Libyan Journal of Medical and Applied Sciences LJMAS

Online ISSN: 3006-1113

Volume 3, Issue 4, 2025, Page No: 01-10

Website: https://ljmas.com/index.php/journal/index

Study of the mineral composition and color intensity of some honey samples circulating in the Libyan market

Rabia Omar Eshkourfu [1], Aisha ALfituri Benjuma[2*], Warda Mohamed Altogbia [3], Hanan Saleh Abosdil [4]

[1,2,3,4] Department of Chemistry, College of Science, Elmergib University, Al-Khoms, Libya

[*]*Corresponding author:* aabenjuma@elmergib.edu.ly

Received: August 01, 2025 Accepted: September 27, 2025 Published: October 02, 2025

Cite this article as: R, O, Eshkourfu., A, A, Benjuma., W, M, Altogbia., H, S, Abosdil. (2025). Study of the mineral composition and color intensity of some honey samples circulating in the Libyan market. Libyan Journal of Medical and Applied Sciences (LJMAS). 2025;3(4):01-10.

Abstract:

Considering that honey is a complete food rich in minerals, which are an important reason for honey acquiring different colors, this study aimed to determine the levels of sodium (Na), potassium (K), lithium (Li), calcium (Ca), and barium (Ba) in six samples. Sample H1 was recorded the highest concentrations of these elements (12.7, 48.8, 1.4, 8.8, and 17.3 ppm), respectively. UV spectroscopy results showed that sample H3 had the highest iron concentration (7.05 mg/100 g), while H6 had the highest phosphorus level (317 mg/g). In addition to the elemental analysis, the honey color was evaluated using the Pfund scale, a commonly used standard that measures the color and intensity of honey samples on a spectrum from light to dark. Sample H6 recorded the highest Pfund value and density (134.73 mm Pfund and 805 mU), showing a dark amber color. In contrast, sample H3 was recorded the lowest Pfund value and density (52.29 mm Pfund and 204 mU), showing a light amber color.

Keywords: Honey Bee, UV Spectrophotometer, Elements, Colour Intensity of Honey.

دراسة التركيب المعدني والكثافة اللونية لبعض عينات العسل المتداولة في السوق الليبي

ربيعة عمر اشكورفو[1]، عائشة الفيتوري بن جمعة[2*]، وردة محمد التقبية[3]، حنان صالح أبوسديل[4]

[1,2,3,4] قسم الكيمياء، كلية العلوم، جامعة المرقب، الخمس، ليبيا

الملخص

باعتبار أن العسل غذاء متكامل وغني بالعناصر المعدنية والتي تكون سببا مهما لاكتساب العسل ألوانا مختلفة. فقد تناولت هذه الدراسة تحديد مستويات الصوديوم (Na)، والبوتاسيوم (K)، والليثيوم (Li)، والكالسيوم (Ca)، والباريوم (Ba)، في ست عينات. حيث قد سجلت العينة 1H أعلى تركيز في هذه العناصر 17.3، 8.8، 1.4، 48.8، 12.7 (ppm) على التوالي. بينما بينت نتائج التحليل الطيفي بجهاز UV أعلى تركيز للحديد للعينة 3H (ملغم/ 100 غرام7.05)، في حين أن أعلى مستوى للفوسفور 6H (ملغم/ غرام317). علاوة على تحليل العناصر، تم تقييم لون العسل باستخدام مقياس بفوند Pfund هومعيار مستخدم بشكل شائع، والذي يقيس لون وكثافة عينات العسل على طيف مُتدرج من الفاتح إلى الداكن. سجّلت العينة H6 أعلى قيمة وكثافة لـ Pfund (134.73 ملم Pfund و805 مللي وحدة)، مُظهرةً لونًا كهرمانيًا داكنًا. في المقابل، سجّلت العينة 3H أدنى قيمة وكثافة لـ Pfund (52.29 ملم Pfund) و204 مللي وحدة)، مُظهرةً لونًا كهرمانيًا فاتحًا.

