





Physicochemical characterization of honey bee (*Apis mellifera* L.) from the Peruvian Amazon region

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ABSTRACT: The Peruvian Amazon represents 62% of the national territory and within its area of influence there are ecosystems rich in biodiversity with great potential for honey production. The objective of the study was to determine the physicochemical characteristics of honeys from the Alto Mayo and Huallaga Central ecosystems of the San Martín region, using the Peruvian Technical Standard and AOAC. Twenty honey samples were taken from apiaries in the provinces of Rioja (Alto Mayo ecosystem) and Mariscal Cáceres (Huallaga Central ecosystem). Different physicochemical parameters were determined such as acidity (19.72 to 13.25 meq kg⁻¹), pH (4.19 to 3.96), moisture (19.06 to 17.82%), water activity (0.53 to 0.54), density (1.41 to 1.42 g mL⁻¹), reducing sugars (63.00 to 64.95%), soluble solids (73.08 to 74.18 °Brix), color (62.57 to 85.55 mm PFund), electrical conductivity (0.26 to 0.41 mS cm⁻¹), ash content (0.23 to 0.52%), hydroxymethylfurfural (14.30 to 15.0 mg kg⁻¹), and diastase index (10.54 to 10.75 °Gothe). The honey from the province of Mariscal Cáceres expressed a lighter color in relation to the honey from the Alto Mayo ecosystem.

Key words: beekeeping; ecosystem; quality; properties

Caracterización físico-química de miel de abeja (*Apis mellifera* L.) procedentes de la Amazonía peruana

RESUMEN: La Amazonía peruana representa el 62% del territorio nacional y dentro del área de influencia se encuentran ecosistemas ricos en biodiversidad de un gran potencial melífero. El objetivo del estudio fue determinar las características físico-químicas de las mieles procedentes de los ecosistemas de Alto Mayo y Huallaga Central de la región San Martín; mediante la Norma Técnica Peruana y AOAC. Se tomaron 20 muestras de miel de los apiarios de las provincias de Rioja (ecosistema del Alto Mayo) y Mariscal Cáceres (ecosistema del Huallaga Central). Se determinaron diferentes parámetros físico-químicos como acidez (19,72 a 13,25 meq kg⁻¹), pH (4,19 a 3,96), humedad (19,06 a 17,82%), actividad de agua (0,53 a 0,54), densidad (1,41 a 1,42 g mL⁻¹), azúcares reductores (63,00 a 64,95%), sólidos solubles (73,08 a 74,18 °Brix), color (62,57 a 85,55 mm PFund), conductividad eléctrica (0,26 a 0,41 mS cm⁻¹), contenido de cenizas (0,23 a 0,52%), hidroximetilfurfural (14,30 a 15,0 mg kg⁻¹) e índice de diastasa (10,54 a 10,75 °Gothe). La miel de la provincia de Mariscal Cáceres expresó un color más claro en relación a la miel del ecosistema de Alto Mayo.

Palabras clave: apicultura; ecosistema; calidad; propiedades



Introduction

According to the Instituto Nacional de Tecnología Agropecuaria de Argentina, based on FAO data, natural honey production increased its trend by 52.4% from 1990 to 2016. China, remains the main producer in the three-year periods 1994-1996, 2004-2006, and 2014-2016, followed by Turkey, United States, Argentina, Ukraine, Mexico, India, Iran, Russian Federation, Brazil and others. It is also the main exporter in relation to volume, followed by Argentina (INTA, 2018).

In the case of the American continent, honey bees have colonized due to the favorable environment for their proliferation, displacing European bee populations (Esquivel Rojas et al., 2015). Since then, honey, a natural product produced by *Apis mellifera* L., has become a highly consumed food due to its typical and sweet taste, being a product in demand in the international market due to its nutritional and therapeutic properties (García-Chaviano et al., 2022).

In Peru, the Ministerio de Agricultura y Riego reported the existence of 40,000 beekeepers working in 300,000 hives, with the largest production concentrated in the Cusco region (11%) (Minagri, 2015). On the other hand, the Peruvian Amazon has a good climate, orography, and vegetation. Is a potential environment for honey production, as revealed by Coronado Jorge et al. (2019), who found that honey (*A. mellifera* L.) from La Banda de Shilcayo has a good sensory evaluation. Also (Ormeño Luna et al., 2021), who claim that the honey (*M. eburnea*) from San Roque de Cumbaza is the sweetest, both districts of the San Martin region.

