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Functional, rheological and sensory properties of a food from honey and *aloe vera*

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The consumption of honey products has increased in recent years, largely due to the pandemic caused by SARS-CoV-2 (COVID-19). The objective of this work was to evaluate the functional, color, rheological and sensory properties of honey (H) and honey mixtures with *aloe vera* (AV). Among the functionality evaluated, the ability to stabilize DPPH (2,2-diphenyl-1-picrylhydrazyl) and ABTS (2,2'-Azino-bis(3-thylbenzothiazoline-6-sulfonic acid)) radicals was determined, as well as the reducing capacity and the content of phenolic compounds. Rheological behavior was analyzed by flow and oscillatory tests. Finally, product acceptance was analyzed by means of a sensory panel. The results showed that honey, AV and the mixtures presented antioxidant activity between 52.5 and 57 mg trolox/g sample, (DPPH method), between 1,666 and 1765 µg trolox/g sample (ABTS method), between 217 and 506 µg trolox/g sample (FRAP method) and between 62.5 and 164.6 mg GAE/100 g sample (phenolic compounds in gallic acid equivalents, GAE). The values obtained were in the order of those reported in the literature for these products. Rheological tests showed a pseudoplastic flow type in the honey and AV mixtures, and a Newtonian tendency for the honey. In addition, the oscillatory tests showed a dependence of the oscillation frequency on G' and G." It can be concluded that the mixtures of honey and AV maintain functionality in terms of antioxidant properties after processing and are also accepted by consumers in terms of taste, smell, color and texture.

KEYWORDS

antioxidant activity, color, flow, viscoelasticity, functionality

1. Introduction

In recent years, the production of foods that provide some kind of functionality or health benefits are being increasingly demanded by the population. The functionality of food products or ingredients has been associated with the presence of phytochemicals and micronutrients such as vitamins, which contribute to the ability to complex free radicals or to generate a reducing environment in the organism (Yan et al., 2020). In particular, after the worldwide emergency caused by the SARS COV-2 coronavirus, several investigations on the effect of natural honey-based products on the prevention or treatment of respiratory diseases are being carried out (Lima et al., 2021). Honey is a natural product, rich in energy and with a low

glycemic index, produced by bees and with a wide biological activity. Studies on this subject have shown that it can improve physical performance at moderate levels of activity, which may result in a synergistic interaction between the consumption of this product and exercise (Ali et al., 2021). Likewise, other studies have found that many of the direct and indirect medicinal properties of honey against COVID 19 are related to the content of phenolic compounds and their antioxidant action (Al-Hatamleh et al., 2020). In addition, many honey producers are seeking to expand the diversity of their products to meet consumer demand. In general, these products are based on mixtures with other products derived from beekeeping, such as propolis and pollen, but other options could also be analyzed with vegetable products such as AV, which has health benefits and is sensorially well accepted.

AV is a perennial plant with turgid green leaves. The parenchyma cells contain a transparent mucilaginous gelatin which is called AV gel. Different studies have shown that the bioactive components of AV have an anti-inflammatory effect and promote lipid and carbohydrate metabolism, helping to maintain normal blood sugar and cholesterol levels and proper body weight (Heś et al., 2019). Preliminary work determined that hydrothermal treatments of AV allowed obtaining particles suitable for addition to passion fruit and panela-based beverages (Nieto and Suarez, 2020). In addition, to our knowledge, products made from honey and AV have not been studied in terms of their functional or rheological properties. In view of the above, the objective of this work was to determine physicochemical, antioxidant, rheological and sensory properties of foods produced from honey and AV gel.

2. Materials and methods

2.1. Materials

Honey from the *Apis mellifera scutellata* species, produced by beekeepers in the municipality of La Vega Cauca Colombia and packaged by the APIMACIZO Company, was used for this study. On the other hand, AV of the *Aloe barbadensis* Miller type was used, which was processed as previously described in Nieto and Suarez (2020). Briefly, the edges of the leaves were cut, approximately 1 cm from the border. Then, the outer green epidermis was separated in order to obtain the inner pulp of the leaf. The pulp was cut into cubes of approximately 1 cm³ to be subjected to hydrothermal treatment. Subsequently, the cubes were submerged in water at 20°C for 10 min, and then, three washes were performed with hot water at 55°C for 10 min each, using a water:AV ratio of 3:1. Finally, the pulp cubes were broken down to obtain the AV gel, by means of a Braun Multiquick, Mq5000 manual homogenizer. In addition, xanthan gum (XG) and Carboxymethyl cellulose (CMC) were used as viscosifiers agents.