الكلمات المفتاحية: عسل النحل، مطياف الأشعة فوق البنفسجية، العناصر، شدة لون العسل

Introduction

Honey Bee is considered a divine gift, is a remarkable substance known for its healing properties and nutritional richness. With its sweet, aromatic taste and viscous texture, honey is a product of bees collecting nectar and pollen from plants, transforming them with specific substances, and storing them in honeycombs to mature [1]. This natural elixir is primarily a concentrated water solution comprising a blend of carbohydrates (typically 80%) and water (about 17%), along with various minor components like enzymes, vitamins, amino acids, proteins, lipids, volatile compounds, flavonoids, phenolic acids, pigments, and minerals [2]. As a vital bee product, honey holds significant nutritional and economic value, making it a highly sought-after commodity in agriculture worldwide

[3]. Notably, honey stands out as one of nature's most intricate food items and serves as the sole natural sweetening agent that can be consumed by humans without any processing [4]. The chemical properties of honey, including its mineral content, are impacted by factors such as honey maturity, climate, and collection/storage methods. However, the chemical composition of honey is not solely determined by these factors. It is also influenced by a multitude of variables like ecological conditions, soil quality, bee species, honey maturation process, colony health, environmental influences, and seasonal variations. These interconnected factors play a crucial role in shaping the distinct characteristics and attributes of different types of honey. The mineral composition of honey is intricately linked to the botanical sources of nectar, the flowers from which nectar and pollen are collected, and bee secretions. This connection highlights the diverse range of elements derived from various plant species that contribute to the mineral content of honey [5].

The quality and quantity of mineral elements in honey depend on the elemental composition of the flowers collected by the honeybee, which in turn vary according to the source of vegetation and the of the spatial distribution [6,7]. The colour of honey serves as a crucial quality indicator, reflecting the components derived from plant nectar dissolved in water, including pigments originating from plants and environmental factors. Honey hues range from yellow to darker shades, influenced by factors like the concentration of dyes in the nectar, such as chlorophyll, carotene, and xanthophyll, varying based on plant type and environmental conditions [8]. Darker honey varieties are found to contain higher levels of essential macroelements (e.g., calcium, magnesium, potassium, sodium), microelements (such as copper, iron, manganese, zinc), and trace elements (including aluminum, cadmium, nickel) [9]. Urban pollution and industrial activities can lead to increased concentrations of elements like aluminum, barium, cadmium, copper, manganese, nickel, lead, and zinc in honey [10].

Studies have identified 31 elements in honey sourced from different plants, with macro and microelement concentrations typically falling within the range of 0.04% to 0.2% [11]. The mineral content in honey can exceed 1%, with minerals dissolved in the water content of honey, ranging from 13.4% to 22.9%. These minerals not only influence the color and texture of honey, whether liquid or semi-liquid, but also contribute to its nutritional value [10].

Metals enter honey plants through the soil, absorbed by plant roots and eventually transferred to nectar, becoming part of the honey produced by bees [12]. The chemical composition of honey, especially concerning key metals like calcium, potassium, magnesium, manganese, and sodium, is significantly influenced by soil composition shaped by geographical, climatic, and geological factors [10]. Microelements play essential roles in biological processes, contributing to natural physiological development, metabolism, and overall metabolic functions. Key elements like sodium, potassium, calcium, iron, zinc, copper, and manganese are vital for the biological processes of living organisms [13]. On the other hand, toxic elements like lead, cadmium, mercury, and arsenic are environmental pollutants that can have harmful effects on health when present in high concentrations [13]. Heavy metal contamination in honey can lead to a range of health problems, including headaches, metabolic issues, respiratory distress, nausea, and damage to vital organs, such as the brain, kidneys, and nervous system [14].

Previous research has confirmed the correlation between the visible elements present in the location where bees collect nectar and the composition of honey. Building on this foundation, the current study aims to analyze the elemental content, color and color intensity of honey.

