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EVALUATION OF HONEY QUALITY: COMPARISON OF THE PHYSICO-CHEMICAL COMPOSITION INITIALLY AND AFTER TWO YEARS OF STORAGE

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Abstract

The study presents the evaluation of the quality of honey according to its physico-chemical composition, both at the time of harvesting and after two years of storage. Important parameters such as sugar content, moisture, pH, enzyme activity and other characteristics are measured and compared to determine whether and how storage influences these properties. The results of the study may provide valuable information for the beekeeping industry and consumers, highlighting the impact of long-term storage on honey quality and safety.

Key words**:** *honey, storage, quality, physicochemical composition, analysis*

INTRODUCTION

Bee honey is a natural product that has been known and used for thousands of years for its exceptional nutritional and therapeutic properties. Chemically, honey is a complex mixture of sugars, water, enzymes, organic acids, amino acids, vitamins and minerals, together with several bioactive compounds with antimicrobial and antioxidant properties. Thanks to this unique composition, honey has remarkable stability and can be stored for extended periods of time without significant deterioration [1, 2, 3, 7]. However, optimal storage conditions play a crucial role in maintaining its physic-chemical and biological properties. The storage of bee honey involves factors such as temperature, humidity and exposure to light, which can influence crystallization, fermentation and degradation of active compounds. Research shows that honey has inherent antimicrobial activity due to its low water content and high acidity, but prolonged exposure to unsuitable conditions can lead to degradation of enzymes and changes in taste, colour and texture [8, 9, 10]. Thus, understanding the factors that influence the stability of honey is essential to ensure the long-term preservation of its nutritional and therapeutic qualities [4, 5, 6].

The aim of this work is to analyse the quality of honey before and two years after storage, studying the impact of environmental factors on the stability of its composition and providing practical solutions for efficient storage [17, 18, 20].

MATERIALS AND METHODS

The beehive where the flower nectar was collected is located in Cisnădioara, Sibiu County. The honey is decrystallized and prepared for sale in the laboratory at 117 A, Valea Argintului Street. In addition to the stationary beehive in the village, the farm also has a trailer pavilion with about 120 hives. The breed used is *Apis melifera carpatica Foti*, 1965. We used six varieties (linden, manna, acacia and hawthorn, polyflower and rapeseed) from 2018-2019 and three others (linden, acacia, polyflower) from 2021 for laboratory analysis.

The analyses were repeated after two years of storage.

Electrical conductivity measures the ability of honey to conduct electric current and depends on the concentration of minerals, organic acids and ionic compounds present in honey [11, 12, 13, 14, 15, 16].

Steps to determine the electrical conductivity of honey:

A. Preparation of the test solution

A solution of honey dissolved in distilled water was prepared. The standard proportion is 20% honey in water, which means that 20 g of honey is dissolved in 100 ml of distilled water. The solution must be thoroughly homogenized to avoid any inhomogeneous particles that could affect the measurement.

B. Calibration of the conductometer

A conductometer is a device used to measure the electrical conductivity of a solution. Before measuring the honey solution, the conductometer must be calibrated using standard solutions with known conductivity values. This step is essential to obtain accurate measurements.

C. Conductivity measurement

After calibration, the conductometer electrode is placed in the honey solution. The electrical conductivity measurement is usually made at a standard temperature of 20°C. If the solution temperature differs, a correction factor must be applied to adjust the conductivity value [11,12, 13, 14, 15, 16].

D. Conductivity calculation

Conductivity is measured in microsiemens per centimeter (μS/cm). The value read directly from the meter is the conductivity of the honey solution.

Steps to determine the pH of honey:

Determining Honey pH

1. Prepared a Honey Solution

Accurately weighing 10 grams of honey, Dissolved in 100 ml of distilled water. Distilled water serves to prevent impurities from affecting the pH measurement. I Stirred the solution thoroughly to ensure us that it's evenly mixed.

2. Calibrating the pH meter

Before measuring the honey solution's pH, We calibrated the pH meter using standard buffer solutions. This ensures accurate readings pH meter, a device that must be calibrated before use. Calibration is carried out using buffer solutions of known pH (usually solutions of pH 4, 7 and 9) so that accurate and precise measurements are obtained.

