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Comparative Analysis of Quality Parameters of Honey Collected from Domesticated and Wild Honeybee Species in District Faisalabad, Punjab, Pakistan

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Abstract

Honey is a natural sweet substance produced by honeybees from the nectars of plant flowers and honeydew. This study has performed a comparative analysis of the physicochemical properties of honey collected from wild (*Apis dorsata*) and domesticated (*Apis cerana*) bee species from different areas of District Faisalabad, Pakistan. The quality of honey was evaluated for floral sources in the case of wild bees and sugar syrup in the case of domesticated bees. Our data reveal differences in the two honey groups regarding ash, minerals, and HMF content. The amount of minerals was recorded higher in *A. dorsata* than in *A. cerana* honey. Similarly, the amount of HMF was higher in the honey of *A. dorsata* than in *A. cerana*. The moisture content, pH, glucose, and glucose oxidase levels were similar in both honey types. This preliminary study is very useful for developing strategies for the quality analysis of honey, which ultimately promotes honey business entrepreneurship in the international market.

Introduction

Honey holds a particular place in the food and medical industries, and it has been regarded as a highly nutritive food in many civilizations (Feas et al., 2010). The term “honey” refers to the nectar and sugary secretions from plants that are collected, transformed, and stored in the honeycomb by honeybees (The Honey England Regulations, 2015). The Codex Alimentarius Commission defines honey as a natural sweet substance produced by honeybees from plant nectar or from secretions of plant-sucking insects found on living plant parts. Honeybees collect these substances, mix them with their own enzymes, transform them, and deposit them in honeycombs. There, the honey is dehydrated, stored, and left to ripen and mature. Bees gather sweet juices from various nectar-producing plants, process them internally, and store the final product in wax honeycombs, which are later harvested by beekeepers.

The composition and quality of honey are variable (Ahmed et al., 2018). It depends mainly on the botanical source of nectar from which it is obtained, domesticated bees from different origins, but also depend on the presence of bees in urban gardens is influenced by several factors, including the city's location and climate, the surrounding landscape, the types and origins of host plants, and pesticide use. Identifying the bee species found in these gardens can greatly support effective garden management (Suni et al., 2022). Faisalabad also has environmental conditions conducive to the growth of high floral biodiversity that contributes to honey production.

The physicochemical properties of honey affect its honey storage, quality, granulation, texture, flavor, nutritional characteristics, and medicinal qualities (Iftikhar et al., 2011). The characterization of honey promotes the understanding of its medicinal properties and antibacterial and antioxidant characteristics. Beekeeping is a profitable industry in Pakistan, with approximately 7,000 beekeepers currently raising exotic bee species. *A. cerana*, in the modern beehives. About 300,000 colonies produce 7,500 metric tons of honey annually. Four species of honeybees are found in Pakistan.

Pakistan is home to four honeybee species – three indigenous and one exotic species that has been successfully introduced and established. These species are found across various ecological regions of the country. Many of honey's physicochemical properties can be analyzed to detect adulteration, making it essential to study specific quality parameters to ensure its purity (Khan et al., 2016).

Naturally, honey is a thick, sticky, and viscous solution containing about 15-17% water. However, its water content is not constant due to honey's hygroscopic nature, meaning it can absorb moisture from the air. The water level can change during storage, depending on ambient humidity. Higher water content increases the risk of fermentation (Jovanovic, 2015). Honey typically contains 80-85% carbohydrates, primarily glucose and fructose, which make up about 95-99% of its dry matter. It also contains approximately 0.1-0.4% protein, 0.2% ash, and small amounts of amino acids, enzymes, vitamins, and phenolic antioxidants. It includes around 4-5% fructo-oligosaccharides (Ajibola et al., 2015).

There is some debate among scientists about the origin of the proteins in honey. Some suggest they come mainly from the bees' salivary glands, used in processing nectar and honeydew, while others believe most proteins are derived from pollen, which contains 10-35% protein.

Organic acids contribute to the acidity and characteristic flavor of honey. Despite its sweet taste, honey is naturally acidic, with an average pH of 3.9 (typically ranging from 3.4 to 6.1). This acidity inhibits the growth of many microorganisms that require neutral or alkaline environments (Aurongzeb et al., 2011). The acids commonly found in honey include acetic, butyric, citric, formic, lactic, maleic, malic, oxalic, and succinic acids (Ball et al., 2018).

