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Original research article

Quality Assessment of Honey Samples Collected in Tashkent, Uzbekistan

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Abstract Quality and authenticity of honey are largely determined by its floral origin, geographical factors, and post-harvest handling. Despite the growing significance of apiculture in Uzbekistan, scientific evaluation of domestically available honey remains limited. This study aimed to assess the botanical origin and physicochemical quality of honey samples obtained in Tashkent, Uzbekistan. In 2022, 30 honey samples were collected from apiaries, supermarkets, and Tashkent State Agrarian University. Melissopalynological analyses were conducted to determine the floral origins, classifying samples into 14 groups. Physicochemical parameters, including moisture content, stable carbon isotope ratio (SCIR), hydroxymethylfurfural (HMF), and sugar composition (fructose, glucose, sucrose, reducing sugar, F/G ratio), were analyzed. The results revealed notable variability among floral groups, although most samples adhered to Codex Alimentarius standards. All honey samples produced in Uzbekistan complied with thresholds for moisture (< 20%) and HMF (< 40 mg/kg). However, the Multifloral (*Onobrychis*) sample exhibited a SCIR value of −22.8‰ suggesting possible adulteration. The Multifloral (*Ferula*) sample failed to meet the 60% reducing sugar requirement, and one *Onobrychis* sample exceeded the 5% sucrose threshold. F/G ratios varied significantly, indicating differing crystallization tendencies. Additionally, while the overall effect of local origin was limited, statistically significant differences in moisture and F/G ratio were detected. These findings suggest that environmental exposure, handling, or storage conditions may influence specific quality attributes independently of floral origin. The combined application of melissopalynological, chemical, and isotopic analyses proved effective for comprehensive quality evaluation, highlighting the primary role of floral and geographical factors, along with minor contributions from distribution-related factors, in shaping honey quality.

Keywords Hydroxymethylfurfural, Stable carbon isotope ratio, Reducing sugar, *Alhagi*, *Helianthus*

INTRODUCTION

Blossom honey, the most common type of honey, is the natural sweet substance produced by honey bees from the nectar of plants. The process involves collection and transformation of nectar by bees, followed by deposition in the honeycomb, where it undergoes dehydration, storage, and maturation (Codex Alimentarius, 2001). It comprises approximately 200 compounds, including sugars (fructose and glucose), water, enzymes, amino acids, organic acids, vitamins, phytochemicals, and minerals (Bogdanov *et al*., 2008; Bergamo *et al*., 2019). The characteristics and composition of honey can vary depending on several factors such as bee species, season, environmental factors, beekeeping practice, storage, and especially geographical region and floral origin

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(Leite *et al*., 2000; Bogdanov *et al*., 2008; Kaškonienė *et al*., 2010; EL-Metwally, 2015; da Silva *et al*., 2016). Therefore, an analysis of honey's physical and chemical properties is important for evaluating its quality, confirming its authenticity, identifying variations related to geographical regions and botanical sources, and ensuring consumer safety (Solayman *et al*., 2016).

The moisture level of honey plays a key role in maintaining its physical and microbial stability by preventing crystallization and fermentation during storage (Nanda *et al*., 2003). Alongside moisture, the sugar composition also significantly influences crystallization (Bhandari *et al*., 1999) and ripening (Zhang *et al*., 2021) and serves as a key indicator in detecting sugar adulteration (European Commission [EC], 2021). The level of hydroxymethylfurfural (HMF) in honey serves as an indicator of freshness, with elevated levels typically associated with heat exposure or aging (Bogdanov *et al*., 2004). The stable carbon isotope ratio (SCIR; $^{13}C/^{12}C$) is widely used to detect honey adulteration, since most nectar-producing plants are C_3 species, while common adulterants like sugarcane and maize are C_4 plants (White *et al*., 1978).

Melissopalynology, the microscopic examination of pollen grains in honey, is used to determine its botanical origin (Brodschneider *et al*., 2019; Nunes *et al*., 2024), classify the honey as monofloral or multifloral (Selvaraju *et al*., 2019), and detect cases of mislabeling related to floral sources (Maieves *et al*., 2020). During nectar foraging, pollen grains adhere to the bodies of honey bees and are subsequently deposited into open honeycombs, ultimately remaining in the ripened honey (Jones and Bryant, 1996; Aronne and Micco, 2010; Sniderman *et al*., 2018; Nunes *et al*., 2024). This method is low-cost and practical; however, it demands expert knowledge and is constrained by the challenges of microscopically identifying morphologically similar pollen types (Bruni *et al*., 2015; Soares *et al*., 2015; Özkök *et al*., 2023). Nevertheless, it remains widely used due to its accessibility and affordability (Bodor *et al*., 2021; Kang *et al*., 2023; Shakoori *et al*., 2023; Shakoori and Salmanpour, 2024; Karki *et al*., 2025).