Therefore, rational beekeeping has become an activity that provides excellent economic, ecological, and social results, where the properties of honey are related to the bee flora, nectar, climate, and the species, as well as the post-harvest management by the beekeeper, which together provide quality honey. However, it has been identified that small and medium-sized producers are unaware of the relevance of this quality, restricting their productivity and failing to comply with the standards required by food regulations (Pereira Ferreira et al., 2021).

The physicochemical parameters express quantifiable results that allow estimating the quality of the honeys, for example, low moisture is related to the soluble solids content and preserve the microbiological activity that allows preserving the honey for a long time. Likewise, the light and dark color originate from the amounts of pigments derived from the flora, with dark honeys having more acidity and mineral substances (Sajid et al., 2020).

The reality of the honey trade in Peru, according to Mercado & Rimac (2019), is concentrated in the informal sector (40 to 45%), such as supply markets, fairs, etc. Since little has been discussed about the quality of honey from different wild flora and the parameters involved for its eventual export and improvement of internal and external competitiveness, this study aims to evaluate the physicochemical characteristics of honey from the Alto Mayo and Huallaga Central ecosystems of the San Martin region, which will contribute to the

establishment of the profile of honey produced in the Peruvian Amazon.

Materials and Methods

Honey samples

Twenty samples of honeys harvested between March and June 2019 were collected from two apiaries located in the provinces of Rioja (latitude 6° 3' 45" S and longitude 77° 10' 4" W) with an altitude of 848 m and rainfall of 1,300 mm year⁻¹ (Alto Mayo ecosystem), and Mariscal Caceres (latitude 7° 10' 47" S and longitude 76° 43' 38" W) with an altitude of 276 m and rainfall of 1,200 mm year⁻¹ (Huallaga Central ecosystem) (Figure 1).

Table 1 shows the average maximum and minimum temperature, relative air humidity and total rainfall recorded during the months of harvest of honeys from both ecosystems.

The Alto Mayo ecosystem is characterized by an abundant flora of native and exotic species with a high potential for honey production, such as 'tingana' (*Sapindus sapinaria*), 'guaba' (*Inga* sp.), 'caimito' (*Pouteria caimito*), 'taperiba' (*Spondias cyatherea*), 'yahuarcaspi' (*Pterocarpus ulei* Harms), 'bolaina blanca' (*Guazuma crinita*), 'huairuro' (*Ormosia coccinea*), 'coco' (*Coccus nucifera*), 'limón' (*Citrus limon*), 'plátano' (*Musa* sp.), 'mango' (*Mangifera indica*), and 'naranja' (*Citrus aurantus*). It also has a temperate subtropical climate, abundant water resources, some of which are periodic with certain reserves.

In the ecosystem of the Huallaga Central, exuberant vegetation such as 'guaba' (*Inga* sp.), 'mango' (*Mangifera indica*), 'naranja' (*Citrus aurantus*), 'limón' (*Citrus limon*), 'guayaba' (*Psidium guajava*), 'palto' (*Persea americana*), 'cocotero' (*Coccus nucifera*), and 'zapote' (*Matisia cordata*).

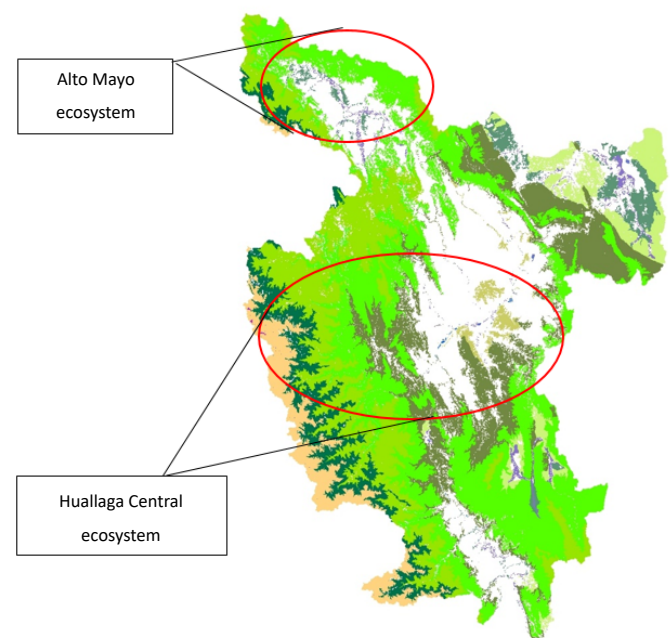


Figure 1. Geographical location of the ecosystems of the San Martín region.

