Characteristic Properties of Kahramanmarash Honey Samples

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ABSTRACT: The characteristic properties of 50 different floral honey samples in Kahramanmarash were evaluated as pH, ash, electrical conductivity, diastase activity, free acidity, 5-hydroxymethylfurfural (HMF) and water insoluble solids. The mean values for pH, ash, electrical conductivity, diastase activity, free acidity, HMF and water insoluble solids were 3.92, 0.34%, 0.48 mS/cm, 17.6, 37.23 meq/kg, 39.28 mg/kg, and 0.089%, respectively. The levels of 10 oligosaccharides of 50 honey samples were also studied. The maltose levels were ranged between 1.55-3.99%. Sucrose, isomaltose were ranged between 0.05-0.76 and 1.19-0.70%, respectively, Turanose, nigerose, melibiose and panose were ranged between 0.76-2%, 1-2.5%, 0.04-0.14%, 0.03-0.09%, respectively. Maltotriose, melezitose and raffinose were present with the values of 0.22-1%, 0.2-0.3% and 0.1-0.21%, respectively.

Key words: Honey, Oligosaccharides, Characteristic properties.

INTRODUCTION

Honey is one of the most complex, wholesome, viscous and aromatic food prepared by bees, mainly from the nectar of flowers or honeydew and certainly the only sweetening agent that can be used by humans without processing. Composition of honey is affected by contributions of the plant, climate, environmental conditions and the ability of the beekeeper. The diversity of the physical and chemical properties of honey depends on the nectar and pollen of the original plant, colour, flavour, and contents of moisture, protein and sugars (White and Maher, 1980).

Honey characterization is based on the determination of its chemical, physical or biological properties. Studies have shown that honey has both antimicrobial and antioxidant properties, useful in stimulation of wounds and burns healing and gastric ulcers treatment (Al-Somal et al., 1994; Gheldof and Engeseth, 2002). Physical and chemical, sensory properties of different kinds of honey have been reported by various scientists (Persano Oddo et al., 1995; Yılmaz and Yavuz, 1999; Bath and Singh, 1999; Cano et al., 2001; Anupama et al., 2003).

HMF measurement is used to evaluate the quality of honey; generally not present in fresh honey, its content increases during conditioning and storage. Heating of unifloral honey leads to different HMF levels in honey (Fallilo et al., 2004). HMF is formed during acid-catalysed dehydration of hexoses and, it is connected to the chemical properties of honey, like pH, total acidity, mineral content (Anam and Dart, 1995; Bath and Singh, 1999).

Honey is produced by bees mainly from nectar of flowers or honeydew. Surveys of floral honey composition have established that fructose and glucose are the major carbohydrates, ranging from 65-80% of the total soluble solids (Doner, 1977; Costa et al., 1999). Besides these sugars, other minor carbohydrates, chiefly di- and trisaccharides containing glucose and fructose residues, the levels of disaccharides and trisaccharides have been identified (Siddiqi and Furgala, 1967; Siddiqi and Furgala, 1968; Low and Sporns, 1988; Swallow and Low, 1990). Some studies have been carried out in order to investigate the origin of the honey oligosaccharides. Maurizio (1975), studying the carbohydrates of nectar of the majority of plant families visited by honeybees, noted that variable amounts of sucrose, glucose and fructose were present therein. Other sugars, such as raffinose and melezitose, were mainly found in honeydew, a sweet liquid secreted by some species of plant-sucking insects, which is gathered by bees during periods of low nectar availability (Lombard et al., 1984). Weston and Brocklebank (1999) and Da Costa Leite et al. (2000), also studied oligosaccharides in honey samples. An oligosaccharide is a saccharide polymer containing a small number (typically three to ten) of component sugars, also known as simple sugars.

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The aim of this study was to determine characteristic properties and oligosaccharide contents of Turkish honeys in Kahramanmaraş City (southeast).

**MATERIALS and METHODS**

**Honey samples**

The present study was carried out using 50 honeys of blossom origin from southeast Turkey (Kahramanmaraş city), the samples were collected from beekeepers in 2004 summer. The samples were stored at 4°C in darkness prior to analysis. Before analysis, crystallized honeys were defrosted at 30°C and homogenized by gently stirring thoroughly for 3 min. All samples were analysed in three triplicates.

**Characteristic parameters**

pH was measured by a pH-meter (Jenco 6231) in a solution containing 10 g honey in 75 mL of CO₂ free distilled water (AOAC, 1990). The free acidity, electrical conductivity, HMF, ash contents, diastase activity and insoluble solids were determined as in TS 3036 (Turkish Standard, 2002).