3. pH measurement

After calibration, place the pH meter electrode in the diluted honey solution. It is important that the electrode is clean and completely immersed in the solution, without touching the edges of the measuring vessel.

The temperature of the solution may influence the result, so measurements are usually made at room temperature (20-25°C), or corrections are applied if the temperature differs.

Procedure for determining the alkalinity of honey

The determination of alkalinity is usually done by an acid-base titration method, using a strong acid (such as hydrochloric acid) to neutralize the alkaline substances in the honey [11, 12, 13, 14, 15, 16].

Dissolve a specific amount of honey 5 g in a specific volume of distilled water (about 50 ml). We used distilled water so as not to influence the result. To facilitate visualization of the neutralization point, we added a pH indicator such as phenolphthalein.

This will change the color of the solution when all the alkali in the honey has been neutralized by the acid.

I gradually added a standardized acid solution, usually hydrochloric acid (HCl), until the solution changes color, thus signalling the equivalence point, the point at which all the alkaline substances in the honey have been neutralized.

The volume of acid needed to neutralize the honey solution is used to calculate alkalinity. This is expressed in milliequivalents of acid per kilogram of honey (meq/kg).

RESULTS AND DISCUSSIONS

A. The acid value of honey is a very important parameter that provides essential information about the quality and freshness of honey.

Here are some key aspects that highlight the importance of this index:

1. Freshness and honey quality indicator

The acidity of honey can increase with time and the fermentation process caused by micro-organisms.

Fresh honey has a relatively low acidity, whereas old or fermented honey will have a higher acidity. Thus, the acid value is a direct indicator of the freshness of honey.

2. Fermentation process control

If honey contains high levels of water and yeasts, it can start to ferment. During fermentation, the acidity of honey increases due to the production of organic acids (such as acetic acid).

A high acid number may suggest that honey has started to ferment, affecting both the taste and the safety of the product.

3. Counterfeiting and tampering detection

Low-quality honey that has been adulterated or mixed with other products may have a different acid value than pure honey.

By monitoring this parameter, it can be detected whether the honey has been diluted or chemically altered.

4. Conservation potential determination

Low acidity indicates that honey can be stored for a longer period of time without losing quality.

Conversely, high acidity may signal a shorter shelf life and the need for additional preservation measures.

5. The correlation with floral composition

The acidity of honey is influenced by its floral origin. Honey made from different flowers will have different pH levels because the nectar and pollen have varying acidity.

This parameter can be used to identify the floral source and hence the type of honey, providing a guarantee of authenticity.

6. Influence on taste and flavor

The acidity of honey contributes significantly to the taste and flavor of the final product. Honey with low acidity has a smoother and more balanced taste, while high acidity can impart a sharper and sometimes unpleasant taste. Thus acidity is essential for the organoleptic evaluation of honey.

In general, the acid value of good quality honey should be below 50 milliequivalents of acid per kilogram (meq/kg) according to international standards.

Exceeding this threshold may indicate that the honey is of inferior quality or has been stored under inappropriate conditions Table 1.

Photo 1 provides information about the laboratory tools used for making the chemical analysis of honey.

Table 1. Determination of acidity index

Honey type	Year	Quantity (g)	Titration NaOH (ml)	
Linden	2018	11.07	21.72	30.52
Manna	2018	10.79	35.54	37.42
Acacia	2019	10.34	13.82	34.46
Acacia and Hawthorn	2018	10.08	13.82	17.72
Polyflower	2019	10.84	29.61	12.80
Rapeseed	2018	10.04	9.87	17.72
Acacia	2021	11.37	-	15.75
Linden	2021	11.75	-	22.64
Polyflower	2021	10.30	-	41.35

Source: Own results.

The acid value of honey is an essential parameter in assessing the quality, freshness and authenticity of honey. It helps to detect the fermentation process, check the quality of the product and ensure optimal taste and flavor, thus guaranteeing consumer safety and satisfaction.