Honey's quality is influenced by its composition, color, aroma, and taste (Kruzik et al., 2017), and its rheological (flow-related) properties are considered key physical indicators of quality (Bayram et al., 2023). As a highly viscous sugar solution, honey is often supersaturated and prone to crystallization over time. The crystallization rate depends on factors such as water content, the presence of crystallization nuclei, the degree of supersaturation, and viscosity – which is affected by temperature. While crystallization is natural for certain monofloral honeys (like citrus), it is generally seen as a defect in commercial honey (Piana et al., 2023).

Methodology

The research was conducted in the Insect Molecular Biology Lab and the Central HI TECH LAB, Laboratory of the University of Agriculture, Faisalabad, Pakistan.

Honey sampling and collection

The study included nine honey samples from two different localities, Faisalabad, province of Punjab, Pakistan, including two types of the most specific *Apis dorsata* and *Apis cerana.* Honey samples were collected in 2023 from different geographical origins, mainly including wild and domesticated.

Moisture content

The refractive index method was used to determine the moisture content with three replicates. The obtained refractive index values were then converted into moisture percentage, and the obtained data was analysed by one-way ANOVA (Bogdanov et al., 1997).

pH and free acidity

Free acidity and pH were measured in the samples by pH meter and the potentiometric titration method. The Acidity of honey is based on nectar, bee secretions, or organic acids (tartaric, citric, oxalic, acetic, etc.).

Protein

The micro Kjeldahl method was used to determine the nitrogen contents from the honey samples, and the protein contents were estimated by using the conversion factor 6.25 (N 9 6.25).

Sugar content

Glucose, fructose, and sucrose were analyzed using High-Performance Liquid Chromatography (HPLC) with a Shimadzu Prominence System equipped with a Refractive Index Detector (RID-10A, Shimadzu). The separation was carried out using a Shim-Pack CLC-NH2 column (6.0 × 150 mm, 5 µm particle size) under isocratic conditions. The mobile phase consisted of acetonitrile and water (80:20, v/v), filtered through a 0.45 µm membrane. The flow rate was maintained at 1.3 mL/min, with both the column and detector set at 30 °C. An autosampler (SIL-20A, Shimadzu) was used for sample injection. Quantification was performed using the external calibration method, with calibration curves prepared for glucose, fructose, and sucrose in the concentration range of 50 to 500 µg/mL. Final sugar concentrations were expressed in grams per 100 grams of dry weight.

Ash content

The gravimetric technique was used to measure the quantity of ash (Al-mentafji et al., 2006). 10 g of the material was weighed into the crucible heated for 12 hours at 550 °C in the furnace. After heating and cooling in the desiccator for half an hour, as measured by a spectrophotometric technique, the crucible was sealed to avoid gas ash and particles escaping.

Mineral content

Mineral contents were determined by weighing 0.5 grams of the honey sample into a Teflon digestion dish with 3 mL of ultra-pure water, followed by 2.5 mL of 65% nitric acid (HNO_3). Wet digestion mixture was achieved by microwave

oven heating at 500 W for 2.5 minutes, 1000 W for 20 minutes, 1200 W for 30 minutes. After digestion solution was placed at room temperature and transfered the clear digested solution in to a 50 mL volumetric flask and diluted with ultra-pure water. Spike all prepared solutions with an internal standard to achieve a final concentration of 10 µg/L.

Statistical analysis

The data were analyzed using one-way and two-way analysis of variance (ANOVA) with three replicates, applying appropriate data transformations where necessary. Additionally, t-tests were conducted when applicable. Principal Component Analysis (PCA) and Hierarchical Cluster Analysis (HCA) were performed using Minitab software to classify and differentiate honey samples based on their physicochemical properties.

Results

The study aimed to evaluate the quality of honey collected from both domesticated and wild honey bee species and to check how these samples compare to international honey quality standards. To achieve this, the researchers collected various honey samples from local markets in District Faisalabad. These samples were then stored at room temperature in a dark place to prevent any changes in their chemical composition before testing. After conducting detailed laboratory analyses, the results showed **significant variations** between domesticated and wild honey samples in several key quality parameters. Specifically:

