

## **Evaluation of low-cost, easy-to-prepare protein diets using beer yeast, sugar cane yeast and texturized soy protein for Africanized *Apis mellifera* in apiaries in Santa Catarina, Brazil**

**Avaliação de dietas proteicas de baixo custo e fácil preparo utilizando levedura de cerveja, levedura de cana-de-açúcar e proteína texturizada de soja para *Apis mellifera* africanizada em apiários de Santa Catarina, Brasil**

**Evaluación de dietas proteicas fáciles de preparar y de bajo costo utilizando levadura de cerveza, levadura de caña de azúcar y proteína de soja texturizada para *Apis mellifera* africanizada en apiarios en Santa Catarina, Brasil**

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### **Abstract**

The aim of the study was to evaluate different diets for *Apis mellifera* in three regions of Santa Catarina, Brazil. The bees received the diets weekly, for 60 days. The treatments used and the corresponding crude protein contents were: D1 – sugar syrup; D2 – commercial feed (24.4%); D3 – sugarcane yeast + commercial sugar (17.0%); D4 – beer yeast + commercial sugar (14.9%); D5 – sugarcane yeast + textured soy protein + commercial sugar (20.9%). The parameters evaluated were: measurement of the breeding areas (drone, open, closed and total), measurement of the food deposit areas (honey and pollen) and percentage of infestation by *Varroa destructor*. At the end of study, a physical-chemical analysis of the honey was carried out. Significant differences were observed when comparing protein rations with the negative control group in the variables of areas of open brood, total brood (Sierra region) and infestation by *Varroa destructor* (Santa Catarina western and Sierra region). No changes were detected in the honeys in relation to the current regulations, demonstrating that the supply of the diets, under the conditions of the study, is safe for maintaining the standards of identity and quality of the final product.

**Keywords:** Honey bees; Food; Breeding areas; Food area; *Varroa destructor*.

### **Resumo**

O objetivo do estudo foi avaliar diferentes dietas para *Apis mellifera* em três regiões de Santa Catarina, Brasil. As abelhas receberam as dietas semanalmente, durante 60 dias. Os tratamentos utilizados e os teores de proteína bruta correspondentes foram: D1 – xarope de açúcar; D2 – ração comercial (24,4%); D3 – levedura de cana + açúcar comercial (17,0%); D4 – levedura de cerveja + açúcar comercial (14,9%); D5 – levedura de cana + proteína texturizada de soja + açúcar comercial (20,9%). Os parâmetros avaliados foram: mensuração das áreas de reprodução (zangão, aberta, fechada e total), mensuração das áreas de depósito de alimentos (mel e pólen) e porcentagem de

infestação por *Varroa destructor*. Ao final do estudo, foi realizada uma análise físico-química do mel. Diferenças significativas foram observadas ao comparar as rações proteicas com o grupo controle negativo nas variáveis de áreas de cria aberta, criação total (região da Serra) e infestação por *Varroa destructor* (região oeste e serra de Santa Catarina). Não foram detectadas alterações nos méis em relação às normas vigentes, demonstrando que o fornecimento das dietas, nas condições do estudo, é seguro para a manutenção dos padrões de identidade e qualidade do produto final.