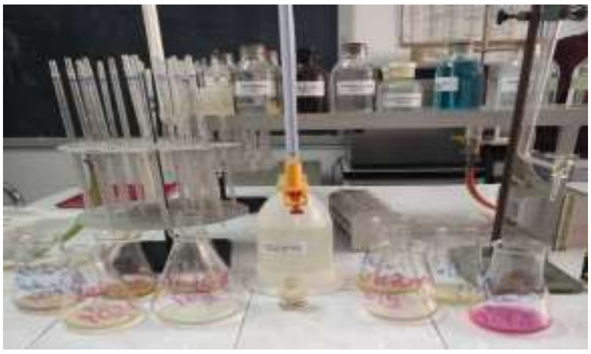

Photo 1. Honey acidity analysis, glassware, reagents and honey assortments
Source: original.

Our research found that four honey varieties — acacia and hawthorn, acacia, manna, and linden — became more acidic over time.

We also identified a decrease in the acidity of rapeseed and polyflower honey (Fig. 1).

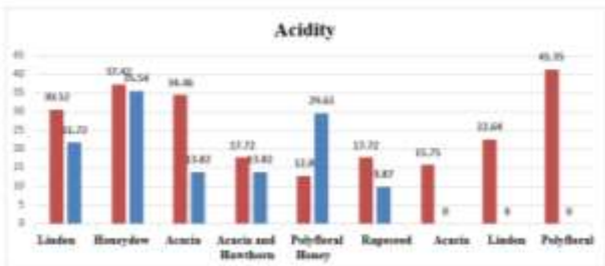

Fig. 1. Graphical representation of the values obtained after the determination of acidity
Source: original.

B. Determination of electrical conductivity of honey is an important method used to assess the purity and composition of honey, as it is an indicator of the floral source and possible contamination or unauthorized additions.

Honey derived from flower nectar typically exhibits low electrical conductivity, generally measuring less than 0.8 mS/cm.

It comes mainly from flower nectar and has a lower mineral content.

Manna honey (forest honey): has a higher electrical conductivity, usually above 0.8 mS/cm, due to its higher concentration of minerals and salts.

This comes from the excretions of insects that feed on plant sap.

Other honeys: The electrical conductivity of honey is influenced by a variety of factors, including the type of honey, the soil conditions where the nectar-producing plants grow, and the surrounding environment.

Measuring honey's conductivity is important because it helps us:

(i)Pinpoint the honey's origin: Conductivity acts like a fingerprint, providing clues about the types of flowers or plants used to produce the honey. This information can be used to verify the honey's source and ensure its authenticity.

For example, blossom honey and manna honey can be clearly differentiated by this test.

(ii)**Purity and quality evaluation**: Increased conductivity may indicate the presence of additional minerals that are not specific to natural honey, suggesting possible contamination or added substances.

(iii)**Determining geographical origin**: The mineral content of honey varies according to geographical region. Determination of conductivity can help to establish the regional origin of the product.

(iv)**Detection of counterfeit:** Abnormal conductivity may indicate that honey has been counterfeited or mixed with syrups or other synthetic products.

The determination of electrical conductivity is an essential parameter in the analysis of honey, providing valuable information about its composition and authenticity.

The method is fast, accurate and non-invasive, contributing to the correct classification of honey and ensuring a high quality standard. Regulations: STAS 784/3-2009 in Romania and 1151/2012 Europe [19].

Table 2**.** Determination of electrical conductivity

Honey type	Year	Electrical conductivity µS· cm-1	
Linden	2018	1.78	1.69
Manna	2018	1.29	1.67
Acacia	2019	0.80	1.73
Acacia and Hawthorn	2018	1.15	1.75
Polyflower	2019	1.38	1.63
Rapeseed	2018	0.61	1.79
Acacia	2021	-	1.72
Linden	2021	-	1.71
Polyflower	2021	-	1.67

Source: original.

The electrical conductance of honey analyzed in 2020 has values between 0.61 and 1.78 µS· cm-1;

The electrical conductance of honey analyzed in the year 2022 has values between 1.63 and 1.79 µS· cm-1.

Here it is easily observed that the electrical conductivity of honey assortments after storage for two years is higher (Fig. 2).