2.2. Reagents

Deionized water (MilliQ™, United States) was used for all assays. Chemical reagents were of analytical grade. All reagents used as standards for calibration curves were from SIGMA-Aldrich (St Louis, MO).

2.3. Production of *aloe vera* honey mixtures

Different preliminary trials were carried out to establish the mixing percentages of honey and AV, taking into account the consistency of the product and sensory acceptability (data not shown). Blends of honey:AV were made in ratios of 70:30, 75:25 and 80:20, homogenizing the mixtures for 10 min in a laboratory mixer (Velp, F201A0155, Italy) at 300 rpm. XG and CMC were used to improve the consistency of the product, at a ratio of 1 and 4 g per kg of product, respectively. Finally, the product was placed in 100 cm³ glass containers and subsequently underwent rapid pasteurization, 72°C, 15 s (Schvezov et al., 2020).

2.4. Antioxidant properties

In order to evaluate the functionality of honey, AV and mixtures, determinations of antiradical capacity, reducing power and phenolic compound content were carried out. These determinations were performed on extracts prepared with acetone, water, hydrochloric acid and the samples based on honey and AV.

2.4.1. Preparation of extracts to determine antioxidant activity

Extractant solution was prepared with acetone:water in a ratio of 70:30, with a final concentration of 0.1% HCl (Palavecino et al., 2019). The extracts were prepared by mixing 0.9 g of each sample with 10 mL of the extractant solution for 30 min at 37°C and 250 rpm. After this time, the extracts were filtered using Whatman N°1 filter paper and subsequently analyzed for each spectrophotometric determination.

2.4.2. Anti-radical capacity

The antiradical capacity was determined spectrophotometrically by two analytical methods, the 2,2-diphenyl-1-picrylhydrazyl free radical (DPPH) method and the 2,2'-Azino-bis(3-thylbenzothiazoline-6-sulfonic acid) radical cation (ABTS) method.

For the DPPH method, the spectrophotometric method of Brand-Williams et al. (1995) was used with some modifications. Briefly, a 6×10⁻⁵ M solution was prepared with the free radical (DPPH) in 96 mL/100 mL ethanol. Different aliquots of each extract were mixed with the DPPH solution and allowed to react for half an hour in the dark. After this time, the absorbance was read at 515 nm. The results were expressed as mg trolox/g sample.

On the other hand, for the ABTS method, the procedure of Wootton Beard et al. (2011) was considered. Briefly, ABTS solution was mixed with each extract. The mixture was left in a thermostatic chamber at 37°C for 30 min in the absence of light and the absorbance at 729.7 nm was recorded. The results were expressed as μg trolox/g sample.

2.4.3. Reducing power

The reducing capacity was determined according to Basanta et al. (2014). Briefly, FRAP reagent was mixed with the extracts and left in a chamber at 37°C in the dark for 30 min. After this time, absorbance was measured at 595 nm. The results were expressed as μg trolox/g sample.

2.4.4. Phenolic compounds

Phenolic compounds were determined according to Shui and Leong (2006). Briefly, the reaction was performed by mixing the extracts with Folin Ciocalteu solution and after 5 min with NaHCO₃ solution. After 60 min in the dark, the absorbance was measured at a wavelength of 765 nm. The results were expressed in milligram gallic acid equivalents per 100 g of product (mg AGE/100 g).

All spectrophotometric determinations to evaluate antioxidant activity were performed at least in triplicate.

2.5. Rheological analysis

Honey and honey-based products were analyzed for flow and viscoelasticity. For both tests a rheometer AR 1500, TA Instruments, New Castel, United States, equipped with a 25 mm plate-plate geometry was used.

2.5.1. Flow tests

Flow assay was carried out at a constant temperature of 25°C in the 0.01 and 100 s⁻¹ shear rate range. The experimental data were fitted to the Power law equation (Eq. 1).

$$\tau = k \cdot \dot{\gamma}^n \quad (1)$$

Where τ represents the shear stress, k the consistency index, $\dot{\gamma}$ the shear rate, and n is the flow index. A Newtonian flow type was determined for values of $n = 1$, pseudoplastic for $n < 1$ and dilatant for $n > 1$. Moreover, the apparent viscosity values for a shear rate of 50 s⁻¹ (η_{50}) were used to compare the different products.