Table 1. Average of climatological variables recorded.

Months	Alto Mayo ecosystem				Huallaga Central ecosystem			
	Temperature (°C)		Relative air humidity (%)	Total rainfall (mm day ⁻¹)	Temperature (°C)		Relative air humidity (%)	Total rainfall (mm day ⁻¹)
	Max	Min			Max	Min		
March	28.15	18.69	82.28	3.49	32.56	21.85	84.92	7.66
April	28.64	18.51	80.76	4.49	33.33	21.63	81.89	2.72
May	28.76	18.35	81.17	5.15	32.67	21.67	82.21	2.59
June	28.66	17.73	80.12	2.53	32.96	20.73	81.45	2.62

Source: [Servicio Nacional de Meteorología e Hidrología del Perú \(2022\)](#).

It also stands out for its herbaceous vegetation made up of grasslands with a predominance of 'brachiaria' (*Braquiaria decumbes*), 'kudzu' (*Pueraria faseoloides*), and 'pungas' (*Punga* sp.). The primary forests are characterized by their floristic variety, located in areas of steep slopes, where the possibilities of agricultural or forestry use are almost nil. There are also valuable species with melliferous capacity, such as the red quinilla (*Manilkara surinamensis*).

Physicochemical analysis

Honey moisture and water activity (Aw) were determined using the Peruvian Technical Standard - NTP-209.171-1999 ([NTP, 1999a](#)). Acidity through NTP-209.174-1999 ([NTP, 1999b](#)) and [AOAC \(2003\)](#). The pH with direct reading by inserting the combined electrode of the Lab850 benchtop pH meter, according to NTP-209.174-1999 ([NTP, 1999b](#)) and [AOAC \(2003\)](#). Color measurements were determined using the portable tristimulus colorimeter (Konica Minolta, Series II, model CR-400, 2002).

Electrical conductivity and ash were measured according to NTP-209.175-1999 ([NTP, 1999c](#)). Hydroxymethylfurfural (HMF) was by the spectrophotometric method established in NTP-209.176-1999 ([NTP, 1999d](#)). The [AOAC \(2003\)](#) and NTP-209.177-1999 ([NTP, 1999e](#)) methods were used to determine diastatic activity, which are based on the rate of hydrolysis of a 2% starch solution by diastase enzymes present in a buffered honey solution. Finally, reducing sugars were identified according to NTP-209.172-1999 ([NTP, 1999f](#)).

Results and Discussion

Acidity

The acidity values in bee honey from the Alto Mayo ecosystem ([Table 2](#)) were 19.72 ± 1.53 (meq kg⁻¹), higher than the bee honey sample from the Huallaga Central ecosystem with a content of 13.25 ± 0.89 (meq kg⁻¹). It also differs from the values reported by [Boussaid et al. \(2018\)](#), in bee honeys of 'menta' (27.03 ± 1.06), 'romero' (7.11 ± 0.20), 'eucalipto' (26.60 ± 0.20), 'toronjil' (27.20 ± 0.20), 'tomilo' (26.20 ± 0.20), and 'naranja' (21.41 ± 0.10) from northern Tunisia. The different results from both ecosystems and from other latitudes corroborate the influence of the type of flora on the total acidity of honey.

On the other hand, from the results of free acidity, none of the samples analyzed from Alto Mayo and Huallaga Central exceeded the limit of 40 meq kg⁻¹, complying with the Peruvian Technical Standard - NTP209.174-1999 ([NTP,](#)

Table 2. Physicochemical parameters of honeys.

