



Original Article

## Assessment of essential minerals and physico-chemical analysis of floral origins fresh honey produced by *Apis mellifera*

Avaliação de minerais essenciais e análise físico-química do mel fresco de origem floral produzido por *Apis mellifera*

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### Abstract

Honey is one of the best nutritious substances in the world, having different services in the body functions regulation. Ten elements (K, Na, Ca, Co, Cr, Mn, Mo, Ni, Pb, Se) from honey samples were analyzed from 80 different locations of Punjab and ten floras. The aim of the present study was to determine the quality and quantity of minerals and Physico-chemical analysis in honey. A flame photometer was used to measure the concentration of major minerals (K, Ca and Na). The concentration of micro minerals (Co, Cr, Mn, Mo, Ni, Pb and Se) was analyzed using Atomic Absorption Spectrometer. The concentration of macro-elements obtained was as follow (in ppm): K (166-1732), Na (107-418) and Ca (07-99), while the concentration of microelements (in ppm) Co (1-2), Cr (>1), Mn (<1), Mo (1.818), Ni (1.911), Pb (<1) and Se (1.968). The most abundant minerals were potassium, calcium and sodium, ranging between 396-810.5, 17.5-640.63 and 169.88-238.62 ppm, respectively. However, the trace mineral elements of honey were obtained in the order of decreasing Se > Co > Ni > Pb > Cr > Mo > Mn. The findings showed that all the heavy metals like Co, Cr, Ni and Pb were present in trace amounts and close to International Honey Quality Standard. The result of given honey samples represented highest value of moisture (31.23%), color (80 mm pfund), pH (8.23), acidity (72.02 meq/kg), electrical conductivity (0.85 ms/cm) and ash contents (0.83%).

**Keywords:** honey, microelements, macro-elements, flora.

### Resumo

O mel é uma das substâncias mais nutritivas do mundo, possuindo diversos serviços na regulação das funções do organismo. Dez elementos (K, Na, Ca, Co, Cr, Mn, Mo, Ni, Pb, Se) de amostras de mel foram analisados em 80 locais diferentes de Punjab e dez floras. O objetivo do presente estudo foi determinar a qualidade e quantidade de minerais e análises físico-químicas em mel. Um fotômetro de chama foi usado para medir a concentração dos principais minerais (K, Ca e Na). A concentração de microminerais (Co, Cr, Mn, Mo, Ni, Pb e Se) foi analisada utilizando espectrômetro de absorção atômica. A concentração de macroelementos obtida foi a seguinte (em ppm): K (166-1732), Na (107-418) e Ca (07-99), enquanto a concentração de microelementos (em ppm) Co (1-2), Cr (> 1), Mn (< 1), Mo (1,818), Ni (1,911), Pb (< 1) e Se (1,968). Os minerais mais abundantes foram potássio, cálcio e sódio, variando entre 396-810,5, 17,5-640,63 e 169,88-238,62 ppm, respectivamente. No entanto, os oligoelementos do mel foram obtidos na ordem decrescente Se > Co > Ni > Pb > Cr > Mo > Mn. Os resultados mostraram que todos os metais pesados como Co, Cr, Ni e Pb estavam presentes em quantidades vestigiais e próximos ao Padrão Internacional de Qualidade do Mel. O resultado de determinadas amostras de mel representou o maior valor de umidade (31,23%), cor (80 mm pfund), pH (8,23), acidez (72,02 meq/kg), condutividade elétrica (0,85 ms/cm) e teor de cinzas (0,83%).