The Republic of Uzbekistan is a Central Asian country, with approximately 85% of its territory consisting of deserts and the remaining 15% comprising mountains and foothills (Belolipov *et al*., 2013; Mamadalieva *et al*., 2017). Given these challenging ecological conditions, the availability of both cultivated and wild-growing melliferous flora becomes essential for the survival of honey bees in the country (Atamuratova *et al*., 2021). Beekeeping in Uzbekistan, accordingly, plays a significant role in supporting regional economic activity through pollination services and honey production (Farmanov, 2025). In addition, honey-based foods, often referred to as East sweets, like *kozinaki*, *halva*, and *chak-chak*, have a long-standing tradition in Uzbekistan (Abdiniyazova *et al*., 2016). Despite the ecological and economic importance of beekeeping and the cultural value of honey, comprehensive studies on the physicochemical characterization of honey in Uzbekistan remain highly limited.

This study aims to compare the physicochemical properties of honey collected in Tashkent, Uzbekistan, including moisture content, sugar composition, HMF levels, and SCIR, based on its botanical origin, as determined through melissopalynological analysis. The central hypothesis is that the physicochemical characteristics of honey vary depending on the floral sources from which the nectar is derived. In addition, the study will evaluate whether the honey samples meet international quality standards. Furthermore, it will investigate whether these parameters differ depending on local sources, such as apiaries, supermarkets, open markets, and the Tashkent State Agrarian University (TSAU). The outcomes of this research could support the development of standardized quality criteria, strengthen mechanisms for honey authentication, and enhance the recognition of Uzbekistan's apicultural products as regionally distinctive and scientifically validated.

MATERIALS AND METHODS

1. Honey samples

In 2022, we collected 30 *Apis mellifera* honey samples from various local sources in Tashkent, Uzbekistan, including apiaries (n=9), supermarkets (n=9), markets (n=7), and TSAU (T; n=5). With the exception of four Russian honeys purchased from supermarkets, all samples were harvested in Tashkent. The study aimed to evaluate the quality of all honey types realistically accessible to consumers in Tashkent, regardless of their

geographical origin. Therefore, all available products at the time of sampling were purchased, while excluding duplicate products from the same producer or identical floral-origin honeys of the same brand to avoid redundancy and maximize sample diversity. According to the product labels, the floral origins on the honey products included, *Alhagi* (camelthorn; n = 2), *Ferula* (n = 1), *Gossypium* (cotton; n = 1), *Helianthus* (sunflower; n = 1), *Psoralea* (n = 1), *Salvia* (n = 1), *Tilia* (linden; n = 1), *Trifolium* (clover; n = 1), and multifloral (n = 21). Although the overall sample size was relatively small, the selected samples reflect a wide range of production and retail sources. Detailed information on the samples is provided in Table 1.

2. Melissopalynological analysis

The botanical source of each honey was determined by identifying the dominant pollen using the melissopalynology (von der Ohe *et al*., 2004). Briefly, an amount of 15 g of honey was mixed with 30 mL of distilled water and centrifuged at 1080 × g for 10 min. The supernatant was discarded, and this washing step was repeated twice. Next, 10 mL of 10% Safranin-O solution was added to stain the pollen, followed by centrifugation under the same conditions and removal of the supernatant. Then, 10 mL of absolute ethanol was added, and the sample was centrifuged again at 1080 × g for 10 min. After discarding the supernatant, the residue was resuspended in 1 mL of ethanol and transferred into a glycerol-containing tube. The sample was left at room temperature for 24 h to allow ethanol to evaporate and was subsequently used for pollen identification.

Morphological analysis of pollen grains was performed using a light microscope (BX53, Olympus, Tokyo, Japan) equipped with IMT i-Solution Lite software (IMT i-Solution Inc., Burnaby, British Columbia, Canada). Pollen identification was based on previously published references and two major palynological databases: PalDat (https://www.paldat.org/) and The Global Pollen Project (https://globalpollenproject.org/).

According to previously established thresholds, a honey can be considered monofloral if it contains at least 20% of *Medicago* pollen (Forcone, 2008), 15% of *Salvia* pollen (Kenjerić *et al*., 2006), or 20% of *Tilia* pollen (Polish Committee for Standardization, 1988). Although categorized as multifloral, the honey samples were further classified based on their predominant pollen types (e.g., Multifloral (*Helianthus*)), as presented in Table 1.

