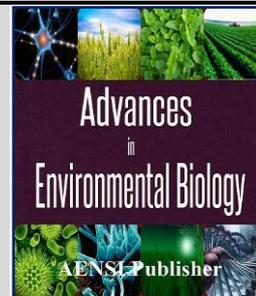




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Physico-Chemical Analysis And Determining Of Toxic Elements In Honey Produced In The Region Of Tiaret

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ABSTRACT

Honey is an important food due to its nutritive and medicinal values. This study was conducted with the objective to determine the physicochemical characteristics and toxic elements. Analysis of physico-chemical properties of honey studied shows a water content of 15 and 17 %, ash content of 0.11 and 0.16. Acidity was estimated to be 21.3 and 24.1 meq/kg and pH was 4.7 and 4.19. For the electrical conductivity, the values were 4.54 and 4.32 $\times 10^4 \mu / \text{cm}$. The values of the HMF content of our two honey samples were 4.6 mg / kg and 9.6 mg / kg. The cadmium content found in our two honey samples was respectively 0.03 mg / kg and 0.01 mg / kg and the lead content found in our two honey samples was respectively 0.05 mg / kg and 0.06 mg / kg. The results of physicochemical analyzes show a composition and physicochemical properties that conform to global standards established for honey nectar. The potentially toxic elements (Pb and Cd) found in our analysis of honey samples, do not present a danger to the human consumption because they are below the detection limit. Hence, it may be used as Bio-Indicator for monitoring the environmental pollution.

KEYWORDS: trace elements, honey, Tiaret, Bio-Indicator, environmental pollution

INTRODUCTION

According to the definition set by the European Union Council Directive [12], "Honey is the natural sweet substance produced by *Apis mellifera* bees from the nectar of plants or from secretions of living parts of plants or excretions of plant-sucking insects on the living parts of plants, which the bees collect, transform by combining with specific substances or their own, deposit, dehydrate, store and leave in honeycombs to ripen and mature". Honey is an important food ingredient due to its nutritive and medicinal values. It contains variety of macro and micronutrients as mainly sugars such as fructose, glucose and sucrose, organic acids, proteins, enzymes, vitamins and traces of minerals. Trace elements are very important for life but can also potentially have harmful effects. Some metals (Fe, Zn, Cu, and Mn) play important roles in a number of biochemical processes and are therefore referred to as essential elements, while others are non-essential elements (Pb, Cd). In contrast, elements such as cadmium, lead and mercury are well-known toxic elements for humans. The presence of heavy metals can pose human health risks. Trace elements in the atmosphere can deposit on the body hair of bees and they are transported to the colonies with pollen or they can still be absorbed with the flower nectar or transported by water or honeydew. It is necessary to measure the levels of heavy metals in honey as a method to make certain product quality and to detect environmental pollution.

Trace elements in the atmosphere can deposit on the body hair of bees and they are transported to the colonies with pollen or they can still be absorbed with the flower nectar or transported by water or honeydew[20].

Concentrations of different metals have been determined in honey in several countries: France [10], Italy [19] and Turkey [24].

A study by Aghamirlou *et al.* [1] has indicated that all types visited four times per year: after winter, before summer, during of honey contain metals and the metal concentrations vary among summer, and before winter. Pollen loads from traps were collected at different regions because of some variables.

Previous studies have suggested that honey may be useful as an environmental indicator of heavy metal pollution (As, Cd, Pb, Hg), as honeybees may be continuously exposed to contaminants. Therefore honeybees and their products can be considered representative bioindicators of the environmental pollution [5]. This work aimed to determine the main quality of honey samples produced in the region of Tiaret, via estimating some physicochemical parameters and also to reveal the presence of trace amounts of trace elements in the subject honey samples, as indicators for the environmental pollution.

MATERIALS AND METHODS

The study was carried out in the municipality of Tiaret. The samples, consisting of 150 mL of honey, were obtained directly from local beekeepers according to the peak of honey production in the region. The physicochemical analyzes were performed after collection of samples not exceeding the period of 30 days.

We assessed the following physical-chemical parameters: Moisture; pH; acidity; electrical conductivity; hydroxymethyl furfural (HMF); and ash content;

2.1. Physical-chemical parameters:

2.1.1 Moisture:

Sample moisture was obtained using a refractometer with the range expressed in percentage (%).

2.1.2 pH:

The pH was measured on a honey solution 10% using a pH meter. The determination was done according to norm CODEX [7].

2.1.3 Acidity:

According to norm CODEX [7] Acidity was obtained by performing the neutralization of acidic solution of honey (10 g of honey dissolved in 75 mL of distilled water) using a sodium hydroxide solution. Titrer with NaOH 0.05 N, Stop adding NaOH at pH 8.5. The result is expressed in meq kg⁻¹ using the equation:

Acidity = V (NAOH) x PA, where:

V (NAOH) = NAOH volume (mL)

PA = sample weight (g)

2.1.4 Electrical Conductivity:

According to the method published in the Official Journal of the French Republic [21], the electrical conductivity was measured using a conductivity meter. A solution containing 10 g of honey dissolved in 50 mL of distilled water was used for the readings (μScm^{-1}) of each sample.