Parameters	Ecosystems	
	Alto Mayo	Huallaga Central
Acidity (meq kg ⁻¹)	19.72 ± 1.53 A	13.25 ± 0.89 B
pH	4.19 ± 0.22 A	3.96 ± 0.18 A
Moisture (%)	19.06 ± 1.48 A	17.82 ± 1.94 A
Water activity (Aw)	0.53 ± 0.0 A	0.54 ± 0.0 A
Density (g mL ⁻¹)	1.42 ± 0.01 A	1.41 ± 0.0 A
Reducing sugars (%)	63.00 ± 1.43 A	64.95 ± 1.36 A
Sólidos solubles (°Brix)	74.18 ± 0.56 A	73.08 ± 1.56 A
Color (mm PFund)	85.55 ± 1.04 A	62.57 ± 1.01 B
Electrical conductivity (ms cm ⁻¹)	0.26 ± 0.01 A	0.41 ± 0.05 B
Ash content (%)	0.52 ± 0.01 A	0.23 ± 0.01 B
Hydroxymethylfurfural (mg kg ⁻¹)	15.0 ± 0.0 A	14.30 ± 0.84 A
Diastase index (°Gothe)	10.75 ± 0.05 A	10.54 ± 0.26 A

Different capital letters on the same line indicate significant differences ($p < 0.05$) between the means of physicochemical parameters of honeys from different ecosystems.

[1999b](#)), Mexican Official Standard - NMX-F-382-S ([Dirección General de Normas, 2008](#)), and Codex Alimentarius - CSX 12-1981 ([Codex Alimentarius, 2001](#)). Honey from Alto Mayo and Huallaga Central do differ significantly in acidity values.

pH

The pH of the Alto Mayo honey was slightly higher at 4.19 ± 0.22 compared to the Huallaga Central honey with a pH of 3.96 ± 0.18 ([Table 2](#)). These values were very close to acacia honeys of 3.99 ± 0.14 , 'pradera' of 3.95 ± 0.12 , and 'girasol' of 3.88 ± 0.25 from the Autonomous Province of Vojvodina (Republic of Serbia) reported by [Sakač et al. \(2019\)](#).

The pH is a fundamental parameter for the extraction, texture, stability, and shelf life of honey. In addition, it is related to the type of honeys according to their origin, if it expresses a pH that fluctuates between 3.30 and 4.60, it is a honey of floral origin. According to NTP209.174-1999 ([NTP, 1999b](#)), the pH values of honey from Alto Mayo and Huallaga Central are within the range of 3.40 - 6.10, therefore, they are within the national standard. Likewise, there is no significant difference.

Moisture content

The average moisture content in the honeys studied ([Table 2](#)) was higher in the Alto Mayo honey with 19.06 ± 1.48 (%), compared to the Huallaga Central honey with 17.82 ± 1.94 (%). These results are very similar to those reported by [Rodríguez-Flores et al. \(2019\)](#) in bee honeys of black oak floral origin from Spain in the years 2009, 2010, and 2011, with moisture values of 17.0 ± 0.8 , 17.6 ± 0.8 , and 18.1 ± 0.8 , respectively.

Moisture content varies with soil and climatic conditions, geographical and botanical origin and season of production and may further vary during honey storage. On the other hand, the moisture content values of honeys remain within international standards despite the diverse geographical regions with a range between 11 and 20% (Nascimento et al., 2018).

In addition, the moisture values of honey from Alto Mayo and Huallaga Central coincide with the maximum values allowed by the Peruvian Technical Standard - NTP209.171-1999 (NTP, 1999a), Mexican Official Standard - NMX-F-382-S (Dirección General de Normas, 2008), and with CSX12-1981 (Codex Alimentarius, 2001). The results of the study between Alto Mayo and Huallaga Central honey showed no significant differences for moisture.

Water activity (A_w)

Both honeys from Alto Mayo and Huallaga Central (Table 2) showed a very close value of water activity (A_w) 0.53 ± 0.0 and 0.54 ± 0.0 , respectively. Water activity is a crucial factor that determines the amount of water available for the survival and growth limitation of microorganisms that cause food spoilage by fermentation or enzymatic activity. This is corroborated by Bobis et al. (2020), who mention that the water activity (A_w) ranges of honeys are generally between 0.49 and 0.65. The water activity results show no significant differences between Alto Mayo and Huallaga Central honey.