- **Moisture Content**: This measures how much water is in the honey. Excess moisture can lead to fermentation. The study found noticeable differences in water content between honey from different sources and regions.
- **Relative Density**: This refers to the thickness or heaviness of honey. It varies with sugar content and water levels. Samples from wild and domesticated sources showed different densities, which affect quality and shelf life.
- **Total Sugars and Reducing Sugars**: These are important indicators of sweetness and energy content. Reducing sugars (like glucose and fructose) are naturally present in honey and give it its main characteristics. The amounts varied significantly between the different types of honey.
- **Sucrose**: Higher sucrose levels can indicate immature honey or added sugar. The study found differences in sucrose content across the regions and honey types, which is important in detecting adulteration.
- **Hydroxymethylfurfural (HMF)**: This compound increases in honey that is overheated or stored for too long. Significant variation in HMF content suggests differences in processing or freshness among the samples.
- **Mineral Content**: Minerals contribute to the nutritional value and can reflect the floral source and soil where the bees forage. There were clear differences in mineral content between the wild and domesticated honey samples.

However, not all tested parameters varied across honey types:

- **pH and Protein Content**: These two parameters did not show significant differences between domesticated and wild honey. Instead, the differences observed were mainly linked to the geographic regions from which the honey was collected, not the source (wild or domesticated). This suggests that environmental factors like soil, plant types, and climate may play a more important role in determining pH and protein levels than the method of honey production.

In summary, the study confirmed that honey quality can vary widely depending on both its source (wild vs. domesticated) and the region of sample collection. These differences are important for consumers, producers, and regulators concerned with honey authenticity, safety, and nutritional value.

Discussion

This study aimed to compare the physicochemical properties of honey from wild (*Apis dorsata*) and domesticated (*Apis cerana*) honeybee species to evaluate their quality and purity. A total of 16 samples were collected from urban and rural regions of District Faisalabad, Pakistan. Four per region for each bee species. Honey samples were obtained from domesticated bee *A. cerana* and wild bee *A. Dorsata*. All samples exhibited notable variation in quality parameters. Moisture content in honey is influenced by harvest season, hive maturity, climate, and nectar water content (Nowak et al., 2021). The moisture levels in *A. cerana* samples ranged from 15.56 ± 0.90% to 18.70 ± 0.36%, while *A. dorsata* samples ranged from 15.90% to 18.00%, both within the Codex Alimentarius limit of <20%. These values align with previous findings from nearby regions (Abselami et al., 2018; Sharif et al., 2018) and indicate good storage potential and resistance to fermentation. The pH of the honey samples ranged from 3.53 ± 0.90 to 4.70 ± 0.90, consistent with nectar-based (blossom) honey. *A. cerana* samples showed no significant differences, except for sample 7, which had a notably higher pH. *A. dorsata* samples had pH values between 3.53 and 4.43, aligning with the typical range of 3.5-4.5 for blossom honey (Faustino et al., 2021; Wu et al., 2022). These values suggest acidic honey, which naturally inhibits microbial growth and confirms nectar origin. The major sugars analyzed were glucose, fructose, and sucrose: Honey obtained from *A. cerana* contained Sucrose: 6.65 ± 0.226 g/100g, Glucose: 34.37 ± 0.263 g/100g, Fructose: 38.50 ± 0.267 g/100, Total sugars: 79.31 ± 0.195 g/100g while honey of *A. dorsata* contained Sucrose: 6.60, ± 0.487 g/100g, Glucose: 34.81 ± 0.843 g/100g, Fructose: 40.51 ± 0.512 g/100g, Total sugars: 81.91 ± 1.614 g/100g.

There were no significant differences ($p > 0.05$) in glucose and fructose contents between the two honey types. The elevated sucrose levels in some *A. cerana* samples could result from artificial feeding practices. Nevertheless, total sugar content adhered to Codex standards. The fructose-to-glucose (F:G) ratios were in line with previous studies from Pakistan

and Bangladesh (Njokuocha et al., 2019; Kamal et al., 2019), indicating low crystallization tendency and good honey quality. Proteins in honey, primarily enzymes like diastase and invertase, are indicators of freshness (White, 1975). Protein content ranged from 0.136 ± 0.06 to 1.254 ± 0.020 g/100g, with *A. cerana* honey showing higher values than *A. dorsata*. This suggests a greater enzymatic activity during ripening in domesticated colonies. Our findings are consistent with prior work showing variation in protein content based on floral and geographical sources (Bogdanov et al., 2016; Gregory et al., 2023). Minerals were analyzed for all 16 samples. Potassium was the most abundant element, followed by calcium, sodium, and magnesium. Concentrations observed were:Potassium: 396.00-810.50 ppm, Calcium: 17.50-640.63 ppm, Sodium: 169.88-238.62 ppm.