**Palavras-chave:** mel, microelementos, macroelementos, flora.

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## 1. Introduction

Honeybees are biological engineers who convert the raw material (flower nectars, plant excretions of plants licking insects) by modifying them via increasing their specific compounds (Andrade et al., 2018). Honeybees belong to order Hymenoptera, family Apidae and genus *Apis*. There are 4 species, including *Apis mellifera*, *Apis cerana*, *Apis dorsata* and *Apis florea* exhibited in Pakistan (Qamer et al., 2013; Sajid et al., 2020). Approximately 25,000 bee species participate in food crop manufacturing and enhancing biodiversity (Gould, 2015). *A. mellifera* enhances crop amounts and fertilization in plants and shows an important function in the honey farming food supply chain. Honey has become one of the most horticulture commodities available commercially in the world and Pakistan at current times (Munawar et al., 2009; Waghchoure-Camphor and Martin, 2009; Adnan et al., 2014; Anjum et al., 2015). Honey is described as “the delicious substance from the nectar of flowers formed by collection, processing and deposition in honeycombs by the bees”. Honey is the most important natural product and graded because of its large dietary contents (330 kcal/100 g) and rapid carbohydrate digestion (Abdulkhalik and Swaileh, 2017; Sajid et al., 2020).

Honey's chemical constitution and characteristics depend on the kinds of plants visited by honeybees and the environmental factors under which the crops are cultivated. Substances such as toxic metals or other chemicals in honey are created by human actions in that particular area from which they are extracted (Pohl et al., 2009, 2012). Honey has a higher nutrient value based on the high percentage of carbohydrates, including maltose, sucrose, fructose, glucose, oligosaccharides, polysaccharides, enzymes, pigments, aroma compounds, pollen grains, minerals, waxes, vitamins and acids (Haouam et al., 2016; Conti et al., 2018). Honey's botanical and regional source is largely decided by its constitution as the raw material and plant's capital portion strongly affected the honey production (Pohl, 2009). The chemical composition of honey also depends on several ingredients, including nectar sources afflicted by environmental conditions, botanical origin of floral sources, soil conditions and geographical origin (Solayman et al., 2016). Honey composition depends on the various plant types, ecological and climatic factors and effective methods of apiculture. Mostly honey comprises water, sugar, proteins, enzymes, nutrients, flavoring agents, phenolic constituents and more than 200 other compounds in trace amounts (Silva et al., 2009).

A wide range of parameters such as sugar, hydroxyl-methyl furfural (HMF), color, pH, humidity, minerals and pollen contents are assumed to differentiate the quality of honey from various regions (Meda et al., 2005; Njokuocha et al., 2019). During honey production, the nature and performance of the honey depend on many ecological factors, including temperature, humidity, and honeydew concentrations in honeycomb. Honey quality depends on different parameters such as humidity, vitamins, amino acids, enzyme activity, electrical conductivity and solid corpuscles (Tuzen et al., 2007; Kiliç Altun et al., 2017; Njokuocha et al., 2019).

Honey is a divine food and is prescribed as the best treatment for allergies (Purbafrani et al., 2014) and gastric ulceration (Al-Waili et al., 2005; Mandal and Mandal, 2011). The ancient Egyptians and Greeks utilized honey as medicine to cure various diseases like gastrointestinal disorders such as burns, ulcers and healing wounds (Pećanac et al., 2013). It plays an important role in producing blood cells, purifying blood, controlling and promoting blood flow, and providing useful protection against arteriosclerosis. It also has essential antioxidant compounds such as organic acids, aromatic acids, ascorbic acid, catalases, flavonoids, glucose oxidase and compounds of carotenoids (Münstedt et al., 2008; Khalil et al., 2010).

Honey comprises essential nutrients and major minerals that perform vital functions (Bogdanov, 2012; Solayman et al., 2016). Macrominerals have been described as being extracted through flowering plants and land. Still, they have also been analyzed to be derived from human-induced resources because different compounds prevail in the atmosphere and pollute the environment. It has also been recognized that trace minerals obtained from flowering plant products play an important role in human welfare (Pohl, 2009). Honey's role as nutrition gives a better body production and a good accumulation of minerals for bone strength (Chepulis and Starkey, 2008; Solayman et al., 2016). The total number of minerals discovered in honey is about 54 till now. The minerals of honey can be categorized as major and trace minerals and elements that are needed in small amounts for organism metabolism are minor minerals (Pohl et al., 2009, 2012; Solayman et al., 2016). Keeping in view the importance of honey, present work had been planned to determine the quality and quantity of minerals in honey of different floral sources and areas of Pakistan and compare mineral levels of fresh Pakistani Honey with International Honey Standards.