2.5.2. Oscillatory assays

An amplitude sweep test was performed at a fixed frequency of 1 Hz and at 25°C to determine the linear viscoelasticity range (LVR) in each product. Subsequently, a frequency sweep was performed at a constant stress to obtain the mechanical spectra of the products in terms of the storage modulus G' and loss modulus G'' and the variation of the angular frequency.

Additionally, the storage modulus G' (Pa) data were fitted to the power law gel model described by Sun et al. (2020) (Eq. 2).

$$G' = \Gamma(1 - n') \cos n\pi / 2 S' \omega^{n'} \quad (2)$$

Where $\Gamma(1 - n')$ is the gamma function for $1 - n'$. The constants S' and n' were used as gel strength and exponent, respectively to model the storage modulus $G'(\omega)$. All rheology analyses were performed at least in triplicate.

2.6. Color analysis

The Lab* coordinates established by the Commission Internationale de l'Éclairage (CIE) were determined in order to establish the differences in ΔE color between honey and AV-added products. The analysis was carried out on a CM-5 spectrophotometer (Konica Minolta, Inc., Osaka, Japan), using Illuminant D65 and an angle of observation of 10° (Shamsudin et al., 2019). Color measurements were performed in triplicate.

2.7. Sensory evaluation

A sensory test was conducted to determine the acceptance and preference of the honey and AV blends. The trial was carried out with the participation of 131 untrained volunteer panelists, between 21 and 55 years of age and regular consumers of products such as honey or its derivatives. In the trial, the parameters texture, color, taste and smell were evaluated using a 5-point hedonic scale, representing: (1) I dislike it very much; (2) I dislike it; (3) I neither like nor dislike it; (4) I like it; (5) I like it very much. Panelists were instructed to eat a cracker and rinse their mouth with water between each sample to avoid carryover effects (Genevois et al., 2018).

For each parameter evaluated, the average score was calculated and additionally the acceptance rate (AR) was calculated according to the equation:

$$AR (\%) = (x / 5) \cdot 100 \quad (3)$$

Where x is the value rated by the panelists for each parameter evaluated (Lucas et al., 2018).

For the entire sensory test, the recommendations established by Lawless (2012) were taken into account. In addition, the studies involving human participants were reviewed and approved by Ethics Committee of Fundación Universitaria de Popayan. Likewise, written informed consent from the participants was not required to participate in this study in accordance with the national legislation and the institutional requirements.

2.8. Statistical analysis

The comparison of the results obtained in each of the properties studied was carried out by means of an analysis of variance ANOVA followed by a Tukey's post-test to identify significant differences. The modeling of the rheological analysis data and the statistical analyses were performed with GraphPad Prism version 7.00 for Windows (GraphPad Software, San Diego California United States).

3. Results and discussion

3.1. Antioxidant properties

It was observed that both honey and AV and blends had antioxidant activity (Table 1). The results showed that honey had a greater antiradical and reducing capacity ($p < 0.05$) than AV. On the other hand, as for the mixtures, it was found that they had the same ($p < 0.05$) antiradical capacity as AV by the (DPPH) method and higher ($p < 0.05$) by the ABTS method. Likewise, the mixtures presented greater reducing capacity (FRAP) than the AV. Regarding the content of phenolic compounds, it was found that honey had a higher content ($p < 0.05$) than AV and the blends. The higher content ($p < 0.05$) of phenolic compounds in honey could explain, to a large extent, the results observed in the analysis of antioxidant activity. Although it is important to highlight that in honeys, other compounds with antioxidant activity have been reported, such as B-complex vitamins and ascorbic acid (Da Silva et al., 2016). As for phenolic compounds in honey, vanillic acid, caffeic acid, syringic acid,

TABLE 1 Antioxidant properties of honey and AV products.

Sample	DPPH mg trolox/g sample	ABTS μ g trolox/g sample	FRAP μ g trolox/g sample	Phenolic compounds mg GAE/100 g sample
Honey	57 \pm 1 ^a	1765 \pm 13 ^a	506 \pm 37 ^a	164.6 \pm 0.9 ^a
AV	53 \pm 1 ^b	1,666 \pm 27 ^b	217 \pm 24 ^d	62.5 \pm 0.8 ^c
Honey-AV 70:30	53.4 \pm 0.2 ^b	1755 \pm 8 ^a	403 \pm 6 ^c	134 \pm 1 ^d
Honey-AV 75:25	52.9 \pm 0.5 ^b	1750 \pm 10 ^a	446 \pm 3 ^{bc}	138.5 \pm 0.5 ^c
Honey-AV 80:20	52.5 \pm 0.6 ^b	1736 \pm 9 ^a	474 \pm 9 ^{ab}	145 \pm 1 ^b

Average and standard deviation are reported ($n = 3$). Different letters in the same column mean significant differences ($p < 0.05$). AV means *aloe vera*.

