion criteria:

- Apiaries with Langstroth hives
- Adequate strength of the colonies chosen (7 combs with bees containing an average of 3 brooding combs) (Vaziritabar *et al.*, 2016)
- Honey production above the mean (15.2 kg/colony) (AGROCALIDAD, 2016)
- No treatment received against varroa prior to the study.
- No introduction of queens in recent years.

Exclusion criteria:

- Hive swarming (exploration every 15 days)
- Transhumance of the apiary.
- Farmer's refusal to participate in the study.

The hives in the study had a brooding chamber and two half-production peaks. Besides, labor continued during the period, and the hives evaluated were the same the samples were collected from. The honeycombs in the brooding chamber of the hives chosen were changed every two years.

The defensive behavior was evaluated according to the methodology of Collins and Kubasek (1982), and modified by Guzmán-Novoa *et al.* (2003), with the use of a black suede flag (10x10 cm). The test was performed between 10:00 am and 3:00 pm simultaneously in all the colonies selected in every apiary, without smoke. The flag was exposed in front of the bees and it was waved for 60 seconds. The results were averaged and grouped by quartiles named gentle (≤ 10 stingers), mid-gentle (10.1-13), aggressive (13.1-17), and very aggressive (17.01+).

The SARH-USDA (1986) methodology was used to evaluate the diameter of worker brooding cells. Three honeycombs were chosen per hive, from the middle of the brooding chamber, and 10 cells in a row were measured on both sides, to average and reduce error, until six measurements were made by hive.

Honey production was measured using the methodology suggested by Büchler *et al.* (2013), and it was evaluated separately. The rises of each colony were weighed before and after honey collection, and the difference was considered harvested honey. All the honey collected throughout the harvest season was summed to estimate total honey production. The honey stored in the brooding chambers was not included.

The normality of the variables studied was evaluated (Kolsmogorov Smirnov), and bivariate correlations among all the variables were made (Spearman). SPSS (IBM Corp. 2012. IBM SPSS Statistics for Windows, version 21.0, was used. Armonk, NY: IBM Corp.).

## RESULTS AND DISCUSSION

The high defensive conduct of a honey bee is one of the main issues of beekeeping in most countries in the Americas, since handling becomes difficult, and and animals and people are involved in accidents (close to lethal).

In response to the physical stimulus used to evaluate the defensive conduct, a mean value of 14 stingers/min were observed, and the maximum and minimum values of this trait were 47 and 4 stingers/min, respectively. The behavior during each sample collection was 15.68, 16.41, and 11.80 stingers/min, respectively. This outcome was inferior to a study in colonies of Africanized bees in Brazil, by Faita *et al.* (2014), who reported mean values of 28 ± 2, and 22 stingers/min, respectively. Also in Brazil, Pinto *et al.* (2016) found a mean of 27 stingers/min. Moreover, defensiveness varied among the samples.

The results achieved show that the defense response of bee colonies in the area of study, compared to bees in other countries, was low. This finding is important and valuable, since it becomes an aspect of interest in terms of genetic breeding, which allows them to be used as parent hives to start selection projects based on this highly inherited trait (Barrera, 2013; Esquivel *et al.*, 2015). Furthermore, Stort and Gonçalves (1991) pointed that this behavior may be controlled by two pairs of recessive genes found in Africanized bees, provided they are crossed with Italian bees. Consequently, this feature may be influenced by the effects of genetic dominance (Guzman-Novoa *et al*., 2002).

No correlation was observed between defensiveness and altitude (r=0.151; p=0.195), so this variable did not have an influence on the defensive behavior of the colonies. Similar results were found by Esquivel *et al.* (2015) in Mexico, and Mantilla *et al.* (1997), in Colombia. Additionally, differentiated defensive behaviors were identified in a single apiary, though the colonies were kept under similar conditions, which coincides with the findings of Pinto *et al.* (2016).

An evaluation of worker's cell diameter showed a mean of 5.28 cm in the 10 cells, with maximum and minimum values of 5.50 and 4.95 cm, respectively. Similar values were found in Nicaragua by Düttmann *et al.* (2013) in Africanized bees, with a mean of 5.22 cm, and 5.54 cm and 4.85 cm as maximum and minimum values, respectively. In relation to the European bees in Cuba, Pérez and Rodríguez (2013) found means of 5.27 cm, with 5.42 cm in motherly hives, and 5.35 cm in fatherly ones, whereas Sanabria (2007) found means of 5.27 cm.

By correlating the variables of defensive behavior with the diameter of the cell, a highly significant (-0.358**) negative correlation was identified, which indicates that the colonies with the greatest defensiveness have the smallest diameter in the cell. It is linked to a smaller size, compared to the less defensive bees. (Fig. 1), as reported by Uribe *et al.* (2003).