## 2. Materials and Methods

### 2.1. Collection of samples

Different samples of honey were collected from the different locations of Punjab, Pakistan. Airtight plastic bottles were used to preserve these samples at room temperature. In order to determine the quality of minerals in fresh honey, 80 fresh honey samples were collected directly from beekeepers at various sites or areas or districts throughout Punjab. The concentration of macro minerals (K, Na, Ca) and micro minerals (Co, Cr, Mn, Mo, Ni, Pb, Se) was analyzed by Flame Photometer and Atomic Absorption Spectrophotometer and was compared with International Standards shown in (Table 1) (Bogdanov, 2012).

### 2.2. Sample preparation

To examine major and trace minerals, 2-3 g of honey was collected in a beaker and weighted on electrical balance. Then added 10 mL of Nitric acid ( $\text{HNO}_3$ ) in it and heated at 160 °C temperature to evaporate  $\text{HNO}_3$ . Subsequently, 5 mL of Perchloric acid ( $\text{HClO}_4$ ) was added and heated until complete evaporation or colorless fumes

**Table 1.** Codex Alimentarius limits for macro and micro elements in honey.

Minerals	Codex Alimentarius Limits
Potassium ( <b>K</b> )	250-2500 ppm
Calcium ( <b>Ca</b> )	200-2300 ppm
Sodium ( <b>Na</b> )	300-1000 ppm
Cobalt ( <b>Co</b> )	<1 ppm
Chromium ( <b>Cr</b> )	<1 ppm
Manganese ( <b>Mn</b> )	<1 ppm
Molybdenum ( <b>Mo</b> )	<1 ppm
Nickel ( <b>Ni</b> )	<1 ppm
Lead ( <b>Pb</b> )	<1 ppm
Selenium ( <b>Se</b> )	<1 ppm

came out. The remaining solution was just 2 ml in the beaker then placed in the water bath for one hour at 100 °C for complete digestion. Eventually, the samples were filtered and transferred to 50 mL Eppendorf tube filled with distilled water up to mark. This procedure was carried out in triplicate (Grembecka and Szefer, 2013; Magna et al., 2018).

### 2.3. Standards for solutions

A stock solution of K, Ca, Na, Co, Cr, Mn, Mo, Ni, Pb and Se were 1000 ppm concentration. The working solutions of different concentrations were prepared from this stock solution using the following Equation 1.

$$C_1V_1 = C_2V_2 \quad (1)$$

where:  $C_1$  = Standard stock solution concentration;  $V_1$  = Standard stock solution volume;  $C_2$  = Working standard stock solution concentration;  $V_2$  = Standard stock solution of working volume.

#### 2.3.1. Standard solution for Sodium (Na)

Working standard solutions of concentration 50 ppm, 75 ppm and 100 ppm were used. These solutions were made following the Formula 2.

$$C_1V_1 = C_2V_2 \quad (2)$$

where:  $C_1 = 50 \times 25 / 1000$ ;  $C_1 = 1.25 \text{ mL}$ ;  $C_1 = 1250 \text{ } \mu\text{L}$ .

The standard stock solution (1250  $\mu\text{L}$ ) was dissolved in distilled water and the final volume was 25 mL. This was a working standard solution for 50 ppm concentration and 1875  $\mu\text{L}$  of stock solution was used to prepare the 75 ppm concentration. Similarly, 2500  $\mu\text{L}$  of stock solution prepared 100 ppm concentration. The final and overall volume in each case was 25 mL.

#### 2.3.2. Standard for Potassium (K)

Potassium was determined in *Apis mellifera* fresh honey by using the working standard concentration of 50 ppm, 75 ppm and 100 ppm, respectively.