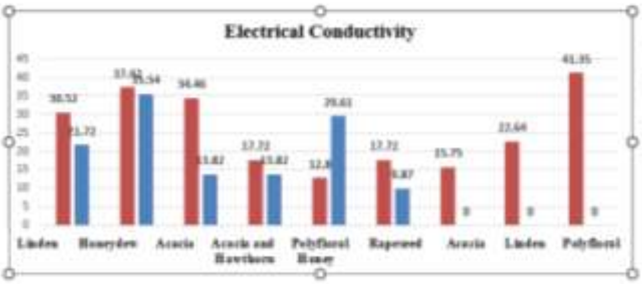

Fig. 2. Graphical representation of the values obtained after determining the conductivity of honey
Source: original.

C. Determining the acidity of honey, measured by its pH, is crucial for understanding its quality and properties. This acidity comes from natural components like organic acids, particularly gluconic acid.

The acidity of honey plays an important role in its microbiological stability and in preventing fermentation, and also contributes to its unique taste.

The pH meter will display a numerical value representing the pH of the honey solution (Fig. 3).

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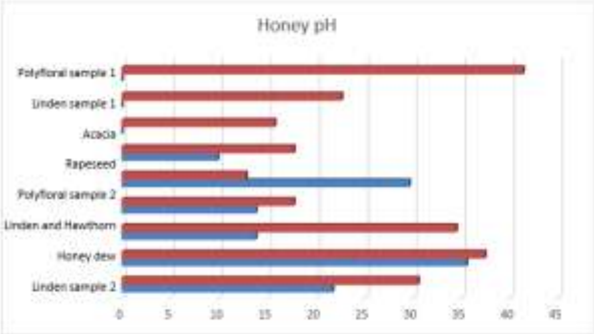

Fig. 3. Honey pH by type
Source: Original.

Honey is naturally acidic, typically with a pH between 3.2 and 4.5. Knowing the pH of honey is important because it can tell us about its freshness and overall quality.

(i) **Fresh honey** tends to have a specific pH, and any changes in that pH might suggest that it's not as fresh as it should be.

A higher acidity may suggest the presence of ongoing fermentation processes, while a lower pH indicates that the honey is less prone to fermentation and bacterial growth. Low pH also contributes to its microbiological stability.

(ii)Determining floral origin

The type of honey influences its pH level. Blossom honey and honeydew honey, for instance, have different amounts of organic acids and minerals, leading to variations in pH. Honeydew honey, with its higher mineral content, tends to be less acidic than blossom honey.

(iii) This **difference in acidity** affects the taste of honey. Honey with a lower pH has a sharper, more tart flavor, while honey with a higher pH tastes milder and sweeter.

Therefore, pH can affect the perception of honey flavor.

(iv)Indicator of degradation processes - If the pH of the honey is higher than typical values, this may indicate fermentation in progress or degradation of the product due to improper storage conditions.

Such a product may present an increased risk of microbiological spoilage and is not recommended for consumption.

The determination of the pH of bee honey is essential to assess its acidity and stability, helping to ensure product quality.

pH also plays an important role in the taste of honey and can be a key factor in identifying the type of honey and its floral source.

Legislation: SR 784-3:2009, 2009 in Romania and 1151/2012 in Europe.

Table 3. Determination of pH

Honey type	**Year**	**pH-ul**	
Linded	2018	5.11	4.37
Manna	2018	4.65	4.07
Acacia	2019	4.90	3.84
Acacia and Hawthorn	2018	5.47	4.30
Polyflower	2019	4.30	4.06
Rapeseed	2018	5.94	4.18
Acacia	2021	-	3.91
Linded	2021	-	3.93
Polyfloral	2021	-	4.03

Source: original.

A difference can be seen between the results in 2020 and 2022 two years after storage. After two years of storage of the honey samples a decrease in pH units can easily be observed.

D. The determination of the alkalinity of honey is a method used in the chemical analysis of this natural product to measure its ability to neutralize acids.

The importance of determining the alkalinity of honey

(i)Indicator of floral and geographical source:

The alkalinity of honey provides information about its content of alkaline substances (especially minerals), which may originate either from the natural composition of honey or from contamination or adulteration.

