Defensive hygienic and productivity behavior analysis in *Apis mellifera* for queen selection and breeding in the Nariño department*

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ABSTRACT. Introduction: Nariño department has great potential for beekeeping activity as its diverse climate supports the growth of a wide variety of melliferous flora. Nonetheless, the beekeeping activity in this region plays a minor role in the agricultural sector. The low development of this activity can be partly explained by the lack of technology transfer, limited unionization, and lack of knowledge of bee handling, diseases, and defensive behavior. **Objectives:** To assess the defensive, hygienic, and productive behavior of ***Apis mellifera*** for queen selection and breeding in the Nariño department. **Methods:** The study was conducted in four towns located in the Nariño department. Five hives in production were selected from one apiary in each town. The variables assessed were: defensive behavior, hygienic behavior, varroa infestation in worker brood, varroa infestation in adult bees, and honey production. Pearson's correlation coefficient was calculated between each of these variables and the climatic factors reported by each town during the assessment period. **Results:** The removal of dead bees ranged between 82.6% and 90.6% in the four apiaries; hence, the hives were considered to be hygienic. A high level of varroa infestation in adult bees was observed in one of the apiaries (8.38%), and the defensive behavior was remarkably high in all the apiaries assessed. **Conclusions:** Honey production showed a negative correlation with precipitation ($P < 0.001$), suggesting the adverse effect of increased precipitation on honey production.

Keywords: defensive behavior, environmental factors, selection, honey production.

Diagnóstico sanitario y productivo de *Apis mellifera* para la selección y cría de reinas en el Departamento de Nariño

RESUMEN. Introducción: el departamento de Nariño posee un gran potencial para la producción apícola, su diversidad climática hace posible una alta variedad de flora melífera; no obstante, la apicultura en la región tiene una baja participación en el sector agropecuario. Algunas de las razones del bajo crecimiento de dicha actividad son la falta de transferencia tecnológica, la escasa agremiación, el desconocimiento sobre su manejo, las enfermedades y la agresividad de las abejas. **Objetivos:** evaluar el comportamiento defensivo, higiénico y productivo de ***Apis mellifera*** del departamento de Nariño para la selección y cría de reinas. **Métodos:** El estudio se realizó en cuatro localidades del departamento de Nariño y en cada uno de ellas se eligió un apiario donde se seleccionaron cinco colmenas en producción. Las variables evaluadas fueron: el comportamiento defensivo, conducta higiénica, la infestación de varroa en cría de obreras, la infestación de varroa en abejas adultas y la producción de miel. Posteriormente, se realizó una correlación de Pearson de dichas variables con los factores climáticos reportados en el periodo evaluado de cada municipio. **Resultados:** se obtuvo un rango de variación de 82,6 % a 90,6% de remoción de crías muertas en los cuatro apiarios, considerando que las colmenas son higiénicas; en uno de los apiarios se evidenció un alto nivel de infestación de varroa en abejas adultas correspondiente al 8,38%, mientras que el comportamiento defensivo reportado fue considerablemente alto en los cuatro apiarios. **Conclusiones:** La variable producción de miel presentó correlación negativa con la precipitación, con un nivel de significancia de $P<0,001$, concluyendo que dicha producción se ve afectada cuando las lluvias aumentan.

Palabras clave: defensividad, factores ambientales, selección, producción de miel

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Introduction

Rustic beekeeping prevails in Colombia since transition processes toward standardized techniques are incipient (Flórez & Ward, 2013). A large percentage of beekeepers are farmers who have acquired knowledge in a traditional way and do not work full time on beekeeping activities (Silvia, Arcos & Gómez, 2006).

Nariño department has immense potential for beekeeping and honey production due to its climate and plant diversity (Government of Nariño, 2016). However, the Ministry of Agriculture and Rural Development (2018) reported that Nariño department ranks 23rd in honey production at the national level, with an estimated production of 20 tons in 2018. The low contribution to the sector may be attributed to lack of knowledge of handling highly aggressive bees, technology transfer, and limited unionization (Gómez, Tello & Muñoz, 2007).

One of the long-term solutions to the challenges faced by the beekeeping industry is genetic selection of honey bees (Cobey, 2007), which demands the identification of the characteristics to be improved in each productive system (Lorenzo, 2010). Defensive and hygienic behaviors as well as productivity are the most relevant characteristics for the selection of queen bees.

