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EVALUATION OF QUALITY PARAMETERS BASED ON THE PHYSICOCHEMICAL CHARACTERIZATION OF HONEY INFORMALLY MARKETED IN REDENÇÃO-PA

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ABSTRACT

This study evaluated the physicochemical quality of honey informally marketed in Redenção-PA. Organoleptic characteristics, microscopic examination, dye test, Lund and Jagerschmidt reaction, diastatic activity, moisture content, pH, soluble solids and titratable acidity. The work used the methodology of the Pharmacognosy Manual and the Adolfo Lutz Institute. The results of the honey meet the required quality standards. The organoleptic and microscopic analyses revealed satisfactory quality, the dye tests indicated the absence of addition of artificial substances, in the Lund reaction, there was a precipitate between 2 and 3 ml, in the Jagerschmidt analysis it had an amber color with a violet tone indicating the absence of commercial sugars. The analysis of diastatic enzymes showed a brown color, attesting that the honey was not heated or adulterated. The moisture content was

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Revista ARACÊ
ISSN: 2358-2472

11%, pH 3.4, soluble solids was 82° Brix, and titratable acidity of 28 mEq/kg are within legal standards. Vitamin C had a concentration of 7.04 mg/100 g. The study reinforces the importance of complying with regulations; The producer was also offered a brief training in good handling practices, encouraging the formalization of the product, with a view to the safety of consumers and the appreciation of honey in the local market.
Using as a reference Normative Instruction No. 11, RDC No. 216 and the Municipal Inspection Seal (SIM), to ensure the safety and quality of honey by promoting formalization.

Keywords: Honey. Analyze. Quality. Tampering.

Revista ARACÊ
ISSN: 2358-2472

INTRODUCTION

Beekeeping is an activity widely explored by farmers, as it is an alternative that generates profit, needs little maintenance and low initial cost compared to other agricultural practices (Aguiar et al., 2018). It also contributes to the maintenance and preservation of existing ecosystems, and does not cause environmental impact (Costa Junior, et al., 2017). It is the practice of raising bees, with the aim of producing honey, propolis, royal jelly, pollen and beeswax (AGUIAR, 2018).

Factors such as inadequate management, lack of minimum infrastructure, and informal marketing practices can compromise the quality of honey. The lack of control over these aspects, added to the difficulty in transmitting correct information to consumers, especially about natural phenomena such as crystallization, represents significant challenges for the beekeeping sector (Silva, 2016; SEBRAE, 2009).

Honey is undoubtedly the most well-known product among those that bees offer. Since ancient times, it has been a part of human food and civilizations, not only as a nutritious food, but also as an important medicinal resource. Currently, honey is still used as food and medicine, due to its properties (Moreira; Maria, 2001).

According to the legislation, honey is a food product produced by honey bees, from the nectar of flowers or excretions of plant-sucking insects, which the bees collect, transform, combine with specific substances of their own, store and allow to mature in the combs of the hive (Brasil, 2000). Some factors can influence the composition of honey, such as the type of vegetation in the region, floral species, climatic conditions, and bee breed (Fujii; et al., 2009).

According to Pereira (2007) you can find in honey small concentrations of B vitamins, such as: B1, B2, B3, B5, B6, B8 and B9, and also vitamin C and vitamin D. Minerals are also present in a small percentage in honey, and some of the inorganic chemical elements found are: aluminum, boron, calcium, lead, chlorine, copper, sulphur, tin, iron, phosphorus, iodine, magnesium, manganese, nitrogen, osmium, potassium, radium, silicon, sodium, titanium and zinc. It should be noted that the mineral content in honey is directly related to its color and floral origin (Pereira, 2007).

Honey is a natural product with limited supply and has often been the target of adulteration, causing extreme distrust in traditional consumers, being the main barrier to the expansion of its consumption (Azaredo et al., 2003). This ends up occurring as a target

Revista ARACÊ
ISSN: 2358-2472

for the addition of adulterating substances, such as commercial sugar, causing a decrease in the quality of the product (SILVA et al., 2018).