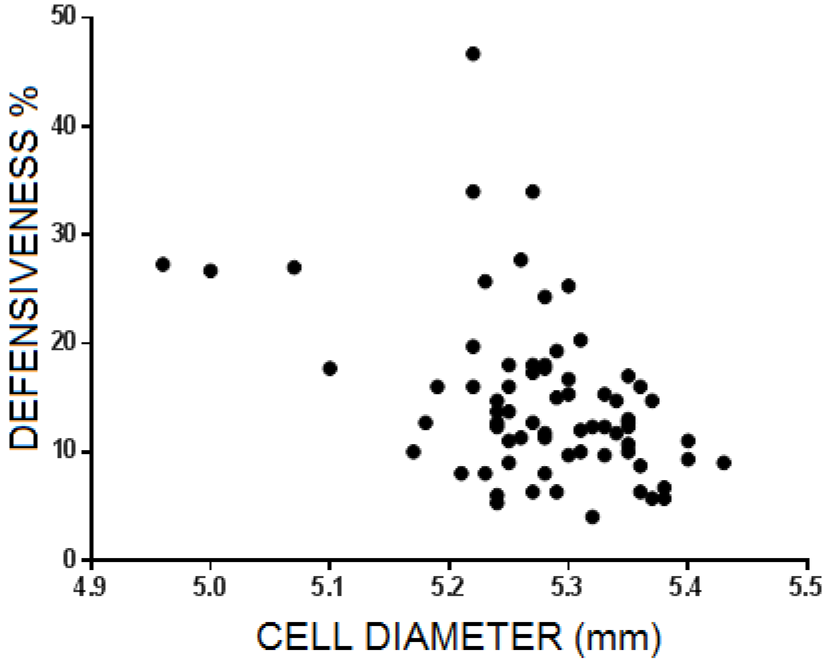


**Fig. 1. Correlation between defensiveness and diameter of worker brooding cells of *Apis mellifera* in the Ecuadoran Highlands; n=75.**

Ruttner (1988) demonstrated that the African bees (*A. mellifera scutellata*) build smaller cells than the Europeans. In ten cells, the Africanized bees built 4.7-4.9 cm cells, and the European bees made 5.2-5.6 cm cells. Similar results were achieved in Brazil by Message and Gonçalves (1995) with 4.7-5.1 cm cells, and Berry *et al.* (2010), with 4.9-5.3 cm cells. Moreover, Hall *et al.* (2015) argued that the Africanized bees did not go beyond 4.90 cm in natural combs, and the European bees built 5.20 cm, in patterned wax sheets, like the ones used in Ecuador.

Zhou *et al.* (2010) said that African bee *A. mellifera scutellata* and its hybrids are smaller and live in smaller cells than the European breeds or their crossbreds, and that this is a highly hereditary trait. Likewise, Winston (1992) said that the Africanized bees build smaller cells because they are 10% smaller (12.7 and 13.9 cm long of Africanized and European workers, respectively), and 33% less heavy than the European bees (62 mg of Africanized workers vs 93 mg of European workers).

Concerning honey production, the mean observed was 25.08 kg (hive, and this variable showed no correlation to altitude (r=-0.015; p=0.897), defensiveness (r=-0.047; p=0.688), or cell diameter (r=0.146; p=0.211). These results indicate that honey production may depend on other factors, like the size of the population, dedication of bees, and the environment (Medina-Flores *et al.*, 2019). Hence, the occasional existence of low or no correlation with certain variables indicates that their effect is diluted within a large variety of factors involved in production. Similar results were found in Mexico by Medina-Flores *et al.* (2014), with a mean production of 27.5 ± 18.9 Kg in the fall, and 21.6 ± 14.9 Kg in spring.

The similarity in honey productions at different altitude levels may be explained by the evaluation performed during the period with the highest nectar flow, which coincides with massive flowering of eucalyptus (*Eucaliptus globulus Labill*), the main nectar-producing species in the area (Masaquiza *et al.*, 2017). The local native vegetation has been almost completely replaced by eucalyptus, so the natural variations in the vegetation have been drastically reduced.

Likewise, the lack of correlations between the studied variables is advantageous, since highly productive hives and low defensiveness can be found. This indicates that hives could be chosen for genetic improvement leading to high yields, low defensiveness, and larger cell diameters, which are linked to the presence of European individuals.

## CONCLUSIONS

No relation was observed between production and the other variables studied, thus allowing the selection of less defensive colonies and greater cell diameters, without affecting honey production.

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## AUTHOR CONTRIBUTION

Author participation: (include the initials of each author separated by comas): Conception and design of research: DAMM, LMCR, BD, and AAC, data analysis and interpretation: LMC, BLDM, and AAC, redaction of the manuscript: LMC, BLDM, and AAC, redaction of the manuscript:

## CONFLICTS OF INTERESTS

None