Figure 4 presents the values of total alkalinity by honey type.

The alkalinity of honey varies according to its floral source and the geographical environment in which the bees were collected. For example, honey from certain regions with mineral-rich soils may have a higher alkalinity. In contrast, floral honey, which comes from the nectar of flowers, usually has a lower alkalinity than honeydew honey, which comes from tree sap.

(ii)Verification of purity and quality: The alkalinity of honey can be a useful parameter to identify contamination or adulteration. An abnormal alkalinity value could suggest the addition of unapproved substances or exposure to improper storage conditions.

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(iii)Evaluation of the production process: Abnormal alkalinity levels may indicate m mishandling of the honey, including overheating, which can change the chemical composition of the honey, leading to deterioration in the quality of the product.

(iv)Determination of chemical stability: Too high or too low a level of alkalinity can influence the long-term stability of honey, affecting both taste and microbiological properties.

1. *Flower honey*: The alkalinity of floral honey is generally low, due to its lower content of minerals and alkaline compounds. It typically ranges between 0 and 1 meq/kg.

2. *Manna honey:* On the other hand, manna honey has a higher alkalinity due to the higher concentration of minerals and salts. Typical values for peanut honey are higher than 1 meq/kg.

3. *Deviation from normal values:* Abnormally high alkalinity may suggest the presence of contaminants or chemical changes during honey processing (e.g., excessive heating or the addition of additives). A very low alkalinity could indicate poor quality honey or possible dilution.

Honey from different flowers can have different levels of alkalinity, depending on the mineral composition of the nectar and pollen.

Apple honey usually has a higher alkalinity due to its plant source and higher mineral content.

Improper handling, such as heating the honey at high temperatures, can affect its chemical composition, changing its alkalinity. Determining the alkalinity of honey is a useful test for assessing the composition and quality of honey. Alkalinity can provide important information about the floral source, purity and potential contamination of the product. This is a simple and effective quality control method that helps to maintain high standards in honey production.

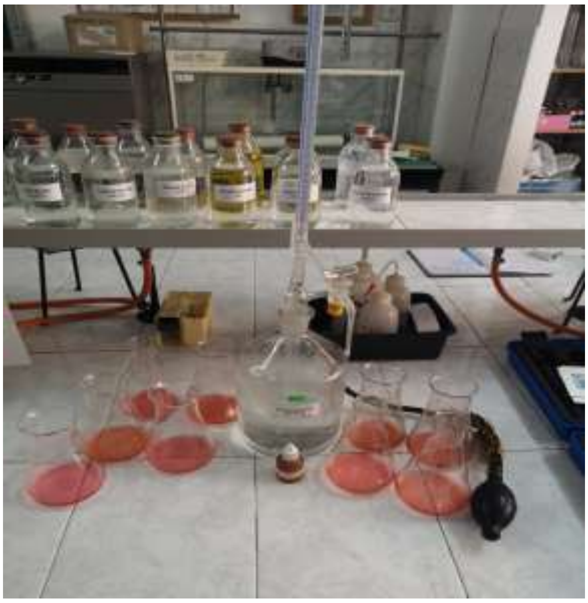

Photo 2. Honey alkalinity analysis, glassware, reagents and honey assortments
Source: original.

Comparing the results of alkalinity determination of bee honey between the years 2020 and 2022, we can observe:

-an increase in total alkalinity in honey from linden, acacia, acacia and hawthorn and polyflorous honey;

-a decrease in the total alkalinity of honey.

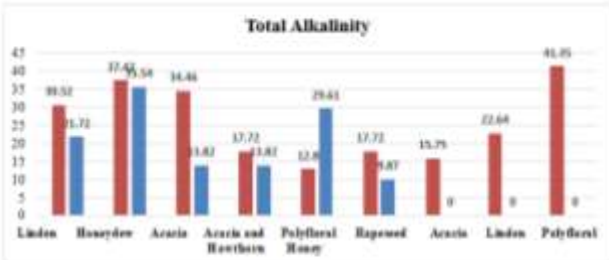

Fig. 4. Total alkalinity by honey type
Source: Original.