Honey is a product of simple handling, which facilitates its adulteration, especially in contexts where inspection is insufficient or non-existent, adulterating substances compromise its quality (Gois et al., (2013), such as the addition of commercial sugar, corn syrup, molasses, invert sugar solution and glucose (Bera & Almeida-Muradian, 2007). It is necessary to perform the physicochemical analysis to prove the quality of the product (Mendes et al, 2009). Obtaining physicochemical parameters of honeys is important for their characterization as well as essential to ensure the quality of this product in the market (De Souza et al., 2021; Nascimento et al., Alves et al., 2020).

In order to avoid such fraud, Normative Instruction No. 11, of October 20, 2000, was instituted, which establishes the Technical Regulation of Honey Identity and Quality. This regulation has the function of establishing the guidelines and tests required to assess the physicochemical parameters of honey quality, verifying the moisture content, pH and acidity, reducing sugars, non-water-soluble solids, minerals and ashes, as well as the diastasis activity and hydroxymethylfurfural (HMF). All quality determination requirements are compared with the reference in force in the legislation (Brasil, 2000).

According to Andreeva (2017), bee honey is the third most adulterated food product, probably because it has high nutritional, medicinal and commercial value, with limited supply and high price. The most common adulterations are the additions of inverted sucrose solutions, glucose and corn syrups, and especially concentrated sugarcane juice. These adulterations have as their fundamental purpose the growth of profits from the sale of the adulterated product (Pinto; Lima, 2010; Calixto, 2018; Buligon et al., 2015).

In this context, this study aims to evaluate the quality parameters of honey informally marketed in Redenção-PA, based on its physicochemical characterization. Thus, it seeks to provide subsidies for the regularization and improvement of local production, ensuring compliance with current standards and promoting consumer confidence.

METHODOLOGY

The work was carried out at the Food Laboratory of Campus XV - University of the State of Pará - UEPA, located in the city of Redenção, in the southeast of the state of Pará. Apis *Meliferas bee honey* was the product evaluated for the analysis, being acquired by informal purchase in the city. The honey is extracted in a colony near the city, in the

Revista ARACÊ
ISSN: 2358-2472

neighboring municipality, Santa Maria das Barreiras, in the Continental Apiary. The criterion evaluated in the analyses was to ascertain the quality of the honey if it was in accordance with the regulatory standards and or if it had been adulterated.

PHYSICAL-QUIMIC ANALYSIS

Methodology of the Manual of the Brazilian Society of Pharmacognosy

The analyses of the honey included the evaluation of the Organoleptic Characteristics (color, aroma, flavor and texture) and Microscopic Examination to verify purity, identifying pollen grains and absence of unwanted residues. Tests such as the Dye, Lund Reaction, Jagerschmidt Reaction and Lugol Test were performed to detect adulterations, such as the addition of dyes, diluters or commercial sugar. The analysis of Diastasic enzymes evaluated heating or mixtures, while the Determination of Moisture verified liquid adulterations or premature harvest.

The following analyses followed the methodology of the Adolfo Lutz Institute

The analyses included pH, soluble solids (°Brix), titratable acidity and vitamin C. pH was measured with a digital pH meter in triplicate. Soluble solids were determined with a manual refractometer using 1 drop of honey. Titratable acidity was evaluated by titration with 0.1 M NaOH, using phenolphthalein as an indicator. Vitamin C was analyzed by titration with potassium iodate, following the method of the Adolfo Lutz Institute, with determination by the blue color point.

INTERVIEW WITH THE PRODUCER

A brief interview was conducted with the honey producer in order to identify the reasons for the absence of labeling on the product. The interview was based on the questionnaire model by Sousa, Belém and Rosa (2024), Chart 1, with adaptations to the questions, including questions about the producer's knowledge of good manufacturing practices, applicable regulations and the challenges for compliance with standards.