**Palavras-chave:** Abelhas melíferas; Alimentos; Áreas de reprodução; Área de alimentos; *Varroa destructor*.

### Resumen

El objetivo del estudio fue evaluar diferentes dietas para *Apis mellifera* en tres regiones de Santa Catarina, Brasil. Las abejas recibieron las dietas semanalmente durante 60 días. Los tratamientos utilizados y los correspondientes contenidos de proteína bruta fueron: D1 - jarabe de azúcar; D2 - alimento comercial (24,4%); D3 - levadura de caña + azúcar comercial (17,0%); D4 - levadura de cerveza + azúcar comercial (14,9%); D5 - levadura de caña de azúcar + proteína de soya texturizada + azúcar comercial (20,9%). Los parámetros evaluados fueron: medición de áreas de crianza (zánganos, abierta, cerrada y total), medición de áreas de depósito de alimento (miel y polen) y porcentaje de infestación por *Varroa destructor*. Al final del estudio se realizó un análisis fisicoquímico de la miel. Se observaron diferencias significativas al comparar las dietas proteicas con el grupo control negativo en las variables áreas de cría abierta, crianza total (región Serra) e infestación de *Varroa destructor* (región oeste y sierra de Santa Catarina). No se detectaron cambios en las mieles con relación a las normas vigentes, demostrando que el suministro de las dietas, en las condiciones del estudio, es seguro para el mantenimiento de los estándares de identidad y calidad del producto final.

**Palabras clave:** Abejas Melíferas; Alimentación; Zonas de cría; Zona de alimentación; *Varroa destructor*.

## 1. Introduction

Beekeeping has different forms and schemes in different parts of the world, varying their patterns over the centuries, mainly with the colonization of new regions, being modified every decade as a result of new agricultural methods, affecting the bee flora. With the technification of agriculture, rapid harvests and control of invaders are promoted even before flowering, which has directly or indirectly influenced the quantity and quality of food available to bees (Crane, 1975).

Over the years, several reports have emerged about the weakening and significant decline of honey bee colonies in Brazil, mainly in the states of São Paulo and Santa Catarina. Several factors have an influence, such as parasite infestation, climate changes, overuse of pesticides in crops and lack of appropriate food resources (Pires et al., 2016; Freitas et al., 2017).

The scarcity of floral resources at certain times can result in a nutritional deficiency of swarms. This factor considerably affects food reserves, causes a decrease in the number of offspring, an increase in cannibalism within the hives and impairs the nutrition status of subsequent generations of workers (Brodschneider, & Crailsheim, 2010; Araújo et al., 2013). Considering the lack of natural food, the supplementary diet can supply the bee's needs, in order to keep the swarms in the apiaries. In these cases, artificial feeding can be decisive in its maintenance (Lima, Silva, Soares, & Evangelista-Rodrigues, 2015; Lima et al., 2017).

Several studies have demonstrated the effective action of different supplements for bees around the world (Manning, 2018; Rizwana et al., 2019; Lamontagne-Drolet et al., 2019). However, it is necessary to provide a diet that has characteristics of nutrition similar value to those provided by a natural diet and that are viable for beekeepers (Somerville, 2005). Thus, the objective of the study was to evaluate five diets, using low-cost raw materials, which are easy to acquire and can be prepared by the beekeeper himself.

## 2. Materials and Methods

The parameters established for the evaluation of diets were adopted due to their practical importance for beekeeping. In order to assess the performance of the colonies in relation to receipt of different diets, the criteria evaluated were: open brooding area, closed brooding area, drone brooding area, total brooding area, pollen and honey deposit and degree of infestation by *Varroa destructor*. To verify the maintenance of the quality of the product obtained, as well as a possible

alteration caused by the use of diets, samples of honey from the hives treated according to the specific technical regulation were analyzed (Brazil, 2000).

## **2.1 Place of implementation of the Apiaries**

Apiaries were established in the city of São Joaquim, Rio das Antas and Florianópolis, all in the state of Santa Catarina. The locations adopted differ from each other, mainly in terms of climate, topography and geographical characteristics.

### **2.1.1 Apiary São Joaquim**

Plateau Region, Sierra Mesoregion, Campos de Lages Microregion, municipality of São Joaquim/SC: humid mesothermal temperature climate, mild summer and altitude above 800m (Ide et al., 1980). According to the agroecological zones, the average temperature is between 13.8 to 15.8 °C. The average minimum and absolute minimum temperatures for August, September and October are respectively, 6.8 °C e -10 °C; 7.6 °C and -7.5 °C; 9.2 °C and -2.4 °C. The hours of cold weather are between the months of April to October 642 e 847h and 2231 e 2808h, when considered temperature below 7.2 °C and below 13 °C, respectively. Annually, 20 to 29 frost episodes occur, with relative humidity between 79.9 and 83.4%, average annual precipitation between 1360 and 1600 mm and total annual sunshine that can vary from 1824 to 2083h (EPAGRI/CIRAM, 2009).

