

## CHARACTERIZATION AND PRODUCTION OF TRADITIONAL MEAD AND MEAD FERMENTED WITH COCOA NIBS (*Theobroma cacao* L.)

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### ABSTRACT

Mead, a globally consumed alcoholic beverage, has added value in the market due to the use of honey (*Apis mellifera*) in its composition. On the other hand, cocoa (*Theobroma cacao* L.) is a product utilized in the cocoa industry due to its antioxidant properties and flavor. The aim of this study is to compare traditional mead (H0) produced with water, honey, and *S. cerevisiae*, and mead (H1) with mixed fermentation of honey and cocoa. The production of the beverages complied with the Brazilian quality standards established by the Ministry of Agriculture, Livestock, and Food Supply (MAPA). In terms of concentration of bioactive compounds, the mead fermented with cocoa nibs (H1) showed superior results compared to mead (H0), with concentrations of polyphenols (mg Eq. Gallic acid/100g) 69.75 (H0) and 89.64 (H1), reduction of free radicals ( $\mu\text{mol Eq. Trolox/g}$ ) 6.42 (H0) and 8.88 (H1), acidity (mg acetic acid/100g) 67.8 (H0) and 69.1 (H1), and alcohol content (%) 4.59 (H0) and 5.25 (H1). The mead with nibs presented distinct characteristics in its flavor, influenced by acidity and alcohol content, as well as providing an enhancement of phenolic and antioxidant compounds, contributing to the added value and quality of the drink in the market.

**Keywords:** Bioactive. Compounds. Characterization. Cocoa. Mead.

### 1 INTRODUCTION

Mead is known as the oldest alcoholic beverage in humanity<sup>1</sup> with high added value in the market<sup>2</sup>, its production consists of honey, water, and yeast through a fermentative process, varying the alcohol content according to the beverage's production<sup>3</sup>. The alcoholic beverage has a sweet flavor and aroma, often containing a golden-yellow to amber coloration, directly associated with the floral origin of the chosen honey for its production<sup>4</sup>. The honey from the *Apis mellifera* bee can vary its sensory characteristics and bioactive compounds, reflecting the diversity and richness of biodiversity depending on the region in which it is found<sup>5</sup>; however, its identity and quality must comply with Brazilian standards for honey and derivatives for consumption<sup>6</sup>. Mead production can also involve the use of fruits and spices during its fermentation and/or afterwards for flavoring in a second fermentation, enhancing the phenolic compounds and distinct sensory characteristics of the beverage, as each one has different acidity, sugars, and phenolics in its composition<sup>7</sup>. Cocoa nibs (shell- and germ-free cocoa bean) from *Theobroma cacao* L. possess bioactive compounds and antioxidant capacity greater than in teas and red wine<sup>8</sup>, such as methylxanthines, epicatechin, polyphenols, and anthocyanins, and a wide variety of volatile compounds responsible for the astringent and bitter taste of the beans<sup>9</sup>, obtained after several stages and predominantly used for chocolate formulations. The aim of this study is to produce and compare traditional mead with mead fermented from honey and cocoa nibs of *Theobroma cacao* L. to find unique sensory characteristics and enhance the bioactive compounds of the fermented beverage with the presence of cocoa nibs during fermentation.

### 2 MATERIAL & METHODS

For quality control, the honey (*Apis mellifera*) used was analyzed based on pH, free acidity (mEq/kg), moisture (g/100g), total soluble solids ( $^{\circ}\text{Brix}$ ), reducing sugars (%)<sup>10</sup>, total polyphenols (mg Eq. Gallic acid/100g)<sup>11</sup>, and DPPH ( $\mu\text{mol Eq. Trolox/g}$ )<sup>13</sup>, while pH tests, total acidity (mg acetic acid/100g)<sup>10</sup>, DPPH ( $\mu\text{mol Eq. Trolox/g}$ )<sup>13</sup>, and total polyphenols (mg Eq. Gallic acid/100g)<sup>11</sup> were conducted on the cocoa beans (*Theobroma cacao* L.) used for the fermentation of one of the beverages. Subsequently, two meads were prepared for the artisanal production of sweet mead<sup>12</sup>. All equipment was sterilized, and for the formulation of the must of 500 mL, 189.30 g of honey (*Apis mellifera*) and 310.70 mL of mineral water adjusted to 32 degrees on the  $^{\circ}\text{Brix}$  scale were measured, pasteurized at 65 °C for 30 minutes, cooled in a water bath to a temperature of 25 °C, and added to the fermenter with an airlock. For the yeast starter, 0.5 g of *Saccharomyces cerevisiae*/L of must was used, which was hydrated with 5 mL of water at 38 °C for 20 minutes, homogenized, and inoculated into 10% (v/v) of the must when the temperature difference between them reached a maximum of 10 °C. For one of the fermented beverages, 15g of cocoa nibs (*Theobroma cacao* L.) were added. The fermenters were stirred for aeration, sealed, and kept sheltered from light and at room temperature to ferment for 21 days. At the end of fermentation, the meads were filtered, bottled, and stored under refrigeration. For beverage quality control, aliquots were taken from the mead and flavored mead for analysis of total soluble solids (TSS) on the  $^{\circ}\text{Brix}$  scale, pH, acidity (mEq/L), alcohol content (%)<sup>10</sup>, and quantification of total polyphenols (mg Eq. Gallic acid/100g) and DPPH ( $\mu\text{mol Eq. Trolox/g}$ )<sup>13</sup>. For the sensory characterization, 30 individuals were subjected to sensory analysis to characterize the flavor of traditional mead and mead with cocoa nibs.