The interview was conducted only with the beekeeper responsible for supplying the honey.

Revista ARACÊ
ISSN: 2358-2472

RESULTS

ORGANOLEPTIC CHARACTERISTICS

The observed and felt organoleptic characteristics correspond to those described in the pharmacognosy manual about the characteristics of honey, also according to MAPA (2000). The taste was sweet with a slight acidic sensation, this is due to the presence of small amounts of formic and malic acids. The aroma was pleasant, and is characteristic of normal honey, being a satisfactory result.

Figure 1 honey on the glass plate.

Figure of the honey analyzed.

MICROSCOPIC EXAMINATION

This analysis evaluated the physical characteristics of the honey; The visualization on the slide was made in triplicate, in its general characteristics the honey did not present substances foreign to its natural composition, confirming that the product was extracted according to the parameters of good practices.

Figure 2- Microscopic Lens

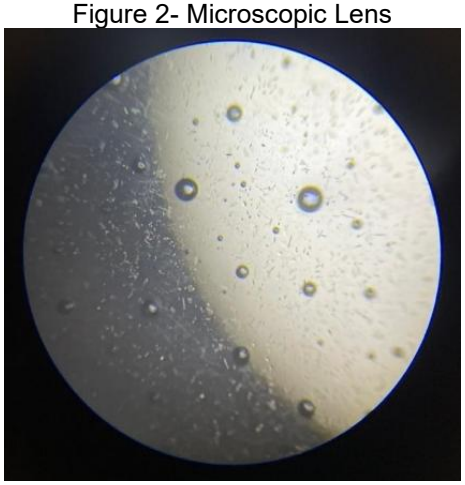


Figure without foreign substances

Revista ARACÊ
ISSN: 2358-2472

DYE TEST

The honey remained with the color unchanged; If there were dye substances added to the honey, the color would gradually change from violet to pink. Thus, the observed result of honey did not have the addition of dyes.

Figure 3- Dye Test

Figure with natural honey coloring

LUND REACTION

This analysis contributes to the identification of fraud involving the addition of water to honey, since this practice hinders the formation of precipitate (Finco; Moura; Silva, 2010). The precipitate volume of the triplicates of the honey analyzed varied between 2 ml and 3 ml, which is in accordance with the Manual of Pharmacognosy and MAPA (2000). According to these sources, a honey without the addition of water or other diluents should not produce precipitate, or present only traces.

Revista ARACÊ
ISSN: 2358-2472

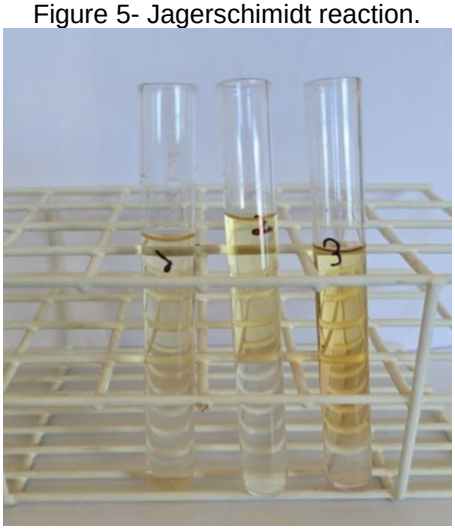
Figure 4- Lund reaction.

Figure with 3 ml precipitate.

REACTION OF JAGERSCHIMIDT

The analysis was carried out to verify whether the honey had adulteration by the addition of commercial sugar. The honey presented an amber color, which turned into violet after a certain time, indicating the absence of added sugar, this behavior is in accordance with the pharmacognosy manual, which establishes that a quality honey should not exhibit an intense violet color, which would be indicative of commercial sugar, which was not observed in the honey analysis.

Figure 5- Jagerschimidt reaction.



Amber and violet figure.