Methods

Apparatus and equipment

volumetric flasks, beakers, measuring cylinders, spatula, funnel, filter papers, pipettes, water bath, drying oven (Memmert), digital Analytical Balance (Model CTG1200-1200), Burning oven (Nabertherm), BWB-XP Technologies flame photometer, and ultraviolet- visible spectrometer (JENWAY 6300UV spectrophotometer).

Chemicals and reagents

Reagents that were used in the analysis were HNO3(69.5%), Hydrochloric acid (HCL) 37%, (deionized water. that was used for diction of honey samples. And also used during optimization of honey samples. Standard solutions for sodium, potassium, boron, calcium, barium, iron, and phosphorus, respectively that are used for the preparation of calibration standards and in the spiking experiments.

Determination of Elements

The elements analyzed in this study include sodium (Na), potassium (K), lithium (Li), calcium (Ca), and barium (Ba). Sample preparation followed the method outlined by Smith [15], and element concentrations were quantified using a BWB-XP Technologies flame photometer (UK, ISO, 9001 certified). While, the iron (Fe) and phosphorus (P) content were analyzed using JENWAY 6300UV spectrophotometer following the procedures outlined in references [16,17]. The examination of minerals and trace elements adhered to specific instrumental parameters for individual elements, with concentrations derived from the relevant standard calibration curves.

Honey colour analysis

The colour of honey plays a significant role in assessing its quality, serving as a visual indicator of the nutrients and essential components present, reflecting its overall vitality. Honey color evaluation can be conducted through various methods, with the Pfund scale or spectroscopic analysis being the most commonly employed techniques, as outlined in method [18]. In this process, samples are diluted to create a 50% honey solution and subjected to UV analysis at a wavelength of 635 nm, with the average absorption measured repeatedly. The obtained average absorption values are then converted using the Pfund scale equation for color assessment, enabling comparisons with other types of honey such as manuka honey.

$$Pfund = -38.70 + 371.39 \times Absorbance\lambda 635$$

Colour intensity

The colour intensity of honey is a significant quality attribute that plays a crucial role in determining the overall appeal and value of this natural sweetener. The color of honey can vary widely, ranging from light golden to dark amber, depending on factors such as the floral source, processing methods, and storage conditions.

Color intensity was evaluated following the procedure outlined in reference [19], involving the preparation of a 50% solution of the honey sample heated to a temperature range of 45-50°C. Subsequently, the samples underwent filtration using filter paper to prevent the presence of any clustered particles. The absorption levels were then measured and quantified in milli absorbance units (mAU), representing the variance between the absorption values at wavelengths of 450 nm and 720 nm.

$$mAU=Abs450-Abs720$$

Results and Discussion

Mineral analysis

The primary mineral elements present in honey include potassium (K), sodium (Na), calcium (Ca), and magnesium (Mg), with iron (Fe), copper (Cu), manganese (Mn), and chlorine (Cl) [20]. Also being significant. Smaller amounts of elements such as boron (B), phosphorus (P), sulfur (S), silicon (Si), barium (Ba), and nickel (Ni) are also found. These minerals are essential for human health. As they contribute to various metabolic functions, help maintain normal physiological processes, influence the circulatory and reproductive systems, and act as catalysts in numerous biochemical reactions [22].

The mineral composition of honey varies between different types; floral honey typically contains 0.1–0.2% mineral content [23]. For the Potassium (K) is the dominant mineral in honey, making up about 80% of the total mineral content [24]. Followed by phosphorus (P), calcium (Ca), magnesium (Mg), sodium (Na), and iron (Fe). Studies have confirmed the presence of potassium as the main mineral in honey samples from regions such as Turkey [25]. Serbia [26]. and Morocco [27].