E. The water content of honey is an essential parameter for determining its quality and stability over time.

Too much water can lead to fermentation, affecting the flavor, texture and nutritional value of honey. In general, quality honey has a water content of around 16-18%. Accurate analysis is necessary to ensure these standards.

Why is it Important to Know the Water Content of Honey?

(i)To avoid spoilage: If honey contains too much water (above 18%), it can ferment. This happens because yeasts that thrive in sugary environments can grow and produce alcohol and carbon dioxide.

(ii)To ensure long-lasting quality: Honey with lower water content is more stable and can be stored for longer periods without spoiling or changing its texture and flavor.

(iii)To maintain high quality and value: Honey with a lower water content generally has a better texture and taste, making it more desirable and valuable.

What Factors Affect the Water Content of Honey?

(I'll need more information to answer this part! Factors that influence water content in honey include things like the nectar source, beekeeping practices, climate, and processing methods.)

Air humidity: During extraction and storage, honey can absorb moisture from the air. It is therefore recommended that honey is stored in dry, well-ventilated rooms.

Flower type: Different types of honey can have varying water contents depending on the source of the flower from which bees collect nectar.

The moment of honey collection from the combs. If honey is collected too early from the combs, it may contain more water than optimal.

In conclusion, determining the water content in honey is crucial to ensure the quality, safety and stability of this product.

Modern methods, such as refractometry and oven dehydration, provide accurate solutions for this process, helping to maintain high standards in honey production and marketing. Regulations: STAS SR 784-3: 2009 in Romania and 1151/2012 in Europe.

Table 4 presents the determination of water content by honey type.

Table 4. Determination of water content in honey

Honey type	Year	Refractive index	Water (%)	Dried substance	Density
Linden	2018	1.4896	19.1%	80.9 %	1.4885
Manna	2018	1.4910	17.6%	82.4 %	1.4296
Acacia	2019	1.4785	23.4%	76.6 %	1.3946
Acacia and Hawthorn	2018	1.4873	20.1%	74.6 %	1.4580
Polyfloral	2019	1.4915	16.8%	83.2 %	1.4310
Rapeseed	2018	1.4920	18.9%	81.1 %	1.4930
Determination of water content					
Honey type	**Year**	**Refractive Index**	**Water (%)**	**Dried substance**	**Density**
Linden	2018	1.4896	19.1%	80.9 %	1.4885
Manna	2018	1.4910	17.6%	82.4 %	1.4296
Acacia	2019	1.4785	23.4%	76.6 %	1.3946
Acacia and Hawthorn	2018	1.4873	20.1%	74.6 %	1.4580
Polyfloral	2019	1.4915	16.8%	83.2 %	1.4310
Rapeseed	2018	1.4920	18.9%	81.1 %	1.4930
Acacia	2021	1.4885	19.2%	80.8%	1.4226
Linden	2021	1.4865	20.0%	80%	1.4170
Poliyfloral	2021	1.4925	17.6	82.4%	1.4334

Source: original.

CONCLUSIONS

This study is a rigorous evaluation of honey quality after a two-year storage period. The research was carried out on nine varieties of honey harvested from the Cisnădioara apiary and subjected to detailed physic-chemical analysis to assess changes in their composition. The nutritional value of the honey, mainly due to its high sugar content (70-80%), classifies it as a high energy food. In addition, the presence of vitamins, which come exclusively from the pollen and nectar of beekeeping plants, contributes to its quality. In this research, honey varieties were re-evaluated after two years of storage under controlled conditions, without exposure to moisture and light, in sealed containers, to observe whether and to what extent the physic-chemical composition was affected. The results showed changes in some parameters such as pH, acidity, alkalinity, conductivity and water content, but the organoleptic properties and water and dry matter content remained stable. These findings confirm that all assortments analysed are natural, with no added additives or sweeteners, retaining their authenticity even after two years of storage.

For not losing its quality and healing properties, honey must be preserved on a shelf in well-sealed glass jars, in a dark and dry place or with maximum 60% humidity, a cold room with a temperature of +20 degrees and also well ventilated and without any fungi. and molds.

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