Aggressive behavior among bees has been reported to be one of the reasons for the low increase in the number of new producers (García et al., 2017). In contrast, Pérez & Demedio (2014) and Principal et al. (2008) state that hygienic behavior is a natural resistance mechanism to diseases, and its variability and heritability turn it into a basic desirable trait in genetic selection programs. This behavior allows for the identification, removal, and cleaning of the cells housing dead, sick, or parasitized brood (Medina et al., 2014). In addition, it is important to select bees already adapted to the climate conditions of the area to obtain good for them to withstand food shortages and achieve good honey and/or pollen production.

Furthermore, Padilla et al. (2010) state that the regular replacement of queen bees can remarkably improve the honey production and disease resistance; in fact, in productive systems, it is necessary to identify progenitor hives with the abovementioned characteristics for the breeding and replacement of queen bees. This study aimed at assessing the defensive, hygienic, and productive behavior of ***Apis mellifera*** for queen brood selection in the Nariño department.

Materials and Methods

The research was conducted from February to November 2018 in four apiaries. The apiary in ***Centro Internacional de Producción Limpia–Lope*** and the one in ***Granja Experimental Botana*** at Nariño University are located in the municipality of Pasto. Pasto is situated 2527 m above sea level, with an average temperature and annual precipitation of 13.3°C and 700 mm, respectively. (Benavides & Marcillo, 2016). The third apiary is located in ***Finca San Juan*** in the municipality of Consacá, at an altitude of 1710 m above sea level with average temperature and annual precipitation of 19°C and 1700 mm, respectively (Territorial Development Plan, 2016–2019). The apiary in ***Finca La Primavera*** in the municipality of Chachagüí is situated 1650 m above sea level, with the respective average temperature and humidity values of 20°C and 76%(Territorial Development Plan, 2016–2019).

The following variables were assessed:

Defensive behavior: Analysis of defensive behavior was evaluated according to Payró, Vandame & Gómez (2010) using a five-level scale to determine defensive response, where 1 is the most aggressive and 5 the most docile.

Hygienic behavior: Hygienic behavior analysis was carried out by means of a deep puncture test which involves choosing a capped brood frame, selecting 100 cells, and piercing the pupae by deep puncture. After 24 hours, the number of uncapped cells removed by the bees was recorded (Vandame et al., 2012).

The removal percentage was determined by the following formula:

$$HB = \frac{Total\ perforated\ cells - Total\ unremoved\ cells}{Total\ perforated\ cells}$$

Varroa infestation in brood (VIB): the methodology proposed by Payró, Vandame & Gómez (2010) was used for this analysis. Hives were decapped and bee brood carefully removed. The body and the inside of each cell were inspected to count the total number of varroa mites found in each brood. The VIB percentage was determined by the following formula:

$$VIB = \frac{Total\ varroa\ mites\ found}{Total\ cells\ inspected} \times 100$$

Varroa infestation in adult bees (VIAB): Samples of ~100 bees each was collected in jars containing 70% alcohol and subsequently shaken to dislodge the varroa mites from the bees. Next, the bees and varroa mites were separated and counted. The VIAB percentage was determined by the following formula:

$$VIAB = \frac{Total\ varroa\ mites\ found}{Total\ number\ of\ bees\ in\ the\ sample} \times 100$$

Honey production: Honey was extracted according to the nectar flow in the area, and the number of frames containing honey in each hive was recorded. Subsequently, the weight of the decapped honey frame was subtracted, and the frame centrifuged in stainless steel. Finally, the empty frame was weighed, and the weight difference was recorded as the amount of honey in a frame.

Amount of honey per frame = ***Weight of decapped honey frame*** - ***weight of the empty frame***

Climate characteristics of the assessed municipalities: Monthly data concerning precipitation, minimum temperature, and maximum temperature in 2018 and 2019 were obtained from the Institute of Hydrology, Meteorology, and Environmental Studies (IDEAM, for its Spanish acronym). The data were used to determine the hydrologic year of each season, as well as maximum and minimum peaks of precipitation and temperature for each location.

The student version of InfoStat software was used to analyze the obtained results. Correlation between the behavior or honey production and the climate factors in 2018 and 2019 (data provided by IDEAM) was studied by Pearson's correlation analysis.

Results and Discussion

Defensive behavior

The bees isolated from hives in the apiaries located in ***Granja Experimental Botana*** and ***Finca La Primavera*** demonstrated the highest defensive response, with values of 1.2 and 1.8, respectively (Table 1). These values set both apiaries in the defensive response level No. 1 and indicate the highly aggressive nature of the isolated bees. Hence, the handling of such bees is difficult, and it is crucial to keep them isolated from other productive systems. By contrast, the defensive response values for the bees from apiaries in ***Centro Lope*** (Pasto) and ***Finca San Juan*** were 3.8 and 3.6 respectively, setting them in the defensive response level No. 3, which indicates that these bees are slightly more docile.