Table 1. Concentration of minerals in six honey samples**.**

samples	The mineral (ppm)				
	Na	K	Li	Ca	Ba
H1	12.7	48.8	1.4	8.8	17.3
H2	9.2	33.2	0.4	7.2	12.7
H3	5.7	9.2	0.0	3	1.1
H4	7.3	25.1	0.2	6.7	10.2
H5	11.6	18.8	0.1	7.3	8.2
H6	9.3	20.1	0.3	5.6	6.5

The above table presents the mineral content (ppm) of sodium (Na), potassium (K), lithium (Li), calcium (Ca), and barium (Ba) in six different honey samples (H1 to H6). Analysis of the concentrations of these minerals can provide insight into the chemical composition of the samples and may provide useful information for a variety of fields, including environmental science, geology, and materials chemistry. From this table, we can see that the mineral content varies greatly across different honey samples, with some elements showing higher concentrations in certain samples. For the Sodium (Na) and potassium (K) show higher concentrations in H1, while barium (Ba) is more concentrated in H1 and lower in the other samples.

Potassium (K)

Concentrations are highest in sample H1 (48.8 ppm) and decrease progressively in subsequent samples, reaching 9.2 ppm in H3. The Potassium (K) was identified as one of the four major nutrients inadequately consumed according to the Dietary Guidelines for Americans 2010. The benefits of K, particularly when linked to organic anions as found in foods like honey, are well recognized. Potassium levels can vary significantly within the range (1.18 to 268 ppm), and this is consistent with the previous studies which have also reported higher concentrations of K in honey samples from Eastern Slovakia [28]. The Serpentin Area in the Eastern Rhodopes Mt. In Bulgaria [29,30,31].

Sodium (Na)
concentrations in the samples range from 5.7 ppm (H3) to 12.7 ppm (H1). The highest concentration is found in sample H1, the Sodium (Na) is crucial for normal cell function, maintaining plasma volume, regulating acid-base balance, and facilitating nerve impulse transmission [32].

Lithium (Li)
concentrations are generally low across all samples, with the highest concentration (1.4ppm) observed in sample H1. The other samples, such as H3, the presence of lithium in honey and suggests that plants growing in lithium-rich soils can absorb the element. The bees then collect nectar from these plants, which leads to trace amounts of lithium being incorporated into the honey. the natural occurrence of lithium in honey but notes that the concentrations are typically very low and not sufficient to have any significant therapeutic effect [33].
Calcium (Ca) concentrations range from 3.0 ppm in H3 to 8.8 ppm in H1. The Calcium (Ca) is essential for bone health and the calcification of bones [34]. In our study, Ca concentrations varied significantly, with the lowest values being <1 ppm (from Hatay and Şanlıurfa) and the highest being 4.50 ppm (from Adana). These values are considerably lower than those reported for Eastern Slovakia [28] (20.3 ± 3.09 ppm) and Argentina [30] (6.92 ± 4.35 ppm), as well as in a study by Pisani et al.[6],where calcium levels were notably higher.
Barium (Ba) is another element that shows significant variation across the samples. H1 has the highest concentration of barium (17.3 ppm), followed by H2 (12.7ppm) and H4 (10.2ppm). Samples such as H3 and H6 have much lower barium concentrations. The concentration of barium (Ba) in honey is typically found in trace amounts 0.1 - 0.5 mg/100g. Barium is a trace element that can enter honey through the soil, plants, and environmental factors, particularly from industrial sources and pollution. Its concentration varies depending on the geographic area, the type of honey (e.g., honeydew or blossom honey), and environmental conditions [35].