Density

The density of the Alto Mayo honey with 1.42 ± 0.01 g mL⁻¹ was very similar to the density of the Huallaga Central honey 1.41 ± 0.0 g mL⁻¹. The density of both honeys (Table 2) was lower than the range (1.55 ± 0.01 - 1.71 ± 0.01) reported by Aumeeruddy et al. (2019) in eucalyptus and commercial honey. The variation observed among honeys could be due to geographical origin, climatic conditions, floral source, and storage conditions. The honey values under study are within the range allowed by NTP209.168-1999 (NTP, 1999e), which establishes that this parameter should be in the range of 1.4 and 1.6 g mL⁻¹ at 20 °C. The results of the honeys showed no significant differences for density.

Reducing sugars

The average value of reducing sugars in the Alto Mayo honey (Table 2) was $63.00 \pm 1.43\%$, a lower content than the Huallaga Central honey of $64.95 \pm 1.30\%$. These contents were lower than the Ethiopian honeys (68.55%) reported by Tigistu et al. (2021). The lower content of reducing sugars in the Alto Mayo honey could be due to the higher moisture content present. This decreases the solid component of the honey resulting in a lower content of reducing sugars. In addition, the difference between honey sources in terms of reducing sugar content could be due to variation in the plant sources from which the honey was produced.

The values of reducing sugars of the honeys studied are within the ranges established by NTP209.172 -1999 (NTP,

1999f), NMX-F-382-S (Dirección General de Normas, 2008), and CXS12-1981 (Codex Alimentarius, 2001), which establish that this parameter should be at 65.00% of reducing sugar. The results showed no significant differences for reducing sugars.

Soluble solids (%)

The value of soluble solids in the Alto Mayo honey was $74.18 \pm 0.56\%$, slightly higher than the Huallaga Central honey of $73.08 \pm 1.56\%$ (Table 2). These values are lower than those reported by Nemo & Bacha (2021) with honey from Koli, Shebel, Yali, and Yati of the Amphilo-Ethiopia district ranging from 82.85 ± 0.63 to $84.8 \pm 0.33\%$. Soluble solids content is correlated with moisture and with sugars, particularly glucose, and fructose. Sugars in honeys are an excellent source of energy and a great antimicrobial activity due to the high osmotic pressure, acidity, hydrogen peroxide (H₂O₂), and non-peroxide components such as methylglyoxal.

The soluble solids values of the honeys studied were within the ranges established by NTP209.171-1999 (NTP, 1999a), which establishes 75% (°Brix) as an acceptable soluble solids value. No significant differences were observed for soluble solids.

Color

Color ranged from 85.55 ± 1.04 mm Pfund for the Alto Mayo honey and 62.57 ± 1.01 mm Pfund for the Huallaga Central honey in the Pfund classifier, corresponding to amber to light amber, respectively (Table 2). These color values of the studied honeys are higher than those reported by Ramos et al. (2018) such as eucalyptus, purple flower, and trefoil based honey of 49.25 ± 8.39 mm Pfund (extra light amber), eucalyptus, purple flower, and sunflower honey of 48.71 ± 7.43 (extra light amber) and eucalyptus, black broom, and purple flower honey of 53.0 ± 000 mm Pfund (light amber).

The color is determined mainly by its botanical origin, as well as depending on the ash content and the temperature at which the honey is in the hive and the storage time. In addition, the color of honey is due to coloring matters in the nectar of the floral source from which it has been released by the bee (Al-Farsi et al., 2018). It was observed that there were significant differences for color among the honeys studied.

Electrical conductivity

The electrical conductivity for the Alto Mayo honey had a value of 0.26 ± 0.01 mS cm⁻¹, much lower than that of the Huallaga Central honey of 0.41 ± 0.05 mS cm⁻¹ (Table 2). While the electrical conductivity values in samples of chestnut honeys from Turkey reported by Demir Kanbur et al. (2021) ranged from 0.18 to 0.38 mS cm⁻¹. Electrical conductivity is a function of salt content and is used to differentiate nectar honey from honeydew honey. In addition, when compared to NTP209.174-1999 (NTP, 1999b), the results of the study are within the standard. Significant differences were found for the Alto Mayo and Huallaga Central honey samples for electrical conductivity.