Revista ARACÊ
ISSN: 2358-2472

LUGOL ANALYSIS

Lugol's analysis showed a blue color with a negative result for adductation, considering that when sugars or starch are added fraudulently to honey, Lugol's reaction identifies the fraud, changing the color that can vary around the red color.

Figure 6- Analise Lugol.

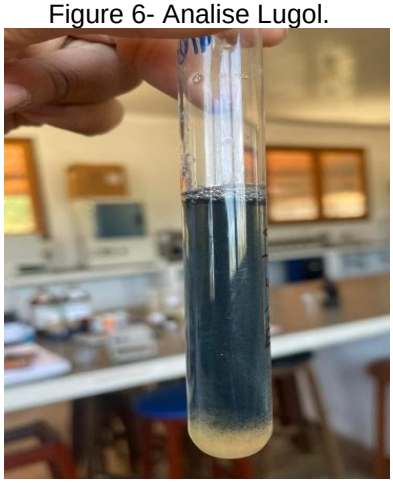


Figure with blue coloration.

DIASTATIC ENZYME ANALYSIS

Analyzing diastatic enzymes in honey is a crucial evaluation to determine the quality and purity of honey. Diastasic enzymes, also known as amylases, are enzymes that catalyze the hydrolysis of starch into simple sugars. These enzymes are naturally present in honey and are sensitive to heat; which makes it an indicator of honey adulteration, this enzyme is responsible for the breakdown of starch molecules (Dalastra et. al. 2009).

The color of the honey after going through the analysis became natural brown, which indicates that the honey did not go through boiling or addition of artificial honey, however, if the analysis resulted in the colors violet or blue the quality would be doubtful.

Revista ARACÊ
ISSN: 2358-2472

Figure 7- Diastasis Enzyme Analysis

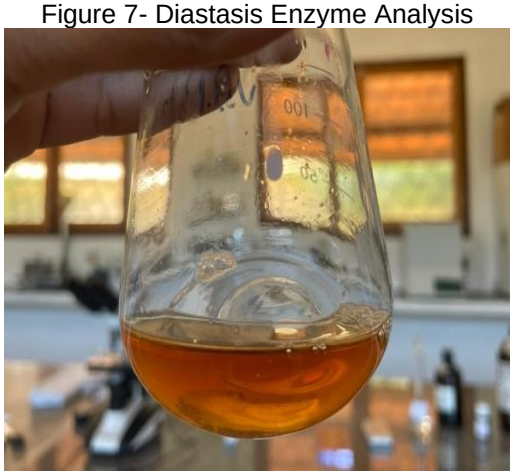


Figure with amber collar

MOISTURE DETERMINATION

The result of the analysis in percentage of water in honey was 11%; which is within the acceptable range for honey, according to the pharmacognosy manual and ABPM, which is between 8.5% and 20% (Brasil, 2000). Therefore, the honey analyzed has an adequate water content, which is evidence that it has not been adulterated or taken prematurely.

HP

The result of the ph analysis was done in triplicate, with an average of 3.4, which was great.

TOTAL SOLUBLE SOLIDS (TSS)

The result for soluble solids was 82° Brix.

TITRATABLE ACIDITY

The result for titratable acidity was 28 mEq kg^{-1}.

ASCORBIC ACID

The result found was 7.04mg/100g;

Table 2 Results of the Physicochemical Analysis of Honey

ANALYSIS	RESULT
Organoleptic characteristics	Color, flavor and aroma within the standards
Microscopic examination	No foreign substances
Dye Test	Unchanged coloration

Revista ARACÊ
ISSN: 2358-2472

Lund's reaction	Precipitate between 2-3 ml
Jagerschmidt's reaction	Amber color, no violet
Lugol analysis	Color: Blue
Diastasic Enzymes	Cor natural a pardo
Moisture Determination	11%
ph	3,4
Total Soluble Solids (TSS)	82° Brix
Titratable Acidity	28 mEq kg^{-1}
Ascorbic acid	7.04 mg/100g

Results in accordance with Normative Instruction No. 11, of October 20, 2000.