The vegetation is composed of mixed ombrophilous forest, savanna and araucaria forest in the Pelotas-Canoas basin. Aeras of capons, riparian forests and pine forests are found (EPAGRI/CIRAM, 2009). The predominant flowers for the production of honey are wild species, brooms (*Baccharis dracunculifolia*) and bracatinga (*Mimosa scabrella*) (EPAGRI/CEPA, 2009).

### **2.1.2 Apiary Rio das Antas**

West Region, Mesoregion West Catarinense, Joaçaba Microregion, Agroecological Zone Vale do Rio do Peixe and Central Plateau, Municipality of Rio das Antas, Santa Catarina. Midsummer humid temperature climate in mild summer. Transition area between the plateau and western regions, with an altitude of 500 to 800 meters (Ide et al., 1980). According to the classification by agroecological zones, the average temperature is between 15.8 and 17.9 °C. The average minimum temperature is between 10.8 and 12.9. The hours of cold between the months of April to October oscillate between 437 and 642h, and between 1653 and 2231h, when considered temperature below 7.2 °C and below 13 °C, respectively. Annually, 12 to 22 frosts episodes occur, the relative humidity is between 76.3 and 77.7%, with average annual precipitation between 1460 and 1820 mm, the total annual sunshine that can vary from 2137 to 2373h (EPAGRI/CIRAM, 2009).

The original vegetation of mixed ombrophilous forest is uncharacterized, predominantly agriculture with cyclic crops and secondary vegetation without palm trees, followed by montain forest (EPAGRI/CIRAM, 2009).

### **2.1.3 Apiary Florianópolis**

Coastal Region, Grande Florianópolis Mesoregion, Florianópolis Microrregion, Municipality of Florianópolis, Santa Catarina: subtropical, humid mesothermal climate with hot summer. The region has an altitude of less than 500m (Ide et al., 1980). Considering that the apiary was located on the island of Florianópolis, the altitude was below 10m. According to the classification by agroecological zones, the average temperatures is between 19.0 to 19.5 °C. The average minimum temperature is between 15.4 to 16.8 °C. Frost rarely occurs, occurring between 0.3 to 3.0 episodes for year. The relative humidity is

between 81.7 and 82.4%, the average annual precipitation between 1270 to 1600 mm and the total annual sunshine can vary from 2021 to 2166h (EPAGRI/CIRAM, 2009).

The original vegetation was composed of dense ombrophilous forest, with secondary vegetation and patches of pioneer formations predominating (marine fluvium and mangrove tree) (EPAGRI/CIRAM, 2009). At the apiary site, in addition to the wild vegetation, there was eucalyptus area (*Eucalyptus* spp.)

## 2.2 Training of experimental apiaries

In each experimental apiary, 20 Langstroth hive were implanted, constituted by a nest with 10 frames. The hives used were already with bees, however, all were uniform in terms of the number of brood combs, food, pulled wax and honeycomb wax before the experiment started.

To standardize the genetic material, the hives were orphaned and afterwards, fertilized queens, daughters of a single queen, were introduced through introduction cages. After a week, there was the release of the queen, as well as the presence of posture.

A draw was made for the formation of groups, distributing the diets for each hive. Each group was formed by four hives randomly distributed and identified by platelets with numbers. Shallow honey super were added as the need for more space was confirmed, by filling them with honey.