### 3 RESULTS & DISCUSSION

The results obtained for the quality of the honey (Table 1) used for the production of mead remained within the quality standards established by the Ministry of Agriculture, Livestock, and Supply (MAPA), with parameters for floral honey being a maximum of 50 mEq/kg for acidity, 20 g/100g for moisture, and 60 mg/kg of hydroxymethylfurfural (HMF), and a minimum of 65 g/100g of reducing sugars<sup>6</sup>. Regarding the quality of the final product, both the traditional mead and the one fermented with cocoa nibs showed satisfactory results (Table 1), which enter within the standard of identity and quality of fermented beverages with total acidity values between 30-150 mEq/L and alcohol content between 4-14% v/v at 20°C<sup>14</sup>. Initially, the worts were at 32 °Brix, and the reduction of this to 23.43 (H0) and 22.03 (H1) indicates the consumption of sugars present by the yeasts and their conversion into alcohol and other byproducts<sup>15</sup>. The pH of 3.61 for the honey is within the recommended range of 3.3 to 4.6<sup>16</sup>; for the beverage, the value of 3.45 for the mead and 3.66 for the mead flavored with cocoa nibs (*Theobroma Cacao L.*) indicates the growth of *S. cerevisiae* yeasts and contributes to the inhibition of bacterial growth, favoring the fermentation of the beverage and providing an acidic taste<sup>17</sup>.

**Table 1** Results regarding the physicochemical characterization of Honey (*Apis mellifera*), Mead (H0), Mead fermented with cocoa nibs (H1), and cocoa nibs (*Theobroma Cacao L.*)

	Free Acidity (mEq/kg)	Total Acidity (mEq/L)	Acidity (mg Acetic Acid/100)	pH	HMF (mg/kg)	Moisture (g/100g)	Total Soluble Solids (°Brix)	Reducing Sugars (%)	Alcohol Content (%)
Honey	43.93 ± 0.05	-	-	3.61 ± 0.5	40.52 ± 0.74	19.65 ± 0.06	79.23 ± 0.58	70.03 ± 1.5	-
H0	-	67.8 ± 0.001	-	3.45 ± 0.01	-	-	23.43 ± 0.05	-	4.59
H1	-	69.1 ± 0.005	-	3.66 ± 0.01	-	-	22.03 ± 0.05	-	5.25
Nibs	-	-	5.48 ± 0.002	5.10 ± 0.03	-	5.73 ± 0.15	-	-	-

In regard to the values of phenolic and antioxidant compounds found in *Apis mellifera* honey at 150.15 (mg Eq. Gallic acid/100g) and 0.07 (µmol Eq. Trolox /g) compared to cocoa nibs at 380.43 (mg Eq. Gallic acid/100g) and 3.50 (µmol Eq. Trolox /g) for the production of mead, the significant difference in the concentration of these compounds in honey and *Theobroma cacao L.* nibs is directly related to the content of bioactive properties found in the final product. The mead made only with water, honey, and yeast showed 69.75 (mg Eq. Gallic acid/100g) of polyphenols and 6.42 (µmol Eq. Trolox/g) of antioxidant activity. In contrast, the mead flavored with 15g of cocoa nibs during the beverage fermentation process showed elevated results with 89.64 (mg Eq. Gallic acid/100g) of total polyphenols and 8.88 (µmol Eq. Trolox/g) of antioxidant activity (Table 2). These results characterize mead fermented with nibs as a functional alcoholic beverage, since it contains higher levels of polyphenols and antioxidants compared to regular mead.

**Table 2** Results regarding the Total Polyphenols (TP) and 2,2-diphenyl-1-picrylhydrazyl (DPPH) of Honey (*Apis mellifera*), Mead (H0), Mead fermented with cocoa nibs (H1), and cocoa nibs (*Theobroma Cacao L.*)

	TP (mg Eq. Gallic acid/100g)	DPPH (µmol Eq. Trolox /g)
Honey	150.15 ± 0.005	0.07 ± 4.35
Nibs	380.43 ± 0.01	3.50 ± 0.26
H0	69.75 ± 0.001	6.42 ± 3.32
H1	89.64 ± 0.002	8.88 ± 5.32

In the sensory test conducted with 30 individuals, the comparative study between traditional mead (H0) and mead fermented with cocoa nibs (H1) highlighted significant differences in the sensory characteristics of both. In the case of traditional mead (H0), 56.67% of individuals highlighted the beverage containing a strong sweet aroma and flavor, while 60% emphasized moderate acidity. This can be attributed to the predominance of natural sugars from honey in its composition, as well as its acidity level related to *Apis mellifera* honey and H0 (Table 1). In contrast, mead fermented with cocoa nibs (H1) was distinguished by a mild astringency and acidity. Approximately 40% of individuals characterized it as having a higher astringency, while 50% highlighted a mild acidic flavor and less sweetness. These characteristics are likely the result of including *Theobroma cacao L.* nibs at the onset of fermentation, which directly influenced the unique sensory characteristics of the beverage made with a combination of honey and cocoa nibs.

## 4 CONCLUSION

Both meads demonstrate conformity with the standards established for the production of alcoholic beverages in Brazil, as specified in Normative Instruction No. 34 of the MAPA. However, the mead made with fermented cocoa fruit (*Theobroma cacao* L.) almonds (nibs) stands out by presenting higher acidity and alcohol content compared to traditional mead, reporting flavor nuances. Additionally, the addition of nibs to the mead resulted in an enhancement of bioactive compounds such as polyphenols and antioxidant activity, revealing higher concentrations compared to conventional mead. This significant increase not only enriches the sensory experience of the beverage but also poses it as a valuable choice within the functional alcoholic beverages segment.

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