Only the dominant nectar sources identified through melissopalynological analysis are presented in this study, based on the detailed palynological data provided in Sun *et al*. (2025), where full pollen frequency distributions and spectra are available.

Table 1. Local source and botanical origin of *Apis mellifera* honey samples collected in Tashkent, Uzbekistan, in 2022, based on product labeling and melissopalynological identification

No.	Local source	Botanical origin	
		Label	Identification
1	Apiary	Multifloral	*Medicago*
2		Multifloral	*Salvia*
3		Multifloral	*Alhagi*
4		Multifloral	*Medicago*
5		Multifloral	*Salvia*
6		Multifloral	*Alhagi*
7		Multifloral	*Helianthus*
8		Multifloral	Multifloral (*Helianthus*)
9		Multifloral	*Helianthus*
10	Supermarket	*Gossypium*	Multifloral (*Helianthus*)
11		*Helianthus*	*Helianthus*
12		*Tilia*	*Tilia*
13		Multifloral	*Onobrychis*
14		Multifloral	*Alhagi*
15		Multifloral*	*Helianthus*
16		Multifloral*	*Medicago*
17		Multifloral*	*Helianthus*
18		Multifloral*	*Helianthus*
19	Market	*Alhagi*	Multifloral (*Alhagi*)
20		*Alhagi*	*Alhagi*
21		*Psoralea*	*Alhagi*
22		Multifloral	*Medicago*
23		Multifloral	Multifloral (*Calligonum*)
24		Multifloral	*Alhagi*
25		Multifloral	*Helianthus*
26	TSAU	*Ferula*	Multifloral (*Ferula*)
27		*Salvia*	Multifloral (*Onobrychis*)
28		*Trifolium*	*Ferula*
29		Multifloral	*Medicago*
30		Multifloral	*Alhagi*

*Asterisk indicates that the honey samples produced in Russia.

3. Physicochemical analysis

1) Moisture content

Moisture content was assessed using a digital honey refractometer (ORL 94BS, Kern & Sohn GmbH, Balingen, Germany). The refractive index was recorded at 20°C and subsequently converted into moisture content using the Chataway table (Chataway, 1932; AOAC, 2012).

2) Sugar contents

Determination of sugars (fructose, glucose, and sucrose) in honey samples was conducted using the modified method of Jalaludin and Kim (2021). 1 g of each sample was diluted with 50 mL of water and vortexed until fully dissolved. The solution was filtered through a 0.20 μm PTFE syringe filter and 10 μL of the solution was injected into HPLC system equipped with refractive index detector (Agilent 1260 series, Agilent Technologies, Santa Clara, California, USA). The system utilized an amino column (Asahipak NH2P-50 4E, 250×4.6 mm, 5 μm particle size; Shodex, Tokyo, Japan) maintained at 40°C. The mobile phase was a mixture of acetonitrile and water (75:25, v/v) delivered at a flow rate of 1.0 mL/min for 15 min.

3) HMF content

HMF analysis was performed with adaptations based on the method of Khalil *et al*. (2010). 1 g of honey samples was dissolved in 10 mL of water, and vortexed to ensure complete dilution. The solution was filtered through a 0.45 μm nylon syringe filter, 20 μL of aliquot was injected into HPLC system equipped with diode array detector (Agilent 1200 series, Agilent Technologies, Santa Clara, California, USA). HMF was separated using a reverse-phase column (CAPCELL PACK C18 MG, 250×4.6 mm, 5 μm particle size; Shiseido, Tokyo, Japan) operated at 40°C. The mobile phase consisted of water and methanol in a 9:1 (v/v) ratio, delivered at a flow rate of 1.0 mL/min for 10 min. Detection was performed at a wavelength of 280 nm.

4) Stable carbon isotope ratio

For SCIR analysis, approximately 1 mg of honey sample was sealed in a tin capsule and combusted in a Flash 2000 Elemental Analyzer (Thermo Scientific, Bremen, Germany) coupled to a DELTA V Advantage Isotope Ratio Mass Spectrometer (Thermo Scientific, Bremen, Germany). The analysis was conducted following the method described by Boschker *et al*. (1999) and Moerdijk-Poortvliet *et al*. (2015).