INTERVIEW WITH THE PRODUCER

In this study, when evaluating the quality of honey, a brief interview was conducted with the producer in order to understand the reasons for the absence of labeling and the SIM Municipal Inspection Seal on the product. During the interview, the producer revealed that despite having an association of beekeepers in the region, the lack of knowledge and incentive has been an obstacle to adapting to regulatory standards.

In response to this situation, a short training focused on good handling practices and the quality of honey was conducted. In addition, the main applicable regulations were clarified, as well as the benefits associated with complying with them, both for consumer safety and for the commercial valuation of the product. This incentive aimed not only to improve the compliance of the honey produced, but also to start the training of the producer, highlighting the importance of following regulatory standards to ensure quality and increase competitiveness in the market.

Table 1. Interview Questions and Answers

QUESTIONS	ANSWERS
There is an association for beekeepers	Yes
Know the types of honey contamination	No
Are you familiar with the types of adulteration of honey	No
Has knowledge about honey quality analysis	Yes
Do you know how honey labeling should be done	No
There is prejudice against honey without a label	Yes
Know Municipal Inspection Seal	No
Do you know what GMP (Good Manufacturing Practices) is	No
Are you familiar with the regulations of the Collegiate Board Resolution (RDC) for food	No
Know About Crystallization	Yes
Know the type of honeys	Yes
Meet the various species of bees	Yes
Are You Interested in Regularizing Production to Obtain Appropriate Inspection Seals and Labels	Yes
Are you open to seeking more information about regulation and quality control in beekeeping?	Yes

Adapted questionnaire table (Sousa, Belém and Rosa 2024).

Revista ARACÊ
ISSN: 2358-2472

DISCUSSION

ORGANOLEPTIC ANALYSIS

Souza's result; Rodrigues; Morais Rodrigues, (2012), were the same as those in the present work.

MICROSCOPIC EXAMINATION

In the study by Gonçalves (2019), similar results were found, as well as in the works of Souza, Rodrigues and Morais Rodrigues (2012), who reported corresponding results.

DYE TEST

Gonçalves, (2019) found the same results.

LUND REACTION

Similar results were obtained by Gonçalves (2019). Salgado et al. (2008) observed that some honey samples presented values below the standards established by the Ministry of Agriculture, Livestock and Supply (MAPA). However, analyses carried out with honeys from different blooms presented results in accordance with the normative guidelines (Souza, Rodrigues and Morais Rodrigues 2012), also found values within the standards required by the Brazilian legislation for honeys from bees. According to Souza et al. (2021), they found in their analyses meis in non-compliance with the legislation.

REACTION OF JAGERSCHIMIDT

Paim et al., (2023) also obtained the same result for the Jagerschmidt Reaction, where the analyzed samples remained in amber color, which indicates the non-addition of sugars and, therefore, no adulteration.

LUGOL ANALYSIS

Wiese (2000) finds that when using iodine and potassium iodide (lugol), adulterated honey presents a characteristic colored reaction due to the presence of starch and dextrin, which does not occur in pure honey (Coringa et. al. 2009). In the work of Silva (2017), he presented adulteration in one of the samples analyzed of industrialized honeys, characterizing it as a counterfeit product. In the work carried out by Souza et al. (2021),

Revista ARACÊ
ISSN: 2358-2472

honeys sold at fairs, some samples showed a positive result, thus being in non-compliance with the legislation.

DIASTATIC ENZYME ANALYSIS

In the work Ludwig et al. (2020) had negative results (blue color) which represents a honey without diastasic activity due to the absence or destruction of positive enzymes , some samples developed the violet color, an indication of decreased diastasic power, this happens in centrifuged honey where there is a certain heating during the process and in mixtures of natural honey with artificial honey.