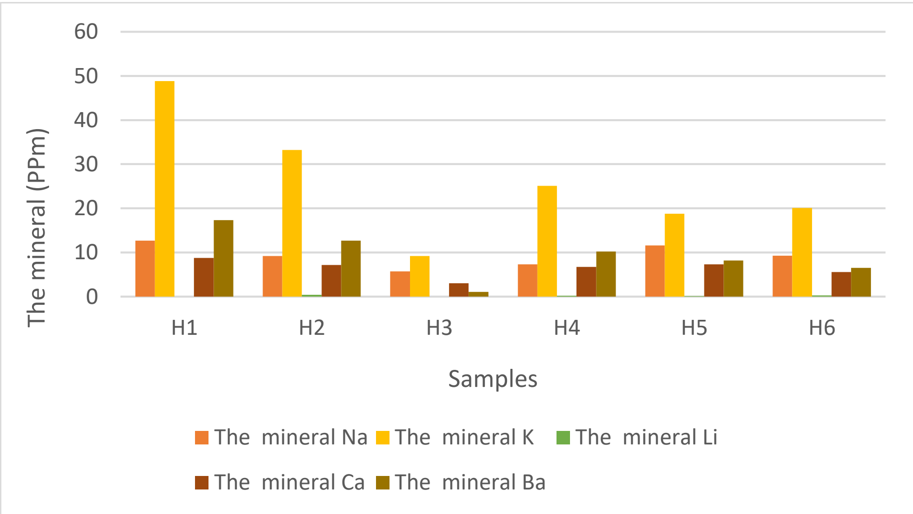


Figure 1. Concentration of minerals in six honey samples.

Iron (Fe)
Iron is a crucial mineral that plays a vital role in various physiological functions in both humans and animals. In bee honey, iron contributes to its antioxidant properties and helps support overall health. Research suggests that iron in honey may help boost the immune system and prevent iron deficiency. Further investigation into the specific mechanisms and benefits of iron in bee honey is warranted to fully understand its significance [36]. The data we obtained is detailed in table 2 and ranged from 4.25 to 7.05 mg/100g, as shown in figure 2. The data indicates varying levels of iron content across the samples, with the highest concentration observed in sample H3 at 7.05 mg/100g and the lowest in sample H5 at 4.25 mg/100g. This variation in iron levels among the samples may be attributed to factors such as floral sources, geographical location, and bee species. Further analysis and comparison of these results could provide insights into the quality and nutritional value of the honey samples. The differing concentrations of iron may be attributed to the climatic conditions of the locality under observation. Fore there, the documented levels of iron (Fe) in the tested samples do not present any health hazards to consumers, as the concentrations fall within safe limits across all samples analyzed [1].

Table 2. Concentration of iron(mg/100g) in six honey samples.

sample	H_1	H_2	H_3	H_4	H_5	H_6
con. mg/100g	**6.25**	**5.75**	**7.05**	**6.15**	**4.25**	**6.7**

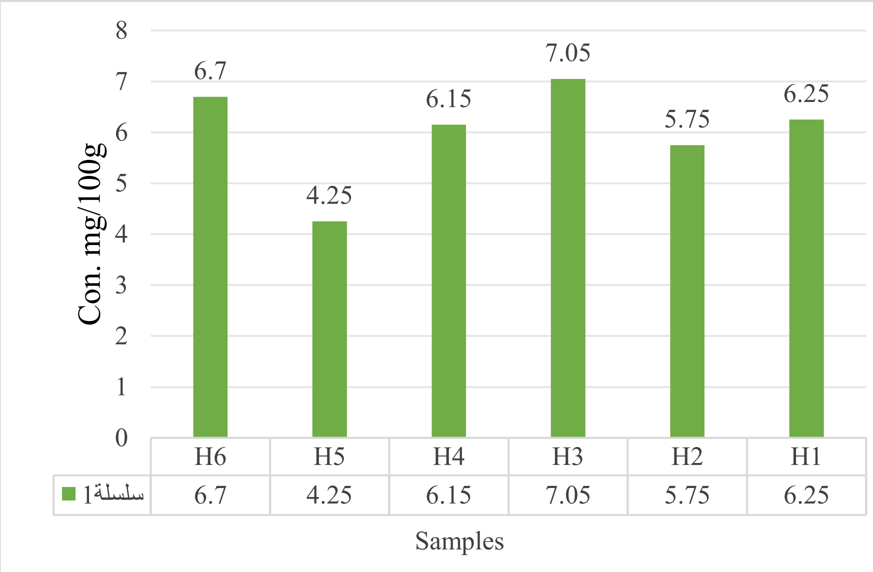


Figure 2. Concentration of iron(mg/100g) in six honey samples.