4. Statistical analysis

Differences in quality parameters among honey samples were analyzed using one-way ANOVA followed by Tukey's post hoc test in R software (version 4.4.1). Prior to analysis, the assumptions of ANOVA were tested: normality was assessed using the Shapiro-Wilk test, and homogeneity of variances was evaluated using Levene's test. In addition to comparing floral origins, the analysis also aimed to detect significant differences based on local resources. A *p*-value of less than 0.01 was considered statistically significant. Distinct letters denote significant differences between groups. All measurements were performed in triplicate for each sample, and the reported values represent the mean ± standard deviation and range (min–max).

RESULTS

1. Pollen identification

Based on melissopalynological analysis, the honey samples were classified into 14 floral origin groups: *Alhagi* (n=7), *Helianthus* from Uzbekistan (n=4), *Helianthus* from Russia (n=3), *Medicago* from Uzbekistan (alfalfa; n=4), *Medicago* from Russia (n=1), *Salvia* (n=2), *Ferula* (n=1), *Onobrychis* (n=1), *Tilia* (n=1), Multifloral (*Helianthus*) (n=2), Multifloral (*Alhagi*) (n=1), Multifloral (*Calligonum*) (n=1), Multifloral (*Ferula*) (n=1), and Multifloral (*Onobrychis*) (n=1) (Table 2).

2. Moisture content

There was a statistically significant difference in moisture content among the honey samples depending on floral origin ($P<0.001$; Table 2). *Medicago* (R) honey exhibited the highest moisture content, followed by Multifloral (*Ferula*). *Salvia* honey showed the lowest moisture content; on average, the moisture content across all samples was around 18%.

Table 2. Physicochemical characteristics, including moisture, stable carbon isotope ratio (SCIR), hydroxymethylfurfural (HMF), fructose, glucose, reducing sugar, sucrose, and fructose-to-glucose ratio (F/G), of honey samples collected in Tashkent, Uzbekistan, in 2022, grouped by botanical origin identified through melissopalynological analysis