TABLE 2 Color properties of honey-based products.

	L*	a*	b	ΔE
Honey	28.85 \pm 0.02 ^d	2.76 \pm 0.03 ^a	5.19 \pm 0.02 ^d	–
Honey-AV 70:30	33.13 \pm 0.04 ^a	1.7 \pm 0.2 ^b	10.8 \pm 0.1 ^a	7.13 \pm 0.05 ^a
Honey-AV 75:25	31.8 \pm 0.4 ^b	1.4 \pm 0.2 ^b	9.62 \pm 0.05 ^b	5.49 \pm 0.03 ^b
Honey-AV 80:20	29.9 \pm 0.2 ^c	0.59 \pm 0.02 ^c	9.2 \pm 0.2 ^c	4.67 \pm 0.04 ^c

Average and standard deviation are reported ($n = 3$). Different lowercase letters mean significant differences ($p < 0.05$). AV means *aloe vera*.

p-coumaric acid, ferulic acid, quercetin, kaempferol, myricetin, pinobanksin, pinocembrin, chrysin, ellagic acid, galangin, 3-hydroxybenzoic acid, chlorogenic acid, 4-hydroxybenzoic acid, rosmarinic acid, gallic acid, hesperetin, benzoic acid and others, have been reported (Trautvetter et al., 2009; Alvarez-Suarez et al., 2012).

Comparing the results with those reported in the literature, it was found that they were in the order of those found by Guldás et al. (2022), who in honey (*Apis mellifera anatolica*) quantified values for antiradical activity of 1950 μ g trolox/g sample (ABTS method) and for phenolic compounds of 35 mg GAE/ 100 g. Likewise, in AV gel used for the production of energy drinks with passion fruit and panela, values of 57 mg GAE/100 mL have been reported (Nieto and Suarez, 2020). AV is considered an interesting product for the formulation of functional products, due to its contribution of bioactive compounds, vitamins, minerals, enzymes, simple/complex polysaccharides, phenolic compounds, organic acids (Hamman, 2008) and antioxidant properties (González-Delgado et al., 2023).

It is important to highlight that honey and honey-based products are desired by many consumers for their benefits in the treatment of viral and other diseases. For example, it has been reported that honey polyphenols have shown an interesting therapeutic potential for regulating the production of inflammatory mediators and decreasing the production of reactive oxygen species, ROS (el-Seedi et al., 2022). In this sense, glutathione peroxidase (GPx), superoxide dismutase (SOD), vitamin E, carotenoids, vitamin C, flavonoids and phenols present in AV may be responsible for the antioxidant properties (Kumar et al., 2019).

3.2. Color analysis

When we talk about honey, it could be said that color is the first attribute observed during its commercialization, and therefore, an important parameter of quality and consumer acceptance. In terms of color analysis, L* values indicate the lightness, a* the redness and b* the yellowness of the sample. Regarding lightness values, it could be observed that the addition of AV tends to increase ($p < 0.05$) the L*

values, slightly lightening the products, in relation to honey. As for the a* and b* coordinates, it could be observed that the values in the products are reduced with the addition of AV. Likewise, the greatest color change ($p < 0.05$) with respect to honey occurred with the addition of 30% AV. On the other hand, in studies carried out by (Boussaid et al., 2018), values were reported for L* between 36.64 and 51.37 for a* between -0.67 and 4.41 and for b* between 6.06 and 17.67 , attributing the differences in the honeys to the different botanical and floral origins. It can be said that the values found for the color parameters in this work are of the order of those mentioned by these authors (see Table 2).