Phosphorus (P) Phosphorus in bee honey is another essential mineral that plays a significant role in various biological processes. The concentration of phosphorus in honey can vary depending on factors such as the type of flowers visited by the bees, the region where the honey is produced, and the specific bee species involved. Further investigation into the levels of phosphorus in different honey samples and its potential impact on the nutritional composition of honey is warranted to enhance our understanding of this important mineral in bee honey.

The table 3 and figure 3 provide information on the phosphorus content (mg/100g) in six honey samples labeled H1 to H6. The data illustrates notable variations in phosphorus levels among the samples, with sample H6 exhibiting the highest concentration at 317 mg/100g, while sample H4 demonstrates the lowest concentration at 8 mg/100g. These differences in phosphorus content could be influenced by diverse factors such as the botanical sources of the nectar, the environmental conditions in which the bees forage, and the bee species responsible for honey production. Further analysis of these findings may offer valuable insights into the nutritional profile and potential health implications of the various honey samples. Consequently, the relationship between the lightness of color and mineral content in the samples indicates that potassium and phosphorus were the most influential factors contributing to the darker hue of this honey compared to other minerals [37].

Table 3: Concentration of phosphorus(mg/100g) in six honey samples

sample	H_1	H_2	H_3	H_4	H_5	H_6
con. mg/100g	204	188	23.2	8	110	317

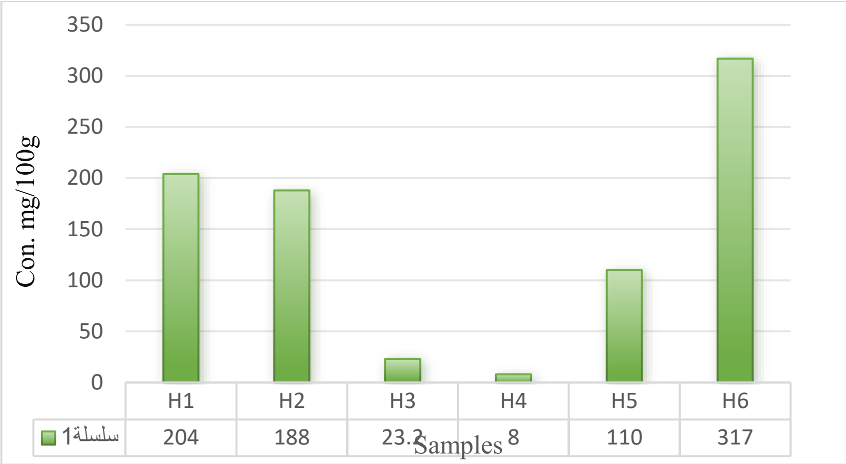


Figure 3. concentration of phosphorus(mg/100g) in six honey samples.

Honey colour

The colour of honey plays a crucial role in determining its market value and consumer acceptance. Honey displays a wide array of colour variations, from almost colorless to deep brown, accompanied by a diverse range of flavors, ranging from mild to bold tastes. These variations in both flavor and color are directly influenced by factors such as the nectar season, specifically the source of the flowers, and the duration between nectar collection and honey harvesting [38]. The table 4 presents the results of samples analyzed using the Pfund scale, indicating the color intensity of each sample. The range of results varies across the samples, with sample H6 showing the highest value at 134.73 mm, categorized as Dark Amber. In contrast, sample H3 has the lowest value at 52.29 mm, classified as Light Amber. These results highlight the diversity in color intensity among the samples, which can impact consumer perception, market value, and overall acceptability of the honey product. The Pfund scale measurements provide insights into the colour of the samples, spanning from Light Amber to dark Amber, determined by millimeter readings. This information allows for a thorough evaluation of the samples concerning their absorbance levels and colour intensity as categorized by the Pfund scale.