These levels fall within or near the ranges reported by other researchers (Boussaid et al., 2015; Sleimi et al., 2022). Lead was detected in all samples but remained below 3 mg/kg, within permissible safety limits. Mineral profiles support the determination of geographical origin and quality.HMF content is a critical indicator of honey freshness and heating during storage. The average HMF values were: *A. cerana* (Urban): 41.00 ± 6.66 mg/kg, *A. cerana* (Rural): 39.33 ± 1.58 mg/kg, A. dorsata: 23.00 ± 0.43 to 28.60 ± 0.57 mg/kg.

Most samples were below the Codex Alimentarius limit of 40 mg/kg, except for one urban *A. cerana* sample (41.70 ± 0.49 mg/kg). Higher HMF values are typically associated with prolonged storage or heating. Our findings are in accordance as reported by Boussaid et al. (2018) and Kadri et al. (2017), suggesting that differences in HMF may be linked to bee species and their foraging behavior.

This research aimed to study the physiochemical comparison of honey between wild and domesticated species of honey. A study was conducted based on different honey samples to confirm the quality and purity of honey. A total of 16 samples were collected from urban and rural areas, four from each area, for both honeybee species. Commercially domesticated *A. cerana* species honey samples were collected from cultured colonies and *A. dorsata* from the wild. All the samples used in the research show markable variations in properties.

Differences in the moisture content of honey depend on harvest season, the degree of maturity reached in the hive, climatic conditions, and water content in the original plant nectar (Nowak et al., 2021). The mean range for moisture content of *A. cerana* honey studied in the honey of all samples was 15.56 ± 0.90 to 18.70 ± 0.36 percent, and *A. dorsata* honey ranged from 15.90 to 18.00 percent, which is a higher range (17.27-19.80%) of moisture content for Tunisian honeys (Boussaid et al., 2014). Honey texture, stability, and shelf life greatly vary on moisture (Thrasyvoulou et al., 2018). Observed moisture content for the honey of domesticated honey in Asia well below the limit (<20%) set by the Codex, indicating honey's degree of maturity against fermentation. Also, (Abselami et al., 2018) observed that the moisture content of honey in the neighboring state of Punjab varied from 17.08-18.89 percent. Therefore, the results obtained in the present study follow the standards set by Codex Alimentarius (2018) for moisture (<20%) and indicate a good storage ability of Pakistan honey samples analyzed. Comparable results were reported by (Sharif et al., 2018) (Table 2).

The pH (Means) of eight samples, *A. cerana* sample 1-4 urban and 4-8 rural, range from 3.56 ± 0.76 to 4.70 ± 0.90. They do not show statistically significant differences. Meanwhile, Sample 7 has a significantly higher pH than the others (Table 2). The variations in the pH values could be due to honey flow sources (foraged plants), salivary secretions of bees, enzymatic process, and fermentative conversion of raw material (Abselami et al., 2018). For *A. dorsata*, the mean range is 4.43 ± 0.90 to 3.53 ± 0.90. These results also follow those of (Wu et al. 2022), who reported a pH range of 3.72-4.02 for pure honey. Honeys with a pH range of 3.5 to 4.5 are considered to have originated from nectar. The present values for pH support the previous findings of (Kamboj et al., 2013) for honey of three major honey-producing states where pH was in the range of 3.90-4.70 and (Imtara et al., 2018) who reported a pH value of 3.80-4.41 for different Pakistan honey brands.

Irrespective of its geographical origin, honey is generally acidic. According to Faustino et al. (2021), honey with a pH range from 3.5 to 4.5 is considered blossom honey, while honey with a pH above 5 is of low quality. Hence, the honey of the domestic and wild bees analyzed in the present study can be categorized as blossom honey. Mostly, the samples had a pH in the normal range of 3.0-6.4. All of them are significantly different from each other.

The sugar composition of honey from *Apis cerana* (domesticated) and *Apis dorsata* (wild) was analyzed for glucose, fructose, and sucrose contents. For *A. cerana*, the mean ± standard deviation values were as follows: sucrose 6.65 ± 0.226 g/100g, glucose 34.37 ± 0.263 g/100g, fructose 38.50 ± 0.267 g/100g, and total sugars 79.31 ± 0.195 g/100g. In contrast, for *A. dorsata*, the mean ± standard deviation values were: sucrose 6.6 ± 0.487 g/100g, glucose 34.81 ± 0.843 g/100g, fructose 40.51 ± 0.512 g/100g, and total sugars 81.91 ± 1.614 g/100g.