Ash content

Regarding mineral content (Table 2), the ash content of the Alto Mayo honey was $0.52 \pm 0.01\%$, much higher than the ash content of the Huallaga Central honey of $0.23 \pm 0.01\%$. These values are very close to the results for nectar molasses honey ($0.5727 \pm 0.05\%$) and heather honey ($0.2111 \pm 0.003\%$), respectively, found by Popek et al. (2017). The mineral content of each honey variety is differentiated by the type of mineral the soil may contain. Darker honeys have higher acidity and higher content of mineral substances.

On the other hand, this can be corroborated by NTP209.175-1999 (NTP, 1999c) which indicates as maximum limit 1% ash content, which points out that the value of ash content in honey is generally small and depends on the composition of the nectar of the predominant plant that contributes to the formation of honey. Significant differences were observed between the two types of honey studied.

Hydroxymethylfurfural

The Alto Mayo bee honey had a slightly higher value of hydroxymethylfurfural of $15.00 \pm 0.0 \text{ mg kg}^{-1}$ relative to the Huallaga Central bee honey sample of $14.30 \pm 0.84 \text{ mg kg}^{-1}$ (Table 2). The closest values in the study were those reported by (Sajid et al., 2020) with fresh honeys ranging from 24.25 to 40.68 mg kg^{-1} . Hydroxymethylfurfural is a fundamental parameter for measuring purity, freshness of honey, and preservation conditions or heat treatment. On the other hand, storage and floral source are the predominant factors in obtaining higher hydroxymethylfurfural (Codex Alimentarius, 2001).

The hydroxymethylfurfural contents of the honeys under study showed very low values for NTP209.176-1999 (NTP, 1999d), which establishes that the limit should be 80.00 mg kg^{-1} of HMF, since our country has a biodiversity of climates such as subtropical climates where honeys can naturally have a higher content without having been overheated or adulterated. No significant differences were demonstrated for hydroxymethylfurfural.

Diastase index

The Alto Mayo bee honey sample expressed a diastase index of 10.75 ± 0.05 °Gothe slightly higher compared to the Huallaga Central bee honey sample of 10.54 ± 0.26 °Gothe (Table 2). These values are close to those reported by Bentabol Manzanares et al. (2014) with avocado honey (14.5 ± 8.3), heather honey (14.5 ± 6.1), and pitera honey (11.5 ± 3.7). Diastase activity expresses the freshness of honey and can also be used as an indicator of poor manufacturing practices.

The diastase values of the honeys under study were found to have a high diastase index, with respect to that established by NTP209.171-1999 (NTP, 1999a), which also coincides with that established by NMX-F-382-S (Dirección General de Normas, 2008), and CSX 12-1981 (Codex Alimentarius, 2001), which indicate a maximum diastase index limit of 8.00 °Gothe. It is likely that the samples could have been affected by different factors such as harvest time, storage time, and heterogeneity

in storage conditions. There were no significant differences between the two honeys under study for diastase index.

Conclusions

In this study, two bee honeys from two different ecosystems of the Peruvian Amazon region, Alto Mayo, and Huallaga Central in the department of San Martín, were characterized. Most of the parameters showed no significant differences between them. However, in the physicochemical characteristics of pH, color, electrical conductivity, and ash content, statistical differences were expressed.

Honey from the Alto Mayo is lighter than honey from the Central Huallaga, as reflected in the values of color, electrical conductivity, and ash content. Most of the physicochemical parameters of the bee honey samples from the Alto Mayo and Central Huallaga ecosystems are within the parameters established and required by national and international regulations.

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Compliance with Ethical Standards

Author contributions: Conceptualization: ASC, MFCJ, JOL; Data curation: ASC, MFCJ, PVR; Formal analysis: ASC, MFC, ETG, EMN; Methodology: ASC, MFCJ, JOL, PVR; Project administration: ASC; Resources: PVR, ETG; Supervision: EMN; Validation: ASC, PVR; Writing – original draft: ASC, MFCJ, JOL, ETG, EMN, PVR; Writing – review & editing: ASC, MFCJ, JOL, ETG, EMN, PVR.

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