MOISTURE DETERMINATION

Other results were observed by Welke et al. (2008), with moisture values ranging from 14.7 to 19.8%, higher than those found in the present study, but still within the limits established by legislation. Marchini et al. (2004) found an average value of 18.91% in analyses of Apis *mellifera honeys* in the State of Tocantins, a result similar to that obtained by Marchini et al. (2005), who identified an average moisture content of 19.1% for wild honey and 21.2% for eucalyptus honey. Humidity can be influenced by the botanical origin of the plant, by climatic and geographical conditions, or by the harvest of honey before its full maturity (Nanda et al., 2003).

HP

According to Feás, et al., (2010) the pH should be between 3.2 and 4.5, however, despite not having a reference pH value in the Brazilian legislation. Changes in pH values may indicate adulteration by fermentation (Gois et al., 2013). The pH value of honey can be influenced by the pH of the nectar, soil or association of vegetables for honey composition (Crane, 1985). Evangelista-Rodrigues et al. (2005), in an analysis of honeys in Paraíba, found values ranging from 3.8 to 4.7 in the pH of the analyzed samples.

TOTAL SOLUBLE SOLIDS (SST)

In the study carried out by Silva et al (2003), the 3 honey samples from the state of Piauí that were evaluated found Brix grade values that ranged from 76.07 to 80.80. In another study by Silva et al (2009), the mean value found was 83.28º Brix.

Revista ARACÊ
ISSN: 2358-2472

TITRATABLE ACIDITY

The value is within the quality standards recommended by the Codex Alimentarius (1993) that up to 50mEq/Kg of acidity in honey using NaOH are allowed, while by the Brazilian Legislation (Brazil, 2000) 40mEq/Kg are allowed, within the maximum limit. Salgado et al., (2008) found values with average acidity between 20.80 and 32.5 mEq/kg, in honeys of different botanical origin. In the study by MARCHINI et al. (2005) they found similar mean values of acidity, of 33.8 meq kg-1, and Azeredo et al. (2003) found a value of 34.3 meq kg-1.

According to Silva et. al. (2004) The variation in acidity can be explicit by the type of flowering, since the acidity of honey has indications in several organic acids contained in the nectar collected by bees. The variation of organic acids caused by the various sources of nectar, the enzymatic activity of glucoseoxidase that originates gluconic acid, the action of bacteria during maturation and the minerals present in its composition that influence the texture and stability of honey (Terrab, 2003).

ASCORBIC ACID

Of the vitamins, ascorbic acid (Vitamin C) is the one found in the highest concentration in honey, with about 4mg/100g of honey (Castro et al., 1998). In the honey of the zamboque bee of the work Silva et al., (2009) presented dark amber color and very high vitamin C content 203.32 mg/100g, this high value of vitamin C, due to the fact that the species and region are different from the present work.

INTERVIEW WITH THE PRODUCER

The producer was presented with some mandatory regulations to ensure the quality and safety of the honey.

Normative Instruction No. 11, of October 20, 2000**,** approved by MAPA Ministry of Agriculture and Supply, defines the Technical Regulation of identity and quality of honey, this document establishes the standards that honey intended for human consumption must meet, including classification, composition, sensory and physicochemical characteristics, as well as labeling requirements and good manufacturing practices. The regulation seeks to ensure the quality, safety and standardization of honey in the market.

The producer reported that there is still a great impasse regarding the crystallization of honey, a relevant question often raised by customers about this natural effect. Kuroishi

Revista ARACÊ
ISSN: 2358-2472

et al, (2012) describe that the crystallization of honey happens due to some factors and can vary according to them, such as the concentration of sugars, the water content in its original composition, the floral origin of the nectar, the handling during processing, processing and storage conditions, Normative Instruction No. 11, of October 20, 2000, corroborates Kuroisshi's explanation about the types of crystallization that can occur in honey, clarifying the different forms and conditions in which this process manifests itself.