The *A. cerana* and *A. dorsata* honey mean values (Table 1) for glucose and fructose displayed insignificant differences ($p>0.05$) within the two types of honey. Considerable variations in sugars are known due to changes during storage, and analysis of sucrose content was instrumental in detecting the adulteration of honey with table sugar or checking the amount of sucrose naturally found in a given honey sample (Baglio et al., 2017).

In the present study, the glucose, fructose, and sucrose contents of honey from various Apis species, as documented by Mustafa et al. (2019) from honeybee farms, were 27% and 34%, respectively. In contrast, honey sourced from natural environments had glucose, fructose, and sucrose contents of 22.50 ± 2.12% and 28.50 ± 3.54%, respectively. Notably, the sucrose content observed in the present study was higher

Table 1 - Comparative physicochemical characteristics of *Api cerana* and *Apis dorsata* honey from different locations.

SAMPLE	Moisture		pH		Protein		Ash		HMF	
Localities of honey	Mean of *A.cerana*	Mean of *A.dorsata*	Mean of *A.cerana*	Mean of *A.dorsata*	Mean of *A.cerana*	Mean of *A.dorsata*	Mean of *A.cerana*	Mean of *A.dorsata*	Mean of *A.cerana*	Mean of *A.dorsata*
S1	17.73	15.60	4.50	3.90	0.39	0.40	0.13	0.44	36.33	28.00
S2	16.62	17.58	3.53	3.66	0.33	0.39	0.14	0.36	36.67	24.33
S3	15.56	16.50	3.83	3.80	0.35	0.40	0.15	0.44	41.00	26.16
S4	16.50	17.43	3.40	4.10	0.37	0.41	0.13	0.40	32.00	28.60
S5	18.03	16.20	3.56	4.43	0.37	0.44	0.12	0.45	36.00	26.33
S6	18.06	16.20	3.56	4.43	0.34	0.41	0.17	0.49	39.33	24.00
S7	18.70	16.36	4.70	4.16	0.33	0.39	0.15	0.44	33.30	28.00
S8	18.00	18.00	4.46	3.53	0.36	0.37	0.18	0.46	34.00	23.00
Mean	17.40	16.73	3.94	4.00	0.35	0.40	0.14	0.43	36.07	26.05

than that reported by Khan et al. (2016), who found sucrose levels to range from 0.19% to 1.02% in *A. cerana* honey from the Punjab district. The higher sucrose content in the present study may be attributed to overfeeding honeybees with sucrose syrup. The total sugar content in the present study falls within the standard range set by Codex (0.95-1.5%) for *A. dorsata* honey, suggesting that this honey has a slow crystallizing nature. Additionally, the findings of the present study are consistent with those of Njokuocha et al. (2019), who reported a fructose-to-glucose (F:G) ratio for honey from Punjab, Faisalabad, Pakistan, ranging from 0.97 to 1.23. Similarly, Kamal et al. (2019) observed the fructose-to-glucose ratio in honey from the northwest region of Bangladesh to range between 1.14 and 1.34.

Honey proteins are primarily enzymes (White, 1975), which bees introduce during the honey-ripening process. Diastase and invertase are the key enzymes commonly used to assess honey freshness. However, their activity levels depend on the botanical origin of the honey, limiting their reliability as freshness indicators based solely on protein content. In Pakistani honey samples collected from various regions, protein content ranged from 0.06 ± 0.136 to 1.254 ± 0.020 grams per 100 grams of honey. A comparison of the protein composition of honey generated by and honey produced by *A. cerana* showed that it had a higher protein level, whereas *A. dorsata* had a lower protein level. The protein content of beekeepers' kinds of honey was less than the detection thresholds. The protein content of commercial kinds of honey ranged from 0.13 to 0.47 g/100 g, depending on the variety.

Honey contains all of the amino acids that are necessary for human health. Most honey proteins are enzymes (White et al., 2013). The bees introduce these enzymes into the honey during the ripening phase. The enzymes diastase and invertase are the most often employed to determine the freshness of honey (Gregory et al., 2023). Varied types of honey have substantially different protein levels, according to the findings. Protein concentrations varied from 0.136-10.06 to 1.25-0.020 grams of protein per 100 grams of honey. Our findings revealed that domesticated honey had the highest protein content, and wild honey had the second-highest protein content. These elements are critical in determining the quality of honey based on its composition (Bogdanov et al., 2016).