## 2.3 Choice, preparation and supply of diets

Initially, seven diets were tested in the laboratory and evaluated for protein concentration in the hemolymph, the weigh of the bees and the consumption of the diets (Pinto et al., 2018), using methodology adapted from Cremonz et al. (1998). Five of them were selected to be tested in the field, of these, four diets were protein and protein-free diet. As a basis in their formulations, the diets contained refined sugar cane (CS), textured soy protein (TSP), inactivated sugar cane yeast (SCY), inactivated brewer's yeast (BY) and water. The crude protein content in the dry matter (DM) of the diets was evaluated using the methodology described by Silva and Queiroz (2006) (Table 1).

**Table 1.** Percent composition and crude protein (CP) content of diets based on sugar cane (SC), textured soy protein (TSP), inactivated sugar cane yeast (SCY), inactivated brewer's yeast (BY).

Diets	Composition	Crude protein (CP)
D1*	50% SC, 50% water (v/v)	0,0 %
D2**	SCY, TSP, vitamin-mineral complex	24,4 %
D3	35% SCY, 65% SC (m/m)	17,0 %
D4	35% BY, 65% SC (m/m)	14,6 %
D5	20% SCY, 20% TSP, 60% SC (m/m/m)	20,9 %

\*Negative control diet, free of protein. \*\*Positive control diet: commercial feed. Source: Authors (2021).

To prepare the diets, the raw material was weighed on a digital scale and the mixture was made using a “Y” feed mixer. After mixing, the diets were divided into portions of 150 g, being kept in a dry environment and protected from light. The diets were provided weekly, for 60 days, in the amount of 150g per hive, in the form of “hamburgers” made by adding enough water to obtain a pasty consistency, except for the protein-free diet, which was provided in liquid syrup form, in the amount of 500 mL. Cover feeder, positioned just above the nest, was used to supply the syrup. Protein diets were placed under the cover feeder, on the nest combs.

In the apiaries located in São Joaquim and Rio das Antas, feeding started in the first week of August 2009 and in the apiary in Florianópolis in the beginning of October 2009.

#### 2.4 Measurement of breeding area and food deposit

The breeding and food reserve areas were measured in cm<sup>2</sup>. For this, a honeycomb holder made of wood was used, with 4 cm<sup>2</sup> area divisions, made with fane wire (Al-Tikrity et al., 1971) (Figure 1).

**Figure 1.** Honeycomb mapping technique according to Al-Tikrity et al. (1971).



Source: Authors (2021).

The measurement of the areas was carried out 60 days after the beginning of the supply of the diets and evaluating the following areas: open, closed and drone breeding; deposit of honey and pollen.

#### 2.5 Infestation for *Varroa destructor*

The percentage of bees infested by *Varroa destructor* was verified through the collection of adult specimens from the hives, done at random. Calculation of the level of infestation was used, according to the David de Jong Test for the diagnosis of varroase in adult bees. For this test, plastic jars of wide mouth, strainer with 4mm mesh and soapy water were used (México, 2007).

#### 2.6 Physicochemical analysis of honey

To verify the identity and quality parameters of the honey produced in the hives evaluated, physicochemical analyzes were carried out as determined by Normative Instruction No. 11, of October 20, 2000, in the Technical Regulation on Honey Identity and Quality (Brail, 2000). The verified parameters were: reducing sugars, humidity, apparent sucrose, acidity and hydroxymethylfurfural (HMF), according to the Association of Official Analytical Chemists methodology (AOAC, 1995). Acceptable levels for humidity of a maximum of 20g/100g were considered; free acidity of a maximum of 50 milliequivalents/kg; reducing sugars at least 65%; apparent sucrose of a maximum of 6g/100g; and HMF at a maximum of 60mg/kg (Brazil, 2000).

#### 2.7 Statistical analysis

The data for each of the variables were analyzed statistically by analysis of variance of Kruskal-Wallis for non-parametric data and by the Tukey test, when parametric ( $P < 0.05$ ), using the program Statistix for Windows, version 8.0 of 2003.

### 3. Results and Discussion

When evaluating the breeding areas, 60 days after the hives were uniform and the feeding started, a significant difference was observed in the open brooding area and in the total brooding area, between the protein diets and the negative control diet (D1), in the apiary of São Joaquim. In the areas of drone breeding and closed breeding there was no significant difference (Table 2).