Overall, it could be stated that the level of defensive response of the bees of the hives in the studied regions is regarded as high, given that neither genetic selection has been performed nor improved biological material has been acquired.

Table 1. General characteristics of behavior and honey production

Characteristic assessed	*Granja Botana*	*Centro Lope*	*Finca San Juan*	*Finca La Primavera*
Defensive behavior	1.2	3.8	3.6	1.8
Hygienic behavior (%)	85.96	90.67	90.24	82.60
Varroa infestation in brood (%)	23.33	0	4	40
Varroa infestation in adult bees (%)	6.96	1.31	0.00	8.38
Honey production per hive (kg)	9.7	9	11	11.5

Hygienic behavior

The hygienic behavior of the bees of the assessed hives ranged from 82.60% to 90.67%. These values are lower than those reported by Contreras et al. (2016) who observed hygienic behavior values between 87.9% and 93.8%. Similarly, Espinosa et al. (20018) observed a hygienic behavior value of 88% in the studied hives when using the same deep puncture method. Lorenzo (2010) states that colonies are to be considered hygienic if ≥80 % of the dead brood are removed within 24 hours. Therefore, it can be concluded that the bees in the apiaries assessed showed good hygienic behavior.

Assessment of VIB

Throughout the assessment period, the highest level of ***Varroa destructor*** infestation in worker brood was observed in the apiaries in ***Finca La Primavera*** (40%) and in ***Granja Experimental Botana*** (23.33%). These were considerably high levels of infestation compared to those reported by Araneda et al. (2008) who observed an average infestation of 7%. Similarly, Sanabria et al. (2015) reported an average infestation of 4.89%. From our results, it can be stated that the infestation in the assessed hives need to be immediately controlled, considering that colony loss is predictable when a high proportion of bees in a hive are parasitized (Medina et al., 2011).

Assessment of VIAB

Apiaries showing high varroa infestation in worker brood also reported the highest levels of varroa infestation in adult bees. The apiaries in ***Finca La Primavera*** and ***Granja Experimental Botana***, exhibited infestation levels of 8.38% and 6.96% respectively (Table 1). Velásquez & Vargas (2016) found that 75.29% of the assessed hives showed low infestation levels (≤3%), while a moderate level of infestation was observed in 14.11% (3 %–5 %), and a high infestation level was observed in 11% (>5%). They further concluded that the type of handling technique used has a direct impact on the presence of mites.

The assessed hives showed very high levels of infestation, and require the immediate implementation of a plan to monitor and control varroa, since a mite population approaching 10% of that of adult bees within a colony has a negative effect on productivity (Espinosa & Guzmán, 2007).

Contreras et al. (2016) reported that the ***Varroa destructor*** population index varies seasonally, being higher during the peak of the flowering season. Therefore, to ensure that the mite population variation does not affect the hives adversely, constant monitoring should be performed throughout the year.

Honey production

Honey production amounted to 11 kg and 11.5 kg/hive/year in the towns with mild weather, i.e., Chachagüí and Consacá (Table 1). On the other hand, the two apiaries situated in Pasto, a cold weather location, produced 9 and 9.7 kg/hive/year, respectively. According to the Ministry of Agriculture and Rural Development (2018), in regions whose altitude ranges from 1000 and 2900 m above sea level, the average honey production is between 10 and 22 kg/hive/year. In this regard, the production values obtained from the apiaries in mild weather regions are within the reported range, whereas those from apiaries in cold weather are close to the reported range. Nonetheless, if the parameters assessed for the selection of queen brood and improved control of V. destructor mites are taken into account, honey production might increase.

Climatic factors

Figure 1 shows the precipitation levels in each location as a function of the hydrologic year. In ***Granja Botana***, the hydrologic year is from August to July, in Chachagüí from February to January, and in Consacá from April to March. All the three locations show two maximum precipitation peaks; in December and January and in April and May.

Ramos et al. (2017) state that honey production is closely related to plant phenology, which, in turn, has a close link with climatic conditions. Hence, honey harvest from the apiaries in the municipality of Pasto was carried out in February and August, times of the year when the precipitation starts to decrease after its maximum peak. By contrast, in Chachagüí and Consacá, honey was harvested in May and November, when the precipitation begins to increase, as shown in Figure 1.