Floral source	n	Moisture (%)	SCIR[a] (‰)	HMF (mg/kg)	Fructose (%)	Glucose (%)	Reducing sugar (%)	Sucrose (%)	F/G[b]
Alhagi	7	17.0 ± 1.07 *bcd* (15.4, 18.4)	−25.0 ± 1.13 *abc* (−26.3, −22.4)	0.1 ± 0.07 *bc* (0.0, 0.2)	38.8 ± 2.21 *ab* (36.2, 43.5)	33.0 ± 2.46 *abc* (28.3, 36.0)	71.8 ± 2.44 *ab* (67.3, 74.5)	2.4 ± 1.25 *b* (0.8, 4.4)	1.2 ± 0.15 *ab* (1.0, 1.5)
Helianthus (U[c])	4	18.3 ± 1.60 *abcd* (15.9, 20.4)	−25.6 ± 0.29 *bc* (−26.1. −25.2)	0.1 ± 0.08 *bc* (0.0, 0.2)	38.0 ± 1.55 *ab* (36.0, 40.1)	32.5 ± 2.26 *bc* (29.6, 35.4)	70.5 ± 2.44 *ab* (67.3, 74.1)	1.9 ± 1.66 *b* (0.0, 4.4)	1.2 ± 0.11 *ab* (1.1, 1.4)
Helianthus (R[d])	3	18.8 ± 1.06 *abc* (17.7, 20.2)	−23.8 ± 1.95 *ab* (−25.5, −21.2)	0.9 ± 1.26 *ab* (0.0, 2.7)	38.2 ± 0.34 *ab* (37.8, 38.7)	35.9 ± 1.18 *ab* (34.3, 37.1)	74.0 ± 1.20 *ab* (72.3, 74.9)	0.9 ± 0.7 *b* (0.0, 1.7)	1.1 ± 0.04 *b* (1.0, 1.1)
Medicago (U)	4	16.9 ± 0.52 *abc* (16.0, 17.5)	−24.2 ± 1.28 *ab* (−25.3, −22.2)	0.0 ± 0.04 *bc* (0.0, 0.1)	39.0 ± 1.20 *ab* (37.5, 40.8)	34.9 ± 0.69 *ab* (34.1, 35.8)	73.8 ± 1.59 *ab* (72.6, 76.6)	2.4 ± 1.18 *b* (1.3, 4.4)	1.1 ± 0.03 *ab* (1.1, 1.2)
Medicago (R)	1	20.4 ± 0.15 *a* (20.2, 20.5)	−26.0 ± 0.03 *bc* (−26.0, −26.0)	ND[e]	37.8 ± 0.09 *ab* (37.7, 37.9)	34.3 ± 0.15 *abc* (34.2, 34.5)	72.2 ± 0.22 *ab* (72.0, 72.4)	1.5 ± 0.06 *b* (1.5, 1.6)	1.1 ± 0.00 *ab* (1.1, 1.1)
Salvia	2	16.3 ± 0.53 *d* (15.7, 16.8)	−25.7 ± 0.41 *bc* (−26.1, −25.2)	0.1 ± 0.08 *bc* (0.0, 0.2)	38.6 ± 0.19 *ab* (38.3, 38.8)	33.7 ± 1.55 *abc* (32.3, 35.3)	72.3 ± 1.73 *ab* (70.6, 74.0)	2.8 ± 0.89 *b* (1.9, 3.7)	1.1 ± 0.05 *ab* (1.1, 1.2)
Ferula	1	17.7 ± 0.10 *bcd* (17.6, 17.8)	−25.3 ± 0.17 *bc* (−25.5, −25.1)	ND	38.6 ± 1.31 *ab* (37.8, 40.2)	35.8 ± 0.05 *ab* (35.7, 35.8)	74.4 ± 1.27 *a* (73.6, 75.9)	ND	1.1 ± 0.04 *b* (1.1, 1.1)
Onobrychis	1	17.6 ± 0.10 *bcd* (17.5, 17.7)	−24.9 ± 0.34 *abc* (−25.3, −24.6)	0.0 ± 0.00 *bc* (0.0, −0.0)	34.1 ± 0.06 *c* (34.1, 24.2)	30.4 ± 0.07 *c* (30.4, 30.5)	64.6 ± 0.08 *c* (64.5, 64.6)	5.6 ± 0.11 *a* (5.5, 5.7)	1.1 ± 0.00 *ab* (1.1, 1.1)
Tilia	1	17.9 ± 0.10 *bcd* (17.8, 18.0)	−25.9 ± 0.01 *bc* (−25.9, −25.9)	ND	38.5 ± 0.08 *ab* (38.4, 38.6)	33.0 ± 0.15 *abc* (32.8, 33.1)	71.5 ± 0.11 *ab* (71.4, 71.6)	2.9 ± 0.06 *b* (2.9, 3.0)	1.2 ± 0.01 *ab* (1.2, 1.2)
Multifloral (*Helianthus*)	2	18.9 ± 0.14 *abc* (18.7, 19.0)	−25.0 ± 0.36 *abc* (−25.4, −24.6)	0.2 ± 0.05 *bc* (0.1, −0.2)	37.6 ± 0.27 *ab* (37.3, 37.9)	35.2 ± 1.11 *ab* (34.2, 36.3)	72.8 ± 1.38 *ab* (71.5, 74.2)	1.2 ± 1.35 *b* (0.0, 2.6)	1.1 ± 0.03 *b* (1.0, 1.1)
Multifloral (*Alhagi*)	1	16.9 ± 0.10 *bcd* (16.8, 17.0)	−26.0 ± 0.21 *bc* (−26.3, −25.8)	1.2 ± 0.07 *a* (1.2, −1.3)	40.6 ± 0.25 *a* (40.4, 40.9)	33.4 ± 0.11 *abc* (33.3, 33.5)	74.0 ± 0.36 *ab* (73.7, 74.4)	1.1 ± 0.08 *b* (1.0, 1.2)	1.2 ± 0.00 *ab* (1.2, 1.2)
Multifloral (*Calligonum*)	1	18.1 ± 0.10 *bcd* (18.0, 18.2)	−22.8 ± 0.10 *a* (−22.9, −22.7)	0.0 ± 0.01 *bc* (0.0, −0.0)	37.7 ± 0.30 *ab* (37.4, 38.0)	32.2 ± 0.15 *bc* (32.1, 32.4)	70.0 ± 0.16 *b* (69.8, 70.1)	2.8 ± 0.09 *b* (2.7, 2.9)	1.2 ± 0.01 *ab* (1.2, 1.2)
Multifloral (*Ferula*)	1	19.1 ± 0.10 *ab* (19.0, 19.2)	−27.1 ± 0.34 *c* (−27.5, −26.8)	ND	37.2 ± 0.09 *bc* (37.1, 37.3)	36.5 ± 0.10 *a* (36.4, 36.6)	73.7 ± 0.16 *ab* (73.6, 73.9)	2.2 ± 0.0 *b* (2.1, 2.2)	1.0 ± 0.0 *b* (1.0, 1.0)
Multifloral (*Onobrychis*)	1	18.4 ± 0.10 *abcd* (18.3, 18.5)	−27.1 ± 0.18 *c* (−27.3, − 27.0)	0.0 ± 0.00 *c* (0.0, 0.0)	28.4 ± 0.30 *d* (28.1, 28.7)	21.9 ± 0.15 *d* (21.8, 22.1)	50.3 ± 0.40 *d* (50.0, 50.8)	1.0 ± 0.10 *b* (0.9, 1.1)	1.3 ± 0.01 *a* (1.3, 1.3)
Total	30	61.7 ± 21.39 (13.0, 100)	−25.1 ± 1.32 (−27.5, −21.2)	0.2 ± 0.51 (0.0, 2.7)	38.0 ± 2.41 (28.1, 43.5)	33.4 ± 3.00 (21.8, 37.1)	71.4 ± 4.69 (50.0, 76.6)	2.3 ± 1.5 (0.0, 5.7)	1.1 ± 0.10 (1.0, 1.5)
$F_{(13,76)}$		7.635	6.016	$F_{(9,68)} = 5.363$	14.966	16.211	39.539	$F_{(12,74)} = 4.69$	2.889
P		< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	< 0.001	0.002