**Table 2.** Average of the measures of the drone breeding areas, open breeding, closed breeding, total breeding area of *Apis mellifera*, in cm<sup>2</sup>, 60 days after the beginning of feeding. Apiaries São Joaquim, Rio das Antas and Florianópolis, Santa Catarina, Brazil.

Place	Diets	Drone breeding	Open breeding	Closed breeding	Total breeding
São Joaquim	D1	32 a	889 b	816 a	1737 b
	D2	53 a	2347 a	1045 a	3445 a
	D3	0 a	2258 a	1422 a	3680 a
	D4	53 a	2715 a	1173 a	3941 a
	D5	0 a	2315 a	1267 a	3581 a
Rio das Antas	D1	120 a	3532 a	2432 a	6084 a
	D2	383 a	5118 a	3296 a	8797 a
	D3	220 a	3444 a	2792 a	6456 a
	D4	320 a	6010 a	4085 a	10416 a
	D5	364 a	4914 a	4080 a	9358 a
Florianópolis	D1	238 a	4448 a	4424 a	9200 a
	D2	264 a	4160 a	4152 a	8576 a
	D3	568 a	4228 a	4788 a	9584 a
	D4	192 a	5472 a	4288 a	9952 a
	D5	272 a	5336 a	5752 a	11360 a

\*Significant difference indicated by different letters in the same column, for the same location (P<0,05). Source: Authors (2021).

In the apiaries of the municipality of Rio das Antas and Florianópolis there was no significant difference between treatments for any of measures of breeding areas. This factor may be related to the pollen deposit, which, because it is available and there is free access by the bees to it, naturally suppressing the nutritional needs of the bees, thus influencing, in the absence of statistical difference regarding the breeding areas, as also observed by Lengler (2000) and Couto (1998). For the three regions, the drone breeding area had little importance over the total breeding area, according to results also obtained by Pereira et al. (2006).

According to Farjan et al. (2015) it is necessary to include a supplementary diet in case of shortage of pollen and nectar, as in the winter season for example. Souza et al. (2019), also describe the influence of food supplementation for maintaining hives, since the correct diet can increase or decrease the reproductive potential of the queen of bees. Studies in several countries are carried out with differentiated supplementation to bees, such as Ullah, Shahzad, Iqbal, and Baloch (2021) who describe that when testing different diets, greater foraging efficiency of bees was noticed, leading to increased production and consequently greater profitability of honey, especially by the alternative soy-enriched pollen diet. With the popularization of the benefits of microalgae, Khan and Ghramh (2022) investigated different diets supplemented with the microalgae *Arthrospira platensis* (spirulina), where it was found that the consumption of bees was higher with the pollen and ajeena diet (commercially available pollen substitute), with or without spirulina, when compared to the date paste diet, with or without spirulina.

Wang et al. (2014) discuss that nutrition availability affects both the longevity and genetic expression of worker bees, which can also interfere with the routine of the hive. Retschnig et al. (2021) report that a diet rich in macro and micronutrients may be key to maintaining the individual health of bee workers. In the same way, authors describe that the lack

of nutrient can cause the increase of diseases and infestation of parasites such as *Nosema ceranae* and *Varroa destructor* (Farjan et al., 2014; Porrini et al., 2015; Dolezal, & Toth, 2018).

In the study, it was found that the infestation by *Varroa destructor* in the apiaries of São Joaquim and Rio das Antas was higher in the negative control group, when compared to protein diets, showing their efficiency for a lower rate of infestation by the mite (Table 3).

**Table 3.** Mean and standard deviation of the percentage of infestation by *Varroa destructor* mite, 60 days after beginning of feeding. Apiaries São Joaquim, Rio das Antas and Florianópolis, Santa Catarina, Brazil.