Table 4: The Pfund scale colour in six honey samples.

Samples	H1	H2	H3	H4	H5	H6
Sample result range	63.8	67.5	52.29	106.51	71.97	134.73
Pfund scale (mm)	Light Amber	Light Amber	Light Amber	Amber	Light Amber	Dark Amber

The figure 1 shows the absorbance for honey samples using a spectrophotometer measuring honey samples at a wavelength of 635 nm. There are seven colour classifications of honey, from extra white to dark amber [16]. In the present study, H6 sample has the highest Pfund value (134.73 mm Pfund) and has a dark amber colour, while H3 sample given lowest Pfund value (52.29 mm Pfund) which is the light amber colour. The differences in colour intensity among various honey samples are related to the unique ratios of dissolved substances derived from plants, including pigment compounds abundant in chlorophyll and carotene found in the nectar, as well as the concentration of mineral salts and minerals, which affect color differently depending on the plant species. Additionally, organic compounds in honey, particularly those that form complexes with transition elements like iron, significantly impact its colour [38]. These elements collectively determine the colour and intensity of honey, with external factors such as temperature and storage conditions also playing a role in influencing honey's colour [39].

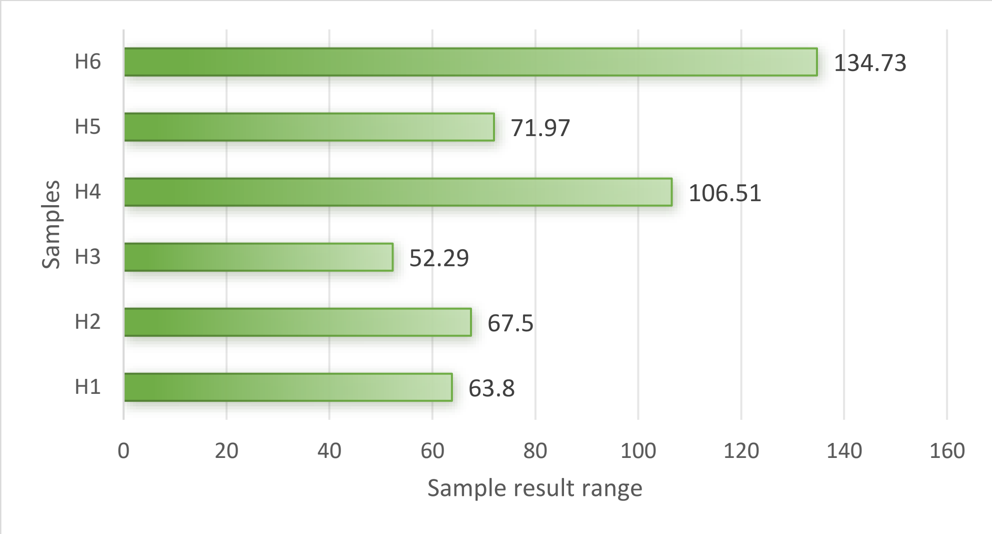


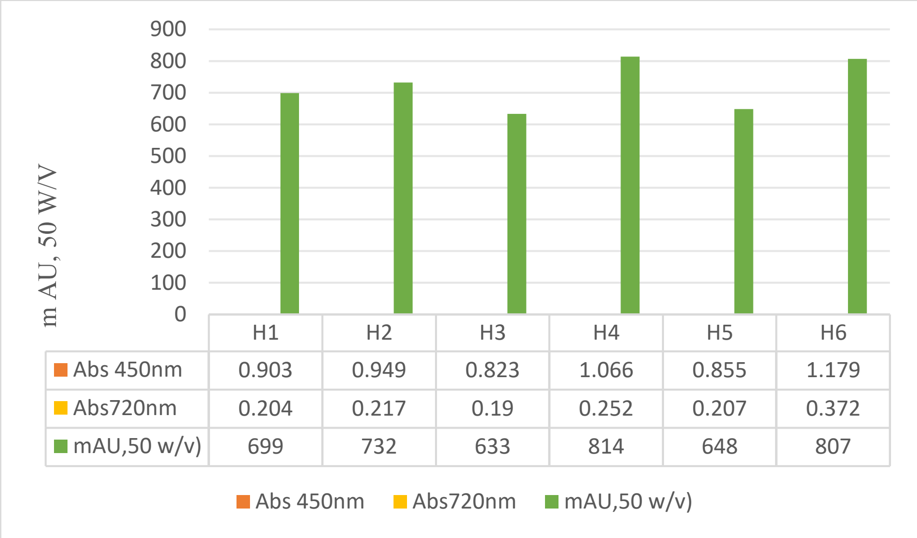
Figure 4. Absorbance values of honey samples measured at a wavelength of 635 nm.

Colour intensity

The intensity of honey colour is primarily influenced by the presence of pigments, such as phenolic compounds and flavonoids, as well as Maillard reaction products that form during heating and processing. Lighter coloured honeys are typically milder in flavour, while darker honeys tend to have a stronger, more pronounced taste. Table 5 and the accompanying figure present Pfund color intensity data for six honey samples, including absorbance values at 450 nm and 720 nm, as well as milli-absorption units (mAU) at 50 w/v. The results reveal differences among the samples, with sample H6 showing the highest mAU value of 807, suggesting a potentially darker color, while sample H3 displayed the lowest mAU value of 633, indicating a lighter colour. The observations regarding colour intensity and compositional differences in the studied honey samples serve to highlight the diverse characteristics and unique qualities present in various honey products. These variations in honey color and intensity can be attributed to the varying proportions of plant and flower contents that serve as the feeding sources for bees [40].