Values are presented as mean ± standard deviation, along with the range (min–max). Different superscript letters within each column indicate significant differences among floral groups (one-way ANOVA followed by Tukey's HSD test, $P < 0.01$).
[a]SCIR indicates stable carbon isotope ratio.
[b]F/G means fructose to glucose ratio.
[c]U indicates honey samples from Uzbekistan
[d]R indicates honey samples from Russia
[e]ND indicates not detected.

In addition to the variations across floral origins, moisture content also differed significantly according to the local origin of the honey ($P<0.001$; Table 3). Samples obtained from supermarkets exhibited the highest average moisture content, which was significantly greater than that of honeys collected from apiaries, markets, and TSAU.

3. Stable carbon isotope ratio

In Table 2, the SCIR significantly differed among the honey samples according to floral origin ($P<0.001$), whereas no statistically significant difference was observed based on local sources in Table 3 ($P=0.057$). Multifloral (*Calligonum*) exhibited the highest ratio, whereas Multifloral (*Ferula*) and Multifloral (*Onobrychis*) showed the lowest values. All samples fell within the overall range of −27.5 to −21.2‰.

1) HMF content

While the HMF content significantly varied among floral groups ($P<0.001$; Table 2), no such difference was observed when the samples were grouped by local resource ($P=0.097$; Table 3). The HMF content of the honey samples ranged from 0.0 to 2.7 mg/kg. No HMF was detected in the *Medicago* (R), *Ferula*, *Tilia*, and Multifloral (*Ferula*) samples, while Multifloral (*Alhagi*) exhibited a noticeably higher HMF value compared to all other samples.

2) Sugar contents

Significant differences were observed among the honey samples in terms of fructose, glucose, reducing sugar, and sucrose contents ($P<0.001$; Table 2), as well as in the fructose-to-glucose (F/G) ratio ($P=0.002$). Multifloral (*Alhagi*) exhibited the highest fructose content among the samples, and it was significantly different from Multifloral (*Ferula*), *Onobrychis*, and Multifloral (*Onobrychis*). Regarding glucose, Multifloral (*Ferula*) showed the highest content, while Multifloral (*Onobrychis*) had the lowest. A similar trend was identified for reducing sugar. Sucrose was not detected in the *Ferula* sample, whereas *Onobrychis* showed the highest sucrose content compared with all other samples. The F/G ratio was highest in Multifloral (*Onobrychis*), while *Helianthus* (R), *Ferula*, Multifloral (*Helianthus*), and Multifloral (*Ferula*) exhibited the lowest values.

Table 3. Physicochemical characteristics, including moisture, stable carbon isotope ratio (SCIR), hydroxymethylfurfural (HMF), fructose, glucose, reducing sugar, sucrose, and the fructose-to-glucose ratio (F/G), of honey samples collected in Tashkent, Uzbekistan, in 2022, grouped by local source (apiaries, supermarkets, markets, and TSAU)