**Oligosaccharide contents**

A 5% (v/v) honey solution was prepared and diluted with acetonitrile (1:1, v/v), than the mixture were centrifuged for 1 min in Heraeus centrifuge (İncelkara, Türkiye). Each sugar standard was dissolved in acetonitrile:water (1:1, v/v) at a concentration of 2 mg/mL. Analyses were carried out in an HPLC system with a pump (Perkin Elmer, Tetra, Türkiye), an injection valve of 20 μL loop and a refractive index monitor.

Separation of oligosaccharides was on a Lichrosphere 5-NH₂ (250x4 mm, i.d., Merck) column using acetonitrile:water (85:15, v/v) as mobile phase and of flow rate of 1 ml/min. Each standard was injected separately. Also both the standard mixture and a honey sample were spiked with each individual sugar to confirm the identity of each oligosaccharide. Quantification was obtained by peak height comparison with standard of oligosaccharides (Da Costa Leite et al., 2000). Undetectable levels have been considered as zero for calculation purposes. Methanol, acetonitrile and the oligosaccharide standards were purchased from Sigma (USA). Sucrose (α-D-glucopyranosyl-β-D-fructofuranoside), isomaltose [O-α-D-glucopyranosyl-(1→6)-D-glucopyranose], maltose [O-α-D-glucopyranosyl-(1→4)-D-glucopyranose], turanose [O-α-D-glucopyranosyl-(1→3)-D-fructose], nigerose [O-α-D-glucopyranosyl-(1→3)-D-glucopyranose], melibiose [O-α-D-galactopyranosyl-(1→6)-D-glucopyranose], panose [O-α-D-galactopyranosyl-(1→6)-O-α-D-glucopyranosyl-(1→4)-D-glucopyranose], maltotriose [O-α-D-galactopyranosyl-(1→4)-O-α-D-glucopyranosyl-(1→4)-D-glucopyranose], melezitose [O-α-D-galactopyranosyl-(1→3)-β-D-fructofuranosyl-(2→1)-α-D-glucopyranoside], raffinose [O-α-D-galactopyranosyl-(1→6)-O-α-D-glucopyranosyl-β-D-fructofuranoside]. Results were expressed as g % for each sugar.

**RESULTS and DISCUSSION**

The characteristic properties and oligosaccharide contents of honey samples are summarized in Table 1.

<table>
<thead>
<tr>
<th>pH</th>
<th>Ash (%)</th>
<th>ec² (mS/cm)</th>
<th>Diastase activity (ID)</th>
<th>free acidity (meq/kg)</th>
<th>HMF (mg/kg)</th>
<th>Water insoluble solids (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>min</td>
<td>3.63</td>
<td>0.004</td>
<td>0.04</td>
<td>8.0</td>
<td>15.76</td>
<td>31.46</td>
</tr>
<tr>
<td>max</td>
<td>4.41</td>
<td>0.65</td>
<td>1.13</td>
<td>29.4</td>
<td>51.3</td>
<td>40.70</td>
</tr>
<tr>
<td>mean</td>
<td>3.92</td>
<td>0.34</td>
<td>0.48</td>
<td>17.6</td>
<td>37.23</td>
<td>39.28</td>
</tr>
</tbody>
</table>

**Characteristics of Honey Samples**

Taking into account that honey is a complex natural food obtained under conditions which are difficult to control, an unequivocal characterization of honey samples requires the use of most of the previously described parameters. In this case, multivariate statistical analysis can be applied for finding trends or correlations among the characterization data, or for establishing the combinations of parameters which are highly related to the objective pretended by the honey characterization. There is no main data for this region and the goal of this study is to determine characteristics and oligosaccharide contents of honey samples in Kahramanmaraş City.
The ash content in honey is generally small and depends on nectar composition of predominant plants in their formation. The soil type in which the original nectar-bearing plant was located also influences the quantity of minerals present in the ash. As such, the variability in ash contents has been associated in a qualitative way with different botanical and geographical origins of honeys which is interesting when considering the production of a wide range of honey types (Abu-Tarbousch et al., 1993; Singh and Bath, 1997; Al-Khalifa and Al-Arify, 1999; Andrade et al., 1999; Latorre et al., 2000). The ash content is a quality criterion for honey botanical origin; the blossom honeys have lower ash content than honeydew honeys. Honeydew and/or mixed honeys have the highest ash content (Diez et al., 2004). Mean ash content of this study was in agreement with the study of Kılıç et al. (2007).

However, little attention has been given to the determination of how much of the variation in ash contents may be attributed to different floral sources or to factors related to honey sampling such as different geographical locations of various honey types or different environmental conditions of producing regions and technological aspects involved in apicultural practices and processing of honeys.

Insoluble solid and ash values were within the admitted limits except 3 samples (Turkish Honey Standard, 2002). According to TS Honey standard ash content of blossom honeys should be maximum 0.6%. Except one sample free acidity results were within admitted levels. HMF, ash and diastase activity values were within admitted limits (Turkish Alimentarus Codex, 2000).