#### 2.3.3. Standard for Calcium (Ca)

For calculating calcium, the working solution of 50 ppm, 75 ppm and 100 ppm were prepared.

#### 2.3.4. Standard for Cobalt (Co)

Similarly, cobalt (Co) was measured using the 1 ppm, 2 ppm, 4 ppm, and 5 ppm working solution of the same element.

#### 2.3.5. Standard for Chromium (Cr)

The working solution of chromium (Cr) had the concentration of 0.1 ppm, 0.5 ppm, 3 ppm and 5 ppm to measure this element in honey.

#### 2.3.6. Standard for Manganese (Mn)

Manganese (Mn) in *Apis mellifera* fresh honey was estimated by a working solution of 1 ppm, 2 ppm, 3 ppm and 4 ppm concentration.

#### 2.3.7. Standard for Molybdenum (Mo)

The standard working solution of 0.1 ppm, 0.2 ppm, 0.3 ppm, and 0.5 ppm strength was used to determine honey's molybdenum (Mo) concentration.

#### 2.3.8. Standard for Nickel (Ni)

Nickel (Ni) was determined in *Apis mellifera* fresh honey using the working standard with the concentration of 0.5 ppm, 1 ppm, 1.5 ppm and 2 ppm, respectively.

#### 2.3.9. Standard for Lead (Pb)

The working standard of 0.1 ppm, 0.4 ppm, 2 ppm, 3 ppm and 5 ppm were used to calculate lead (Pb) in fresh honey samples.

#### 2.3.10. Standard for Selenium (Se)

The honey samples were subjected to analyze selenium (Se) in honey using the working standard of 0.5 ppm, 1.5 ppm, 2.5 ppm and 3 ppm concentration.

### 2.4. Flame photometer

An instrument used in inorganic chemical analysis to determine the concentration of certain metal ions of different macro minerals. Samples were subjected to the Flame Photometer to compare some minerals Potassium, Sodium and Calcium.

### 2.5. Atomic absorption spectrometer

Atomic absorption spectrometry is an analytical technique that measures the concentration of elements. It is so sensitive that it can measure down to parts per billion of a gram in a sample. The technique uses the wavelengths of light specifically absorbed by an element.

For analysis of micro minerals, samples were subjected to the Atomic Absorption Spectrometer. Operational conditions are given in Table S1 (Supplementary Material).

### 2.6. Physicochemical analysis

Moisture contents, color, pH values, total acidity, electrical conductivity and ash contents were analyzed according to the method of (Bogdanov et al., 2002).

### 2.7. Statistical analysis

The data of all honey samples were analyzed to the statistically ANOVA one-way using Tukey's test. The data was analyzed at the significance level 0.05.

## 3. Results

### 3.1. Macro elements

#### 3.1.1. Potassium (K)

Potassium (K) concentration recorded the highest 1732 ppm in honey sample 52 collected from Narowal, whereas the lowest 166 ppm in honey sample 5 from Sheikhpura. Overall, honey samples 5 and 39 had K less than International Standard (250-2500 ppm). While samples 1, 2, 4, 12, 14, 24 and 25 had K in the 201-300 ppm range. Similarly, samples 3, 6, 8, 11, 15, 16, 23, 26, 27, 30, 31, 32, 33, 36, 38, 41, 44, 45, 46 and 50 were in the range of 301-400 ppm. In addition, samples 9, 10, 17, 19, 20, 22, 34 and 47 were in the range of 401-500 ppm. The range of 501-600 ppm for K was observed in honey sample 29, 35, 37, 48, 54, 58, 62 and 79. The range of 601-700 ppm for K had observed in honey samples No. 43, 49, 61, 66, 68, 69, 74 and 75, respectively. Moreover, range of K of 701-1000 ppm was estimated in samples 28, 40, 51, 53, 55, 57, 59, 60, 63, 64, 67, 71, 73, 76, 77, 78 and 80, respectively. Moreover, the range of K of 1001-1800 ppm was estimated in samples 7, 13, 18, 21, 42, 52, 56, 65, 70 and 72. The results showed that Potassium was more dominant element in 70% honey samples than other minerals (Table 2).