Diet	Percentage of <i>Varroa destructor</i> infestation		
	São Joaquim/SC	Rio das Antas/SC	Florianópolis/SC
D1	8,086 (2,687) a	6,96 (1,7) a	0,606 (0,393) a
D2	2,684 (2,571) b	1,35 (1,37) b	0,831 (0,240) a
D3	3,089 (1,700) b	0,80 (0,55) b	0,538 (0,530) a
D4	3,394 (0,836) b	0,31 (0,54) b	0,890 (0,310) a
D5	2,988 (1,660) b	0,73 (0,55) b	0,671 (0,450) a

\*Significant difference in the same column (P<0,05). Source: Authors (2021).

From the percentage of *Varroa destructor* infestation observed in the different municipalities it is possible to infer that the municipality of São Joaquim had higher incidences when compared with the municipalities of Rio das Antas and Florianópolis. This result may be related to the climatic differences between the regions where the experiments were installed, since in the São Joaquim apiary the climatic conditions are more severe. The low temperature, coupled with the low availability of natural food resources during the evaluated period, has been shown to affect the open brood area, total brood area, as well as increased mite infestation. Thus, the data demonstrate that in a place with unfavorable climatic conditions and in times of natural food scarcity, the benefits of artificial protein nutrition are more evident, corroborating observations made by Lengler (2000) and Couto (1998).

In the apiary of Rio das Antas, with intermediate climatic conditions between the studied regions, there was a significant reduction in infestation in the groups treated with protein diets in relation to the negative control. At the Florianópolis apiary, where the experiment was installed at a time with an abundance of natural resources and with favorable climatic conditions, no statistical difference was observed in any of the parameters studied.

According to Nazzi and Le Conte (2016), the mite can live in temperatures similar to those of the bee's nest (34-35 °C), however, it needs humidity for its reproduction. In periods of hot and dry climate there is a decrease in reproductive rates, consequently leading to the death of ectoparasite (Harris et al., 2003). The *Varroa destructor* mite is considered the main harmful parasite to bees around the world, so this evaluation is extremely necessary, considering that these cause bees to lose weight, reduce their learning ability and have a shorter life expectancy. Eventually high levels of *Varroa* infestation can lead to colony-wide varroosis, causing mass death of bees (Reams & Rangel, 2022).

In the three apiaries, no statistical difference was observed in the areas of food deposit between treatments. Assessing the differences between the pollen stored by region, it was found that in the São Joaquim apiary there is a small deposit of pollen in relation to the others. In addition to the availability of pollen, temperature is a determining factor in its collection, with a temperature of around 25 °C being considered ideal for collection and below 10 °C no bees are collecting pollen (Girou, 2003). Such differences observed in the temperature averages between the locations could explain the difference in the pollen deposit area between the regions (Table 4).

**Table 4.** Average measurements of the honey and pollen deposit areas (in cm<sup>2</sup>) 60 days after the beginning of feeding. Apiaries São Joaquim, Rio das Antas and Florianópolis, Santa Catarina, Brazil.

Diet	Honey			Pollen		
	São Joaquim	Rio das Antas	Florianópolis	São Joaquim	Rio das Antas	Florianópolis
D1	2173 <sup>ns</sup>	5128 <sup>ns</sup>	7024 <sup>ns</sup>	210 <sup>ns</sup>	1228 <sup>ns</sup>	2336 <sup>ns</sup>
D2	3776	10780	7092	304	1368	3092
D3	2082	7060	6144	450	1352	3652
D4	3674	11325	5304	232	1088	2440
D5	3664	11408	11832	442	948	3672

\*ns – not significant. Source: Authors (2021).

In the municipality of Florianópolis, larger amounts of pollen collected are perceived, varying from 2336 to 3672, whereas in Rio das Antas there is an intermediate pollen collection, with a variation from 948 to 1368. For the municipality of São Joaquim, it is possible to verify a low amount of pollen, ranging from 210 to 450. According to Barroso-Arévalo et al. (2019), the pollen area is inversely proportional to the infestation by *Varroa destructor*, which could explain the greater infestation in relation to the low availability of collected pollen and, therefore, the efficiency of protein diets administered in non-ideal conditions of floral resources.