Local source	N	Moisture (%)	SCIR[a] (‰)	HMF (mg/kg)	Fructose (%)	Glucose (%)	Reducing sugar (%)	Sucrose (%)	F/G[b]
Apiary	9	17.2 ± 1.18 *b* (15.6, 18.8)	−25.1 ± 1.07 (−26.1, −22.2)	0.1 ± 0.09 (0.0, 0.2)	38.5 ± 0.77 (37.5, 40.1)	34.3 ± 1.97 (29.6, 36.3)	72.8 ± 1.48 (69.6, 74.3)	1.9 ± 1.26 (0.0, 4.4)	1.1 ± 0.09 *b* (1.0, 1.4)
Supermarket	9	18.7 ± 1.26 *a* (16.7, 20.5)	−24.6 ± 1.59 (−26.1, −21.2)	0.5 ± 0.91 (0.0, 2.7)	37.3 ± 1.34 (34.1, 38.7)	33.8 ± 2.05 (30.4, 37.1)	71.1 ± 3.20 (64.5, 74.9)	2.3 ± 1.69 (0.0, 5.7)	1.1 ± 0.04 *b* (1.0, 1.2)
Market	7	17.4 ± 0.81 *b* (15.9, 18.4)	−25.3 ± 1.10 (−26.3, −22.7)	0.2 ± 0.44 (0.0, 1.3)	38.4 ± 1.47 (36.0, 40.9)	33.1 ± 1.20 (31.1, 34.7)	71.5 ± 2.52 (67.3, 74.5)	2.6 ± 1.10 (1.0, 4.4)	1.2 ± 0.03 *ab* (1.1, 1.2)
TSAU	5	17.6 ± 1.28 *b* (15.4, 19.2)	−25.7 ± 1.29 (−27.5, −24.0)	0.0 ± 0.02 (0.0, 0.1)	37.6 ± 5.27 (28.1, 43.5)	31.7 ± 5.89 (21.8, 36.6)	69.3 ± 9.95 (50.0, 76.6)	1.9 ± 0.70 (1.0, 2.9)	1.2 ± 0.19 *a* (1.0, 1.5)
Total	30	17.8 ± 1.29 (15.4, 20.5)	−25.1 ± 1.32 (−27.5, −21.2)	0.2 ± 0.51 (0.0, 2.7)	38.0 ± 2.41 (28.1, 43.5)	33.4 ± 3.00 (21.8, 37.1)	71.4 ± 4.69 (50.0, 76.6)	2.3 ± 1.5 (0.0, 5.7)	1.1 ± 0.10 (1.0, 1.5)
$F_{(3,86)}$		9.059	2.609	$F_{(3,74)}=2.187$	1.460	2.933	1.883	$F_{(3,83)}=1.176$	4.566
P		<0.001	0.057	0.097	0.231	0.038	0.138	0.324	0.005

Values are presented as mean ± standard deviation, along with the range (min–max). Different superscript letters within each column indicate significant differences among floral groups (one-way ANOVA followed by Tukey's HSD test, $P<0.01$).
[a]SCIR indicates stable carbon isotope ratio.
[b]F/G means fructose to glucose ratio.

Although no significant differences were found in the levels of fructose ($P=0.231$), glucose ($P=0.038$), sucrose ($P=0.324$), or reducing sugars ($P=0.138$) across local sources (Table 3), the F/G ratio differed significantly ($P=0.005$). TSAU samples showed the highest F/G ratio, which was significantly greater than that of the sampels from apiaries and supermarkets.

DISCUSSION

Water, the second most abundant component in honey, affects its physical characteristics, including viscosity and crystallization, as well as other quality parameters such as color, aroma, taste, density, solubility, and shelf life (Escuredo *et al*., 2013). In this study, the moisture content of the honey samples exhibited statistically significant differences, with values ranging from 15% to 20%. Yücel and Sultanoglu (2013) reported values ranging from 15 to 21%, depending on the botanical origin of the honey, the degree of hive maturation, processing methods, and storage conditions. Moreover, honey with a moisture content below 20% is considered acceptable for commercial sale due to its resistance to fermentation (Codex Alimentarius, 2001). All the samples produced in Tashkent met this criterion, except for the *Medicago* sample from Russia, indicating generally good post-harvest handling and storage practices in Uzbekistan. In addition, among the four sources, only supermarket honeys exhibited significantly higher moisture content, whereas samples from apiaries, traditional markets, and the university showed comparable levels. This elevated moisture may be attributed to the hygroscopic nature of honey and prolonged exposure to ambient humidity during distribution or storage in indoor retail settings (Karabagias *et al*., 2014). However, further investigation is required to confirm the specific environmental factors responsible.