Table 2 - Mean ± SD values of the concentration of sucrose, fructose, glucose and total sugar for honey *A.cerana* and *A. dorsata* sample.

The concentration of sucrose, fructose, glucose and total sugar for honey sample					
		Sucrose g/100g	Glucose g/100g	Fructose g/100g	Total Sugars g/100g
A. cerana (domesticated)	Mean	6.65	34.37	38.50	79.31
	SE Mean	0.22	0.26	0.26	0.19
	St Dev	± 0.63	± 0.74	± 0.75	± 0.55
A.dorsata (wild)	Mean	6.60	34.81	40.50	81.91
	SE Mean	0.17	0.29	0.53	0.57
	St Dev	± 0.48	± 0.84	± 1.51	± 1.61

The mineral composition of honey also depends directly on the geographical and botanical origin of honey. Different elements of honey were determined from 8 samples, each of *A. cerana* and *A. dorsata* honey. The results of the concentrations of various elements in honey are presented in Figure 1. The mineral composition of honey is strongly influenced by its botanical origin, which makes it a useful indicator for determining both the geographical and floral sources of the honey. The most abundant minerals detected in all honey samples from Punjab were potassium, sodium, calcium, and magnesium, consistent with previous findings (Sleimi et al., 2022). These minerals were found within a broad concentration range of 0.13 to 2220 mg/kg, with potassium typically being the most dominant (>550 mg/kg). Lead was also detected in all samples, but only at low concentrations (<3 mg/kg).

Specifically, potassium, calcium, and sodium concentrations in the honey samples ranged from 396.00-810.50 ppm, 17.50-640.63 ppm, and 169.88-238.62 ppm, respectively. Boussaid et al. (2015) reported sodium concentrations between 497.54-362.55 ppm and 251.34-521.22 ppm, which are comparable to our current findings. Calcium levels in all fresh honey samples were below the international standard range of 200-2300 ppm. Similarly, Abdulkhaliq et al. (2017) reported lower concentrations of potassium (183.86 ppm) and sodium (104.66 ppm), which align with previous research. These results support the general consensus that potassium is the predominant mineral in honey (Liu et al., 2021), accounting for over 30% of its total mineral content. Other frequently observed minerals include sodium, calcium, magnesium, and iron. For mineral analysis, sample preparation commonly involved wet acid digestion using either nitric acid (Sleimi et al., 2022) or hydrogen peroxide (Gregory et al., 2023).

Hydroxymethylfurfural (HMF) is another key parameter used to assess honey freshness and purity (Codex Alimentarius, 2001). In our study, the mean HMF content in *Apis cerana* honey was 41.00 ± 6.66 mg/kg in urban samples and 39.33 ± 1.58 mg/kg in rural samples – both relatively high for domesticated honey. In contrast, HMF levels in wild honey ranged from 23.00 ± 0.43 to 28.60 ± 0.57 mg/kg, with an average below the Codex-recommended limit of 40 mg/kg. Boussaid et al. (2018) reported similar HMF values of 27.43 ± 1.50 mg/kg and 37.31 ± 17.13 mg/kg, while Amruta et al. (2019) recorded significantly lower HMF levels (3.78 mg/kg and 3.87-4.64 mg/kg) in Saudi and Indian honeys. High HMF content is generally attributed to prolonged storage and floral source variations.

According to Codex Alimentarius and EU standards, the maximum acceptable HMF level is 40 mg/kg. In our findings (Table 2), HMF content ranged from 23.80 ± 0.44 to 41.70 ± 0.49 mg/kg, mostly within the recommended limit – except for sample S-3 from urban domesticated honey, which slightly exceeded it. These results suggest that HMF formation is influenced by storage conditions, processing heat, and heating duration. Kadri et al. (2017) reported similar variability, highlighting that differences in HMF levels may also arise from the specific honeybee species and their foraging preferences (Sahney et al., 2017), which affect the chemical composition of the honey.

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Authors' Contribution

U.A.: Conceptualization; investigation; writing-original draft.

Z.U.A.: Methodology; supervision.

H.H.: Formal and data analysis.

U.A.C.: Formal analysis; writing-review & editing.

M.T.: Visualization.

W.A.: Funding acquisition.

M.U.S.: Project administration.

S.N.: Data collection.

S.E.T.R.: Data collection.

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