As the pollen area is inversely proportional to the infestation, which could explain the greater infestation in relation to the low availability of collected pollen and, therefore, the efficiency of protein diets administered in non-ideal conditions of floral resources.

All the physicochemical parameters performed showed acceptable values according to the current legislation, with no changes caused by the diets provided (Table 5).

**Table 5.** Physicochemical analysis of honey collected 60 days after beginning of feeding. Apiaries São Joaquim, Rio das Antas and Florianópolis, Santa Catarina, Brazil.

Place	Diet	Humidity (%)	Free acidity (MEq.Kg <sup>-1</sup> )	Sugars reducing (%)	Apparent sucrose (%)	HMF (mg/kg)
São Joaquim	D1	17,20	19,95	82,00	1,39	1,37
	D2	18,17	17,47	82,22	1,02	0,84
	D3	17,94	19,11	79,97	1,15	1,33
	D4	18,20	17,58	79,14	0,69	1,60
	D5	17,70	18,75	80,67	0,86	0,85
Rio das Antas	D1	17,20	21,35	78,03	4,82	1,91
	D2	16,52	18,68	83,06	2,83	2,26
	D3	17,62	19,38	83,02	1,76	2,10
	D4	18,73	22,72	84,99	3,45	2,01
	D5	17,42	19,95	79,75	0,71	2,30
Florianópolis	D1	18,47	19,39	79,94	1,28	1,17
	D2	17,72	18,19	80,58	1,06	1,02
	D3	17,74	18,76	79,76	0,97	1,30
	D4	17,45	18,03	82,70	0,84	1,22
	D5	17,75	18,09	80,25	0,83	0,86

\*HMF – hydroxymethylfurfural. Source: Authors (2021).

As the hives were kept in a natural environment, the bees were free to perform the flights, thus having access to natural resources, which may be a probable cause of the variations, since the interferences of the environment can end up confusing the data, making difficult to estimate the efficiency of substitute diets (Cremonz et al., 1998). Other authors also kept bees with access to natural resources (Castagnino et al., 2006; Pereira et al., 2006), getting variable results. The influence of environmental factors, such as free access to pollen sources, can be overcome by confining small colonies without access to flowers (Herbert et al., 1977), but this procedure is expensive and time consuming (Cremonz et al., 1998).



The validation of the efficiency of diets previously evaluated in the laboratory, tested in field conditions, demonstrate that these can effectively be used in beekeeping practice, as they present positive result especially under unfavorable conditions, when it is necessary to administer them as food supplementation. The non-alteration of the physical-chemical parameters shows that the diets offered did not alter the final quality of the honeys.

#### 4. Conclusion

The effectiveness of the diets is conditioned to the natural resources available, because when they are available in nature, it becomes difficult to evaluate, due to their interference. Under the conditions in which the experiment was carried out, the protein diets evaluated can be considered efficient for the artificial feeding of *Apis mellifera* bees. Infestation by *Varroa destructor* was verified with greater intensification in the apiary in the municipality of São Joaquim, a result that may be linked to low temperatures and lack of food availability. The protein diets provided were able to reduce mite infestation. Based on the physicochemical analyzes carried out in the present work, both the sugar syrup control diet and the protein diets used in the treatments, in the quantities and conditions that were provided, did not compromise the quality of the produced honey, obeying the levels required in the specific technical regulation.

Due to the numerous environmental, climatic, and floral availability variables that occur in each region, we believe that more studies should be developed in addition to controlled laboratory studies, so that there is a real evaluation of the efficiency of protein diets that can be used in practice by the beekeepers, focusing on regional assessments and with raw materials whose costs can be defrayed by producers, taking into account the ease of acquisition of ingredients.

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