#### 3.1.2. Calcium (Ca)

Calcium (Ca) concentration recorded the highest 90 ppm in honey sample 52 collected from Narowal, whereas, lowest 7 ppm in Murree honey sample 10. The results revealed that Ca, being a macro element, was not more than 100 ppm in all the samples, far less than International Standard limits of 200- 2300 ppm (Table 2).

#### 3.1.3. Sodium (Na)

The amount of Na as macro-mineral was analyzed in all *Apis mellifera* fresh honey samples collected from various Punjab districts. The concentration of Na recorded highest 418 ppm in honey sample 49 collected from Hafizabad. Whereas the lowest 107 ppm in Sheikhpura honey sample 2. Overall, most honey samples had Na less than International Standard (300- 1000 ppm) (Table 2).

### 3.2. Micro elements

#### 3.2.1. Cobalt (Co)

In *Apis mellifera* fresh honey samples 1, 4, 8, 9, 10, 11, 12, 15, 21, 27, 43, 44, 45, 46, 51, 66, 67, 73, 74, 79 and 80 had Cobalt (Co) as a micro element less than 1 ppm as mentioned of International Honey Commission. Similarly, samples 2, 3, 5, 6, 7, 13, 14, 16, 17, 18, 19, 20, 22, 23, 24, 25, 26, 30, 33, 35, 37, 41, 47, 48, 49, 50, 52, 53, 54, 55, 57, 59, 60, 63, 64, 70, 71, 75, 76 and 78 had slightly higher 1-2 ppm level. Likewise, honey samples No. 28, 29, 31, 32, 34, 36, 38, 39, 40, 42, 56, 58, 61, 62, 65, 68, 69, 72 and 77 showed 23.75% (Table 2).

#### 3.2.2. Chromium (Cr)

Recommended International Standard of Chromium (Cr) in honey stated it should not be more than 1 ppm. In present study, samples 1-4, 6-16, 18-32, 38-41, 46, 47, 48, 49-57, 59, 61-80 contained Cr within International limit (<1 ppm). Likewise, honey samples No. 5, 17, 33, 34, 35, 36, 37, 42, 43, 44, 45, 58 and 60 showed Cr concentration higher than 1 ppm (Table 2).

#### 3.2.3. Manganese (Mn)

Recommended International Honey limit for Manganese (Mn) is <1 ppm. Samples 1-18, 20-26, 28-38, and 43-80 had Mn less than 1 ppm. Likewise, honey samples 19, 27, 39, 40, 41 and 42 showed Mn concentrations higher than 1 ppm. Whereas Mn was found in all samples of fresh honey (Table 2).

#### 3.2.4. Molybdenum (Mo)

The concentration of Molybdenum (Mo) was found maximum (1.818 ppm) in sample 43 collected from Sargodha, more than value set by honey standard (<1 ppm). While Mo in samples 9, 29, 33, 34, 43, 44, 45, 46, 47, 48, 49, 50, 58, 61, 75 and 78 was in the range of 1-2 ppm. Similarly, most *Apis mellifera* fresh honey samples had Mo as a microelement of less than 1 ppm (Table 2).

#### 3.2.5. Nickel (Ni)

The concentration of nickel (Ni) was found maximum (1.911 ppm) than the international limit (<1 ppm) in sample No. 3 that was collected from Sheikhpura. Nickel in samples 3, 4, 5, 6, 13, 34, 36, 37, 53 and 60 was in the range of 1-2 ppm (Table 2).