2.1.5 Hydroxymethylfurfural (HMF):

According to the method published in the Official Journal of the French Republic [21], the determination of hydroxymethylfurfural (HMF) in the presence of barbiturate and paratoluidine acid, derived turns red. Given two tubes and placed in the first 2 ml of honey solution, 5 ml of reagent to para-toluidine and 1 ml of distilled water (blank test). In the second tube was poured 2 ml of honey solution (10g/50 ml), 5 ml of reagent to para-toluidine (10g of para-toluidine dissolved in 50 ml of isopropanol and then 10 ml of glacial acetic acid and complete up to the mark of 100 ml with isopropanol) and 1 ml barbituric acid solution (500 mg of barbituric acid dissolved in 100 ml distilled water). HMF content is expressed in mg per 1000 g honey is given by the following formula:

$$\text{HMF} = \frac{192 \times A \times 10}{\text{Weight of honey in grams}}$$

A: extinction

2.1.6 Ashes:

According to the Harmonized Methods of The International Honey Commission [11], the ash content in the samples was determined by the incineration of 5 g of honey (crucible) in a muffle at 600°C.

The result is expressed in percentage (%) according to the equation:

Ashes = $[(m1-m2)/m3] \times 100$, where:

m1 = weight of the crucible with the ash (g).

m2 = weight of the crucible (g)

m3 = weight of honey sample (g)

2.2. Determination of toxic elements in honey:

Digestion procedures:

One gram of sample was placed in a high form porcelain crucible. The furnace temperature was slowly increased from room temperature to 450° in 1 h. The samples were ashed for about 8 h until a white or grey ash residue was obtained. The residue was dissolved in 5 ml of HNO₃ (25% v/v) and the mixture, when necessary, was heated slowly to dissolve the residue. The solution was transferred to a 10 ml volumetric flask and made up to the volume. A blank digest was also carried out in the same way [25].

The heavy metal concentration was measured using the atomic absorption spectrophotometer

RESULTS AND DISCUSSION

3.1. Results Physical-chemical parameters:

3.1.1 Moisture:

The moisture of the two honey samples was 15 and 17 %. Generally, honeys have water content less than 18% [13].

3.1.2 pH:

The pH value of the two honey samples was 4.7 and 4.19. Both samples are acids honeys as they have a pH greater than 4. The pH is a measure for determining the floral origin of honey.

According to the standards of the Official journal of the French Republic [21], the honey with a pH between 3.5 and 4.5 are derived from nectar and honey derived from honeydew have pH between 5 and 5,5

3.1.3 Acidity:

The free acidity of honey is considered 21.3 and 24.1 mEq / kg.

The free acidity values obtained for all the samples are in the norm (<40 meq/kg) [2, 3, 16]. Yücel et al. [28] found, that the lowest mean value for the free acidity was 21.23 meq/kg.

3.1.4 Electrical conductivity:

The electrical conductivity of two honey samples were 4.54 and 4.32 Sx10⁴ μ / Cm) The electrical conductivity is a good criterion for determining the botanical origin of honey [18].

Jean-Prost [15] and Chauvin [6] indicate that the electrical conductivity ranges from 1 to 15 Siemens per cm Honeys derived from nectar had a conductivity of 1 to 5 x10⁻⁴S / cm and honey derived from honeydew 15x10⁻⁴S to 10 / cm; and median values often are based on natural mixtures of two origins [14, 23].

3.1.5 Hydroxymethylfurfural (HMF):

The values of the HMF content of our two honey samples were 4.6 mg / kg and 9.6mg / kg According to the European standard for honey, the maximum value of HMF is 40 mg / kg.

3.1.6 Ashes

The ash content of two honey samples was 0.11 and 0.16.

The measure allows ash generally determine the origin of a secretory honey analyzed and allows to highlight the clear honeys are quantitatively less rich in mineral material that dark honeys.

3.2 Result of determining of toxic elements in honey:

The results obtained of the two heavy metals (lead and cadmium) by atomic absorption spectrophotometer are shown in the following paragraph:

3.2.1 The cadmium:

The cadmium content found in our two honey samples was respectively 0.03 mg / kg and 0.01 mg / kg.

There is no specific maximum residue limits for cadmium in honey, but a value of 0.1 mg / kg was proposed by the European Union [5]. The levels of Cd found in our honey samples are well below this value.

These values are similar to those obtained by Yaiche-Achourand Khali [26] in 05 Algerian variety of honeys which are 0.018 and 0.019 mg / kg

Rached and Soltan [22] detected cadmium values around of 0.5 mg / kg in the Egyptian honey.

Lazoret *al.*, [17] found cadmium values ranges from 0.0942 to 0.0948 mg/ kg in theSlovakian honey.

3.2.2The lead:

The lead content found in our two honey samples honey was respectively 0.05 mg / kg and 0.06 mg / kg. There is no specific maximum residue limits lead in honey, but a value of 1.00 mg / kg was proposed by the European Union [5]. The lead content found in our honey samples are much less than this value.

Our results are belowto those obtained by Yaiche-Achourand Khali [26] in 05 samples of Algerian honeys that were 0.22 mg / kg and lazoret *al.*, [17]found Pb values ranges from 0.1252 to 0.1626 mg / kg in the Slovakian honey.

The values obtained in two honey samples are 0.05 mg / kg and 0.06 mg / kg. These results are similar to that found by Tuzenet *al.*, [25] in Turkish honey which was 0.058 mg / kg.

Higher concentrations were found by Rashed and Soltan, [22] in Egyptian honeys which were 15.5 and 19 mg / Kg. This high concentration of lead is usually related to environmental pollution.

Conclusion:

The results of physico-chemical properties conform to standards designed for nectar honeys, including data for HMF, water content, electrical conductivity, ash content, acidity confirm the good quality this honey. However, detection of pollutants (Pb and Cd) is consired in honey as negligible contamination and do not present a danger to the human consumption because they are below the detection limit. Environment affects the composition of honey. Hence, it may be used as Bio-Indicator for monitoring the environmental pollution.

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