Table 5. The Pfund scale colour in six honey samples.

Samples	**Abs 450nm**	**Abs720nm**	**mAU,50 w/v)**
H1	0.903	0.204	699
H2	0.949	0.217	732
H3	0.823	0.19	633
H4	1.066	0.252	814
H5	0.855	0.207	648
H6	1.179	0.372	807



	H1	H2	H3	H4	H5	H6
Abs 450nm	0.903	0.949	0.823	1.066	0.855	1.179
Abs720nm	0.204	0.217	0.19	0.252	0.207	0.372
mAU,50 w/v)	699	732	633	814	648	807

Figure 5. Colour intensity**.**

Conclusion

In conclusion, and through the results obtained and based on the references cited by the study, we saw that the level of minerals and their availability in honey components has a strong relationship with the colour of honey and its colour intensity. The colour intensity of honey serves as a visual indicator of its composition, with variations reflecting the diverse sources of nectar and the presence of different compounds such as pigments and minerals. Understanding the relationship between colour and mineral content in honey is essential for assessing its authenticity, quality, and potential health benefits. Further research in this area can provide valuable insights into the unique characteristics and properties of honey, enhancing its appreciation and utilization in various applications.

Recommendation

To enhance the quality and consistency of honey in Libya, it is imperative to establish standardized specifications that cover all crucial properties of this natural product. These specifications should be disseminated across all regions involved in honey classification to ensure uniformity and adherence to quality standards. It is essential to include detailed information on honey packaging, such as type, origin, and components, to provide transparency to consumers.

Regular monitoring of bee habitats is necessary to evaluate environmental conditions and nutritional sources, which directly impact the quality of honey. Employing proper preservation and storage techniques for honey samples is vital to maintain their freshness and quality over time. Additionally, conducting routine quality analyses of honey in stores is essential to verify compliance with the established Libyan standards, thereby safeguarding consumer trust and satisfaction.

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