#### 3.2.6. Lead (Pb)

Lead (Pb) is considered as toxic element and its concentration should be (<1 ppm) in honey as recommended by International Commission. In present study, maximum 2.608 ppm Pb was found in sample 36 collected from the Talagang. Similarly, samples 1, 5, 11, 13, 15, 34, 40, 58 and 60 had slightly higher (1-2 ppm) Pb than international limit. Likewise, honey samples 33 and 36 showed Pb in the range of 2-3 ppm. Whereas, majority of honey samples 2, 3, 4, 6, 7, 8, 9, 10, 12, 14, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 26, 27, 28, 29, 30, 31, 32, 35, 37, 38, 39, 41, 42, 43, 44, 45, 46, 47, 48, 49, 50, 51, 52, 53, 54, 55,

**Table 2.** Concentration (ppm) of macro and micro elements in honey samples produced by *Apis mellifera* in different areas of Punjab.

Sr. No.	Dominant Flora	Locations	Concentration									
			Potassium (K) ppm	Calcium (Ca) ppm	Sodium (Na) ppm	Cobalt (Co) ppm	Chromium (Cr) ppm	Manganese (Mn) ppm	Molybdenum (Mo) ppm	Nickel (Ni) ppm	Lead (Pb) ppm	Selenium (Se) ppm
1	Orange ( <i>Citrus xsinensis</i> )	Sheikhupura	292	18	115	0.208	0.27	0.526	0.753	0.537	1.124	0.236
2	Sheesham ( <i>Dalbergia sissoo</i> )	Sheikhupura	290	21	107	1.098	0.49	0.029	0.271	0.628	0.486	0.151
3	Orange ( <i>Citrus xsinensis</i> )	Sheikhupura	353	15	125	1.319	0.063	0.204	0.072	1.911	0.449	0.447
4	Sheesham ( <i>Dalbergia sissoo</i> )	Sheikhupura	282	22	138	0.986	0.067	0.331	0.19	1.217	0.376	0.098
5	Sheesham ( <i>Dalbergia sissoo</i> )	Sheikhupura	166	10	148	1.411	1.086	0.472	0.278	1.519	1.18	0.282
6	Orange ( <i>Citrus xsinensis</i> )	Sheikhupura	312	14	151	1.851	0.655	0.163	0.403	1.476	0.658	0.185
7	Granda ( <i>Carissa carandas</i> )	Murree	1423	12	139	1.668	0.696	0.29	0.356	0.854	0.098	0.589
8	Bhaiker ( <i>Justicia adhatoda</i> )	Murree	382	19	149	0.872	0.263	0.263	0.766	0.845	0.828	0.901
9	Black locust ( <i>Robinia pseudoacacia</i> )	Chakwal	446	11	153	0.412	0.531	0.111	1.06	0.916	0.672	1.968
10	Berseem ( <i>Trifolium alexandrinum</i> )	Murree	464	7	154	0.496	0.845	0.578	0.944	0.235	0.102	0.056
11	Orange ( <i>Citrus xsinensis</i> )	Sargodha	320	17	156	0.932	0.048	0.538	0.999	0.9	1.169	1.208

Table 2. Continued...