The preceding behavior can be attributed to the effect of rains on the phenology of the regional flora. In the municipality of Pasto, the nectar flow is contributed by ***Eucalyptus globulus*** and ***Brassica napus*** (Reina et al., 2019), species characterized by their slow blooming, which may extend from one to two months (Ramírez, 2017). Chachagüí and Consacá are coffee-growing towns (Lagos et al., 2019); hence, honey production in these regions is dependent on the flowering of coffee plants., They start blooming intensively immediately after the beginning of the rainy season.

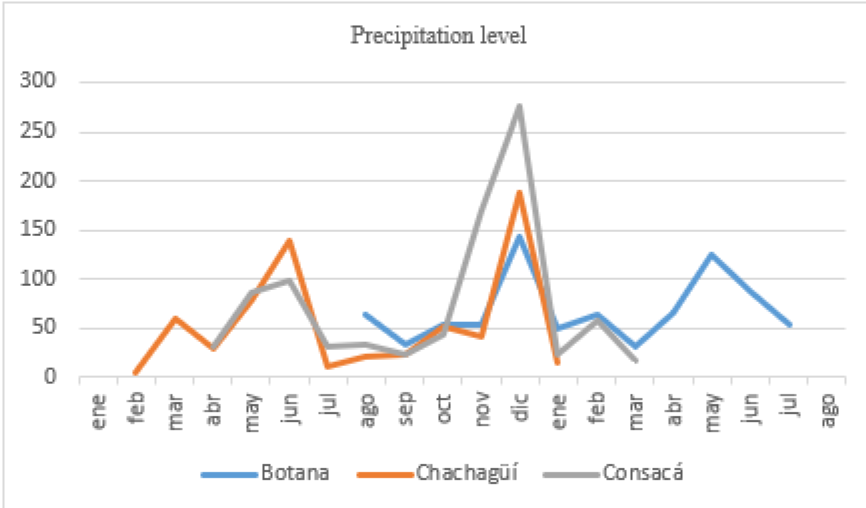

Figure 1. Hydrologic year in the locations assessed

As shown in Table 2, in the correlation analysis between climatic factors and the variables assessed, the variable VIAB is positively correlated with the minimum and maximum temperatures ($P > 0.01$). This suggests that the presence of ***V. destructor*** in the hive tends to increase when temperature rises or falls to the extreme. Furthermore, Lorenzo (2010) states that temperature, precipitation levels, relative humidity, solar radiation, and wind are the physical factors that mostly affect the health of the hives.

Table 2. Correlation coefficients between climate factors and the variables assessed

Variable[1]	Factors[2]					
	Pr (mm)		T_{min} (°C)		T_{max} (°C)	
DB (%)	−0.25863		0.16045		0.1367	
HB (%)	−0.00661		−0.04077		0.1367	
VIB (%)	−0.16696		0.18587		0.24166	
VIAB (%)	−0.42268		0.51238	**	0.54694	**
HP (kg)	−0.69795	***	0.66196	**	0.69755	***

[1] DB= Defensive behavior; HB= Hygienic behavior; VIB= Varroa infestation in brood; VIAB= Varroa infestation in adult bees; HP= Honey production;

[2] Pr= Precipitation; T min= Minimum temperature; T max = Maximum temperature

The variable honey production showed a negative correlation with precipitation (P < 0.001); i.e., the higher the precipitation, the lower the honey production. This behavior can be attributed to the fact that excessive precipitation may affect the regular foraging activity or wash away the nectar from the flowers (Martell et al., 2019).

In addition, there was a positive correlation between honey production and maximum temperature (P < 0.001), which implies that honey production increases when temperature rises. This behavior can be attributed to the occurrence of sunny days, a key player in the photosynthesis process carried out by the plants that produce nectar and pollen to attract bees and other insects (Cuentas, 2017). The maximum temperature reported during the assessment period was 27°C, which, according to Castellanos et al. (2016), does not affect bee behavior. They report that bees decrease their pollen and nectar foraging activity to collect water at temperatures are >32°C.

Conclusions

In conclusion, considering that no genetic selection program is currently employed in the Nariño department and the beekeeping sector uses native wild bees, the bees of the apiaries studied demonstrate acceptable hygienic and productive behavior. Similarly, the beekeepers' lack of knowledge of the activities required to identify and control the ***Varroa destructor*** mite was observed. Environmental factors were shown to have a direct impact on the health and productivity of the hive; hence, beekeepers should take appropriate control measures optimized to handle sudden temperature and precipitation changes to manage pests. In fact, the average honey production in the region might be improved with the implementation of appropriate technical management in the apiaries, including preparation of hives during pre-flowering period, health monitoring and regular replacement of queen bees with those obtained from genetic selection programs that guarantee the beekeepers' honey production goals.

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