3.3. Rheological characterization

3.3.1. Flow assay

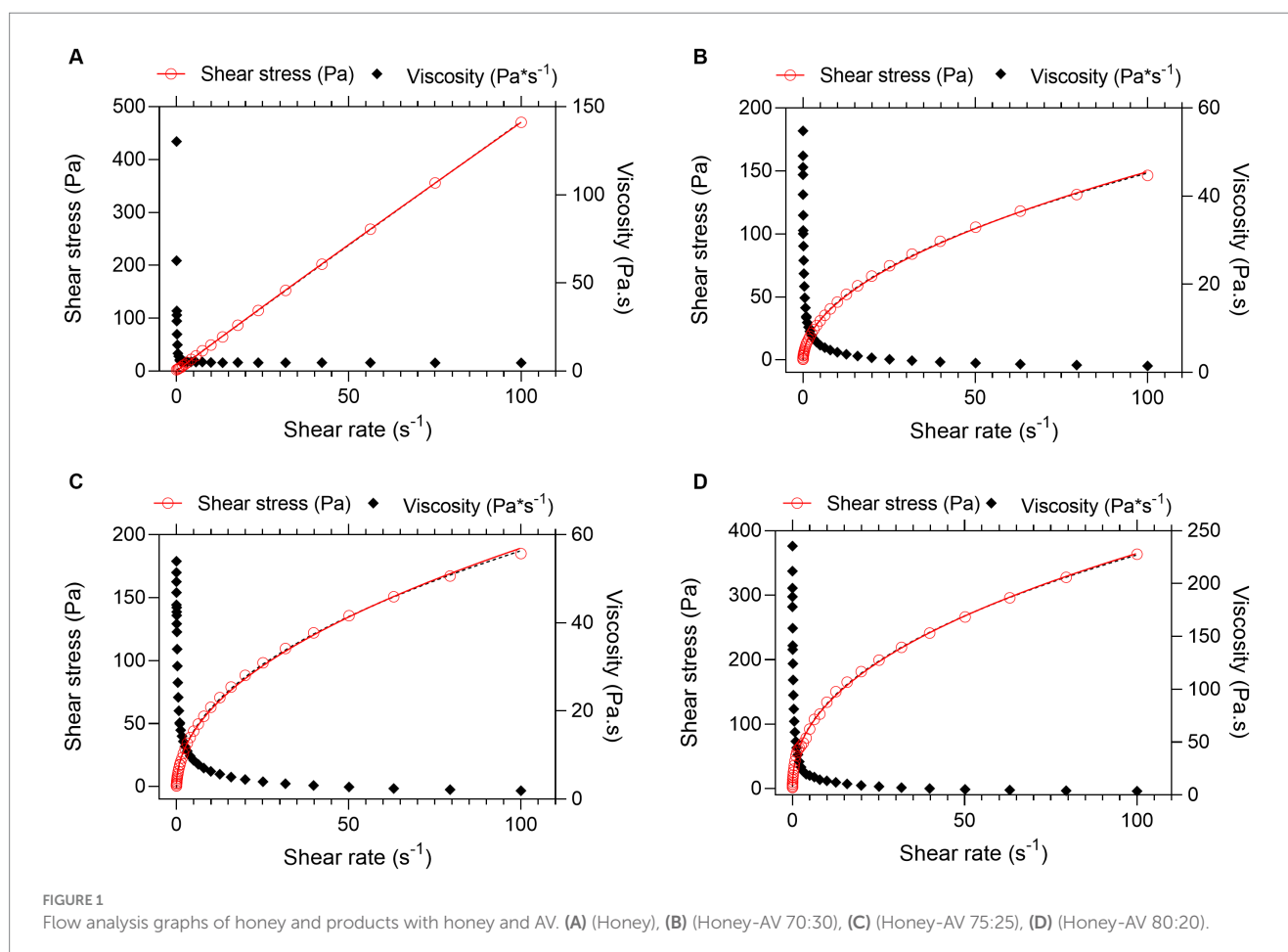
The viscosity of honey and mixtures of honey and AV were analyzed. The η_{50} value was taken to compare the results of this property (Table 3). It was found that honey-AV mixtures 70–30 and 75–25 had a significantly lower viscosity ($p < 0.05$) than honey. On the other hand, the honey-AV 80–20 mixture had the highest viscosity value at this shear rate point.

Also, Figure 1 shows the flow behavior of honey and the mixtures. As for the honey (Figure 1A, red line), a linear flow trend was observed, of Newtonian type, however, when analyzing the viscosity graph (black line), a reduction of its viscosity could be verified as the shear rate increased. Likewise, when analyzing the flow index value (n), a value of 0.975 was found (Table 3) which, being less than 1, denotes a pseudoplastic flow behavior, but it can also be said that it is a value very close to 1, a value corresponding to a Newtonian flow. This same flow behavior has been reported by other authors in the scientific literature. For example, Gómez-Díaz et al. (2006) reported values of n between 0.933 and 0.982 , in honeys from Galicia, which are of the order of the value found in this work. Likewise, as for the value of the consistency index (K) of the honey, it is within the values that have been reported in other works, such as those of Trávníček and Přidal (2017), who found values between 8.4 and 65.4 Pa.sⁿ, in honeys of different origins.