The MERCOSUR/GMC/RES. No. 89/99, establishes standardized quality criteria for honey in the MERCOSUR countries. This resolution has characteristics similar to those of IN 11 of October 20, 2000 and its main objective is to eliminate trade barriers arising from differences in national regulations, ensuring compliance and product safety for consumers.

A pertinent point conveyed to the beekeeper was the issue of possible contamination of honey and the importance of Good Handling Practices. Lengler, (2001) describes that the presence of yeasts in honey occurs due to contamination due to carelessness in handling (hygiene) such as supporting honeycombs on the ground, poorly washed centrifuges, brass centrifuges, very dark combs and prolonged storage of honey in honeycombs, in view of Lengler's description, the need to present RDC N°216 of September 15, 2004 was ratified.

The guidelines of Resolution RDC No. 216, of September 15, 2004, establish standards of good practices for food services, which include requirements for the handling, storage and hygiene of food, ensuring health safety, these guidelines ensure that handling and storage are in adequate conditions, minimizing risks of contamination and preserving their natural characteristics. In addition, the resolution provides guidance on the cleaning of equipment and utensils used in the process, as well as the training of handlers, ensuring that the final product is met.

The Municipal Inspection Seal (SIM) is a certification that attests to the compliance of products of animal origin with local health standards. Its main objective is to ensure food safety, promoting the formalization of producers and ensuring that food is produced and processed in a hygienic and safe manner. To obtain the SIM, producers must comply with Good Manufacturing Practices (GMP) and undergo regular sanitary inspections, in addition to meeting the legal requirements established by the municipality. SIM brings several benefits, such as increased consumer confidence, product appreciation, and the possibility of accessing regulated markets

Revista ARACÊ
ISSN: 2358-2472

The SIM Municipal Inspection Service is aimed at the inspection of products of animal origin sold within the limits of the municipality. This service is especially important for smallholder and family farmers who sell their produce at local markets. SIM ensures that the products meet the quality and safety standards set by the municipal authorities. It is valid for civil society to encourage local industry without registration to adhere to SIM, as it is the way to get out of illegality and, in turn, to improve the competitiveness of food, increasing the standard of quality and safety of the product, due to the sanitary rigor of inspections, which follow the guidelines of technical legislation (Food Safety Brazil, 2024).

In the work by Souza et al. (2021), honey samples sold at street markets in Barreiras-BA, exhibited results with poor characteristics that were not in accordance with specific technical legislation; which indicated the need for greater quality control. However, the honey analyzed in the present study did not present these same deficiencies; however, it has resulted satisfactorily within the standards required by the Legislation; For this reason, the importance of adapting to the current inspection standards was reformulated and reinforced, ensuring more trust and credibility to customers.

Analyzing the beekeeper's responses, it is clear that he has extensive knowledge about honey in several aspects, from handling in the apiary to the bottling process. He demonstrates an understanding of the natural variations of honey, such as crystallization, types of honey, blooms, among other essential knowledge for the practice of beekeeping. However, with regard to the regulations required for the commercialization of the product, he is still unfamiliar.

At the end of the interview, it was recommended that the producer contact the association to evaluate the possibility of providing product labeling, as well as seeking the support of a professional in the area, such as a food technologist, who could offer more in-depth guidance. Given that the main objective of the interview was to provide an introduction to food safety, with a brief approach, following a simple format of questions and answers, with emphasis on the importance of labeling and compliance with Good Manufacturing Practices (GMP).

CONCLUSION

The work in question on the honey informally marketed in Redenção-PA meets the required quality standards, proving its safety and physicochemical compliance. However, the lack of labeling and the SIM Municipal Inspection Seal limits access to new markets.

Revista ARACÊ
ISSN: 2358-2472

The guidance offered to producers on good practices and the importance of regulation seeks to encourage the formalization and appreciation of the product, promoting food safety and the development of the local market.

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Revista ARACÊ
ISSN: 2358-2472

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