Carbon isotope analysis is used to detect sugar adulteration in honey by distinguishing plant types based on their $^{13}C/^{12}C$ ratios, which reflect different photosynthetic pathways. C_3 plants (e.g., sugar beet, apple, grapes) show $\delta^{13}C$ values from −33 to −22‰, C_4 plants (e.g., sugarcane, corn) from −20 to −10‰, and CAM plants (e.g., pineapple, cactus) from −13.5 to −11.0‰ (Padovan *et al*., 2007; Çinar *et al*., 2014). Under AOAC official methods 978.17 and 998.12, honey is considered adulterated when its CSIR value is greater than −23.5‰ (AOAC, 1979, 1998). Although the value of −22.8‰ exceeds the authenticity threshold, this alone is insufficient to confirm adulteration; however, it does raise concern and warrants further investigation. HMF is widely recognized as a marker of honey deterioration. It is primarily formed through the degradation of monosaccharides or via the *Maillard* reaction during prolonged storage or exposure to heat (da Silva *et al*., 2016). In the present study, HMF levels in the samples were generally very low, ranging from 0.0 to 2.7 mg/kg. Several samples, including *Medicago* (R), *Ferula*, *Tilia*, and Multifloral (*Ferula*), showed non-detectable levels. These values fall well below the international quality limit of 40 mg/kg established by Codex Alimentarius (2001). Such low concentrations of HMF suggest that the majority of the samples were either recently harvested or stored under appropriate temperature conditions, with minimal thermal degradation.

Sugars in honey contribute to properties such as energy value, viscosity, hygroscopicity and granulation (Kamal and Klein, 2011). Monosaccharides, mainly fructose and glucose, typically constitute around 75% of the total sugar content in honey, while disaccharides account for 10−15%, with trace amounts of other sugars. In most honeys, the fructose content typically exceeds that of glucose, although exceptions have been reported in specific floral types such as rape and dandelion (Escuredo *et al*., 2013). Codex Alimentarius (2001) stipulates that floral honeys must contain a minimum of 60% reducing sugar. In our study, all samples displayed a higher proportion of fructose than glucose. While most samples met the requirement, only the Multifloral (*Onobrychis*) sample fell below the 60% reducing sugar threshold. The general sugar composition is largely influenced by the botanical origin of the nectar, the geographical region, and can be further affected by climate conditions, processing methods, and storage duration (Tornuk *et al*., 2013; Escuredo *et al*., 2013). Sucrose levels, in particular, are considered an important indicator of possible adulteration with low-cost sweeteners (da Silva *et al*., 2016). To address this, Codex Alimentarius (2001) sets the maximum acceptable sucrose concentration in floral honeys at 5%. All analyzed samples were found to be within acceptable sucrose levels, except for

a single *Onobrychis* honey sample which exceeded the 5% threshold. Taken together, the data confirmed that most samples meet established quality criteria, although isolated deviations underscore the influence of floral and environmental factors on sugar composition.

In addition, the F/G ratio is commonly used as an indicator of honey crystallization tendency, as glucose is less soluble in water than fructose (Escuredo *et al*., 2013). In this study, the F/G ratio varied notably among the samples; Multifloral (*Onobrychis*) exhibited the highest ratio, suggesting a lower tendency to crystallize. In contrast, samples such as *Helianthus* (R), *Ferula*, Multifloral (*Helianthus*), and Multifloral (*Ferula*) showed lower F/G ratios, implying a greater potential for granulation. These differences likely reflect variations in nectar composition among floral sources, which in turn may influence consumer preference and shelf stability. Moreover, the highest F/G values were recorded in samples from TSAU, suggesting that post-harvest handling, storage duration, or environmental exposure may contribute to subtle shifts in sugar balance regardless of floral origin (Tornuk *et al*., 2013; Escuredo *et al*., 2013; da Silva *et al*., 2016).

Overall, the physicochemical characteristics and adulteration markers analyzed in this study reveal considerable variability among honey samples of different floral origins available in Tashkent. While most samples complied with international quality standards, specific deviations, such as low reducing sugar, elevated sucrose, or borderline CSIR values, underscore the influence of their botanical sources. In contrast, only limited differences were observed across local sources, with statistically significant variation detected in a few parameters such as moisture content and the F/G ratio. These selective differences suggest that post-harvest handling or storage conditions associated with distribution channels may contribute to minor shifts in sugar balance or moisture stability. These findings highlight the need for integrated quality management that considers not only botanical authenticity but also environmental and logistical factors affecting honey throughout production and distribution. From an industrial perspective, the results provide a basis for developing regional quality certification schemes and marketing strategies that can enhance consumer trust and export potential. Moreover, the observed compliance with Codex standards in most samples suggests that Uzbekistan's honey sector has a promising foundation for alignment with international quality frameworks, which could inform future policy-making and regulatory development.

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