TABLE 3 Results in terms of mean viscosity analysis, and power law fit of the flow data.

	η_{50} (Pa.s)	K (Pa.s ⁿ)	n	R ²
Honey	4.778 ± 0.004 ^b	5.28 ± 0.04 ^d	0.975 ± 0.003 ^a	0.9998
Honey-AV 70:30	2.103 ± 0.002 ^d	13.69 ± 0.03 ^e	0.519 ± 0.001 ^b	0.9992
Honey-AV 75:25	2.708 ± 0.003 ^c	19.59 ± 0.07 ^b	0.494 ± 0.002 ^c	0.9985
Honey-AV 80:20	5.310 ± 0.002 ^a	46.62 ± 0.05 ^a	0.447 ± 0.001 ^d	0.9988

Average and standard deviation are reported ($n=3$). AV means *aloe vera*. Different lowercase letters mean significant differences ($p < 0.05$).



As for the mixtures (Figures 1B–D), a pseudoplastic curvature is observed in the red lines. Also in the black lines a reduction of viscosity with shear rate. Additionally, Table 3 shows that the values of n for the three mixtures were less than 1, which denotes a pseudoplastic flow. The determination of the rheological behavior of materials is of great importance during the design of processing lines, pumping or storage equipment, quality control and in the development of new products (Fischer and Windhab, 2011; Faustino and Pinheiro, 2021). Finally, it was possible to verify that the power model was adequate to model the flow data, honey and mixtures, obtaining R^2 values above 0.9984.

3.3.2. Oscillatory studies

The rheological properties of the honey-AV-based gels were influenced by both the concentration of AV used and the oscillatory frequency. The results showed that the gels had viscoelastic liquid

characteristics, with a loss modulus (G'') higher than the storage modulus (G') over the entire frequency range analyzed.

Honey is a viscoelastic product, which denotes both elastic and viscous properties when deformed. Honey has a higher resistance to flow than other products with Newtonian tendency such as milk, causing it to flow more slowly. When the rheological properties of honey were analyzed at frequencies between 1 and 100 rad/s, the results showed that the viscous property prevails over the elastic property, since the loss modulus (G'') was higher than the storage modulus (G') (Figure 2A). However, a slight elastic property was also observed, so it could be said that honey is not a simple Newtonian fluid, but a “weakly” viscoelastic fluid. This same rheological behavior was also reported in the work of (Witczak et al., 2011) in heather honey. Figure 2A also shows that the elastic modulus (G') increases with frequency for both modules, increasing by two orders of magnitude between 1 and 100 rad/s. Likewise for the honey-AV