Results about the acidity of honey samples were in agreement with those reported Abu-Tarbousch et al. (1993), Al-Khalifa and Al-Arify (1999), Andrade et al. (1999), Terrab et al. (2004) and Ouchemoukh et al. (2007).

The ash content of the studied honey samples differs widely. These differences in mineral content are dependent on the type of soil in which the original nectar-bearing plant was located (Anklam, 1998). Ash values were in agreement with Terrab et al. (2004) and Ouchemoukh et al. (2007) but lower than Al et al. (2009). The ash content is a quality criterion for honey botanical origin; the blossom honeys have lower ash content than honeydew honeys. Popek (2002), demonstrated that the ash content honeydew honey is 0.56%. Honeydew and/or mixed honeys have the highest ash content (Diez et al., 2004). Electric conductivity analyses were less than 0.75 mS/cm, but three honey sample results were high (1.13, 1.09, 1.05 mS/cm). The electrical conductivity of the honey is closely related to the concentration of mineral salts, organic acids and proteins; it is a parameter that shows great variability according to the floral origin and is considered one of the best parameters for differentiating between honeys with different floral origins (Terrab et al., 2003).

Variation of enzyme activity, from honey to honey, has been shown to occur for a variety of reasons, including the amount of sucrose in food sources, rate of nectar flow and even age of the bees. The minimum Standard value for diastase activity is eight, according to the Turkish Alimentarus Codex, Honey Rescript (2000).

Turkish Alimentarus Codex, Honey Rescript (2000), proposed 50 milliequivalents as the maximum permitted acidity in honey. Our result indicated that one sample was not admitted. The acidity of honey is due to the presence of organic acids, particularly gluconic acid, in equilibrium with their lactones or esters and inorganic ions, such as phosphate and chloride (Echingo and Takenaka, 1974).

The chemical properties of honey, such as pH, total acidity and mineral content, influence the formation of HMF (Bath and Singh, 1999; Anam and Dart, 1995). The honey samples studied showed an appropriate HMF content, below the allowable limit of 40 mg/kg, with the exception of honey coming from countries or regions with tropical climate and blends of these honeys, where HMF content must not exceed 80 mg/kg (Turkish Alimentarus Codex, 2000; Anonymous, 2001).

Our insoluble solids results were lower than Al-Khalifa and Al-Arify (1999), similar with the results of Andrade et al. (1999).

Oligosaccharide Contents of Honey Samples
Sucrose, isomaltose, maltose, turanose, nigerose and four trisaccharides as melibiose, panose, maltotriose; melezitose and raffinose were identified in 50 honey samples.

The results were listed in Table 1. As can be seen from mean values, from major oligosaccharides to minor ones were maltose (3.02%), nigerose (2.02%), turanose (1.72%), maltotriose (0.75%), isomaltose (0.44%), sucrose (0.41%), melezitose (0.28%), raffinose (0.19%), melibiose (0.1%) and panose (0.05%), respectively.

Maltose was the major oligosaccharide found in Kahramanmaras/Turkey honeys, which was in agreement with Földházi (1994) (mean values 3.36%), Mateo and Bosch-Reig (1997) (mean values 3.96%) and Da Costa Leite et al. (2000) (mean values 3.05%), but not with Siddiqui and Furgala (1967) (mean values 1.07%) and Doner (1977) (mean values 7.31%). Nigerose and turanose mean values were lower obtained by Siddiqui and Furgala (1967) of 0.17% for turanose and 0.06% for nigerose in Canadian honeys.

The mean values for maltotriose were similar with Da Costa Leite et al. (2000) (mean values 0.79%) but were higher than Földházi (1994) (ranged between 0.02-0.23%). The isomaltose contents were similar with Mateo and Bosch-Reig (1997). The sucrose content for all the samples was within the limits of the European Codex Honey Standards (5 g/100 g for honeys in...
general, with some exceptions such as not more than 10 g/100 g for citrus honey and not more than 15 g/100 g for lavender honey) (2001). The mean values for melezitose were higher than those reported by Al and Low (1990), Mateo and Bosch-Reig (1997) and Földházi (1994) (ranged between 0.03-0.06%). but lower than Al et al. (2009) (ranged between 0.01-2.75 g/100g). Raffinose mean values were close to honeys from Spanish origin Mateo and Bosch-Reig (1997), (ranged between 0.16-0.34%), but different from samples from Földházi (1994) (ranged between 0-0.06%). Melibiose findings were in accordance with Da Costa Leite et al. (2000) (mean values 0.11%). The low amount of panose (0.03-0.09%) was similar to the results obtained for Canadian honeys (trace-0.09%) (Swallow and Low, 1990; Da Costa Leite et al., 2000).

Our results indicate that the profile of oligosaccharides could be useful for the identification of the Kahramanmaraş region in which honey was produced and may also be useful for testing Turkish honey authenticity.

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