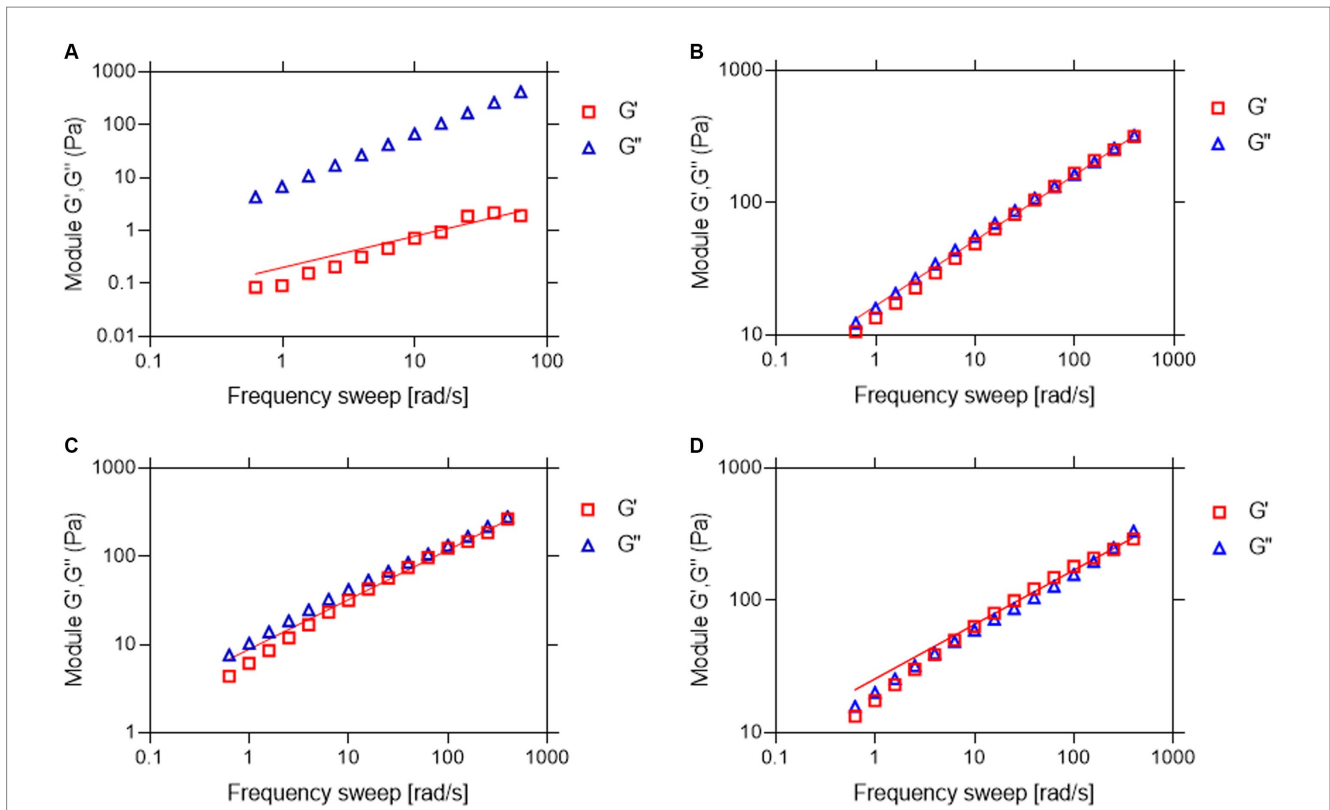


FIGURE 2 Mechanical spectra of honey and honey-AV mixtures, fitted with the power-law gel model. (A) (Honey), (B) (Honey-AV 70:30), (C) (Honey-AV 75:25), (D) (Honey-AV 80:20).

TABLE 4 Results in terms of gel index (n) and gel strength (s) of honey and products by power-law model fitting.

	Gel strength (Pa.s ⁿ)	Gel index (n')	R ²
Honey	0.1541 ± 0.009 ^d	0.59 ± 0.02 ^a	0.9998
Honey-AV 70:30	6.9 ± 0.4 ^c	0.56 ± 0.01 ^a	0.9992
Honey-AV 75:25	13.6 ± 0.9 ^b	0.494 ± 0.009 ^b	0.9985
Honey-AV 80:20	24 ± 2 ^a	0.406 ± 0.007 ^c	0.9988

Average and standard deviation are reported (n=3). AV means *aloe vera*. Different lowercase letters mean significant differences (p < 0.05).

mixtures, a high oscillatory frequency dependence and more similar G' and G'' components were also found over the frequency range analyzed (Figures 2B–D). The modulus (G'') is directly related to ω, as expected for Newtonian fluids. For this analysis, it could be said that the addition of AV, XG and CMC to honey caused the viscoelastic moduli to approach each other, which could be explained by the presence of the AV polysaccharides or thickeners.

Using the power gamma function model, the parameter S' was estimated to represent the gel strength. When comparing the S' values of honey and the honey:AV 70:30 mixture (Table 4), an increase in gel strength could be observed, which is in agreement with what was seen in Figures 2B–D, where an increase in the elastic component was observed. On the other hand, although for the other mixtures the values of n' were not statistically equal (p < 0.05), a tendency of increase in gel strength with the reduction of the percentage of AV was observed. Finally, the model used to describe the data had R² values greater than 0.9985.

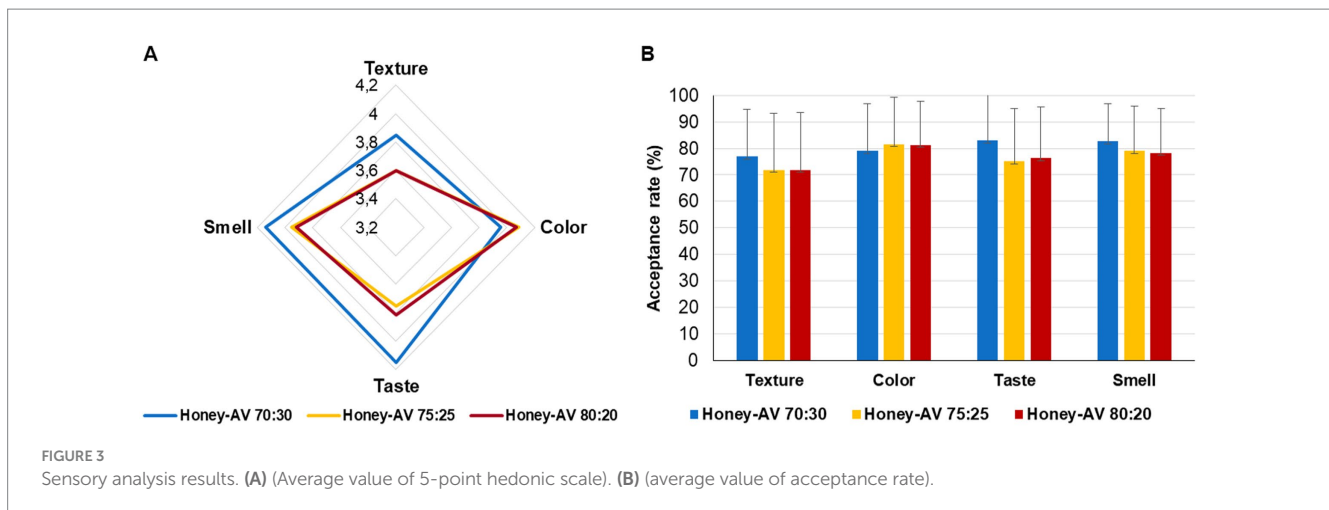
3.4. Sensory analysis

The results of the sensory analysis showed that all three products tasted were accepted with a rating above 3 or an acceptance rate above 70% for all parameters (Figures 3A,B). It has been established that for a product to be accepted with respect to its sensory characteristics, it must have an acceptance rate of over 70% (Lucas et al., 2018). In this work, the honey:AV 70:30 mixture had a significantly higher rating (p < 0.05) in terms of taste (%AR). However, among the other sensory parameters, no significant differences were detected between the products (p < 0.05). Furthermore, it can be said that, although the variability of the responses ranged between 2 and 5, the rating with the highest frequency (mode) in the data set was 4, corresponding to a perception of: I like it, for all the samples and sensory properties evaluated.

The results obtained in this work allowed us to verify that the mixture with the lowest percentage of honey and the highest AV was the one most preferred by the panelists in terms of taste. However, with respect to the other sensory parameters, no significant differences (p < 0.05) were found in the preference of the products. Therefore, producers and consumers should take into account the content of bioactive compounds and the antioxidant and rheological properties demonstrated.

4. Conclusion

Products based on honey and AV were produced to evaluate their functionality and sensory acceptance. The study concluded that all the



products showed antiradical and reducing capacity, largely explained by the presence of phenolic compounds from both honey and AV.

On the other hand, it was also possible to conclude that the products obtained showed pseudoplastic type flows, while honey has a more Newtonian tendency. Also, both honey and the blends showed a dependence on the oscillation frequency in terms of the G' and G'' moduli. Furthermore, mixing higher percentages of AV together with XG and CMC tended to improve the gel strength of the products. Finally, the addition of AV to honey, in percentages of 20, 25 and 30% were sensorially accepted by consumers. In addition, the honey-AV 70–30 mixture was the one that presented the best acceptance in terms of taste. Possibly, a greater replacement of honey with AV makes the product less sweet and more acceptable to consumers. These results are promising for honey and AV producers and allow to visualize the rheological behavior of the products for the design of possible production equipment.

Data availability statement

The raw data supporting the conclusions of this article will be made available by the authors, without undue reservation.

Ethics statement

The studies involving human participants were reviewed and approved by Ethics Committee of Fundación Universitaria de Popayan. Likewise, written informed consent from the participants was not required to participate in this study in accordance with the national legislation and the institutional requirements.

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Author contributions

CS-R and SV-P: experimental development and data analysis. CM-H: statistical analysis. DR-A: application of mathematical rheological models, writing revision and editing. JN-C: conceptualization, study propose, supervision and revision of the manuscript. All authors contributed to the article and approved the version submitted.

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Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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