Sr. No.	Dominant Flora	Locations	Concentration										
			Potassium (K) ppm	Calcium (Ca) ppm	Sodium (Na) ppm	Cobalt (Co) ppm	Chromium (Cr) ppm	Manganese (Mn) ppm	Molybdenum (Mo) ppm	Nickel (Ni) ppm	Lead (Pb) ppm	Selenium (Se) ppm	
12	Bhaiker ( <i>Justicia adhatoda</i> )	Kallar Kahar	297	18	150	0.359	0.203	0.52	0.327	0.984	0.099	0.768	
13	Phulai ( <i>Acacia modesta</i> )	Chakwal	1033	14	141	1.545	0.054	0.047	0.596	1.38	1.13	1.426	
14	Bhaiker ( <i>Justicia adhatoda</i> )	Choa Saidan Shah	279	26	147	1.053	0.409	0.163	0.174	0.95	0.463	1.336	
15	Orange ( <i>Citrus xsinensis</i> )	Sargodha	347	20	177	0.97	0.006	0.52	0.802	0.835	1.135	0.973	
16	Orange ( <i>Citrus xsinensis</i> )	Chakwal	366	32	203	1.933	0.701	0.236	0.171	0.764	0.159	1.557	
17	Berseem ( <i>Trifolium alexandrinum</i> )	Narowal	474	33	126	1.324	1.689	0.1	0.24	0.249	0.425	1.638	
18	Phulai ( <i>Acacia modesta</i> )	Kallar Kahar	1207	29	166	1.342	0.048	0.897	0.423	0.175	0.065	0.733	
19	Phulai ( <i>Acacia modesta</i> )	Hafizabad	437	31	176	1.111	0.025	1.253	0.001	0.437	0.251	0.431	
20	Berseem ( <i>Trifolium alexandrinum</i> )	Hafizabad	412	30	140	1.292	0.255	0.673	0.01	0.084	0.029	0.343	
21	Granda ( <i>Carrisa carandas</i> )	Chakwal	1218	28	168	0.871	0.294	0.84	0.144	0.051	0.166	0.803	

Table 2. Continued...

Sr. No.	Dominant Flora	Locations	Concentration									
			Potassium (K) ppm	Calcium (Ca) ppm	Sodium (Na) ppm	Cobalt (Co) ppm	Chromium (Cr) ppm	Manganese (Mn) ppm	Molybdenum (Mo) ppm	Nickel (Ni) ppm	Lead (Pb) ppm	Selenium (Se) ppm
22	Orange ( <i>Citrus xsinensis</i> )	Chakwal	490	19	186	1.009	0.271	0.597	0.227	0.251	0.232	0.827
23	Phulai ( <i>Acacia modesta</i> )	Chakwal	365	26	162	1.463	0.243	0.64	0.04	0.134	0.782	0.201
24	Black locust ( <i>Robinia pseudoacacia</i> )	Chakwal	288	15	205	1.554	0.66	0.423	0.041	0.007	0.742	0.228
25	Bhaiker ( <i>Justicia adhatoda</i> )	Chakwal	264	22	155	1.363	0.633	0.916	0.508	0.5	0.78	0.713
26	Bhaiker ( <i>Justicia adhatoda</i> )	Chakwal	369	31	118	1.49	0.635	0.594	0.93	0.309	0.906	0.431
27	Clover ( <i>Trifolium repens</i> )	Chakwal	393	27	139	0.92	0.324	1.063	0.919	0.376	0.531	0.489
28	Bhaiker ( <i>Justicia adhatoda</i> )	Chakwal	996	18	209	2.139	0.182	0.061	0.226	0.493	0.71	0.074
29	Phulai ( <i>Acacia modesta</i> )	Chakwal	584	16	130	2.298	0.312	0.844	1.017	0.436	0.758	0.941
30	Bhaiker ( <i>Justicia adhatoda</i> )	Chakwal	308	18	137	1.277	0.084	0.467	0.497	0.411	0.049	0.202
31	Phulai ( <i>Acacia modesta</i> )	Chakwal	347	13	198	2.005	0.35	0.148	0.482	0.572	0.795	0.257

Table 2. Continued...

Sr. No.	Dominant Flora	Locations	Concentration										
			Potassium (K) ppm	Calcium (Ca) ppm	Sodium (Na) ppm	Cobalt (Co) ppm	Chromium (Cr) ppm	Manganese (Mn) ppm	Molybdenum (Mo) ppm	Nickel (Ni) ppm	Lead (Pb) ppm	Selenium (Se) ppm	
32	Orange ( <i>Citrus xsinensis</i> )	Chakwal	384	9	165	2.032	0.283	0.223	0.932	0.646	0.097	1.477	
33	Bari ( <i>Ziziphus mauritiana</i> )	Kotla Haji Shah	360	42	148	1.933	1.807	0.408	1.303	0.889	2.465	0.725	
34	Bari ( <i>Ziziphus mauritiana</i> )	Kotla Haji Shah	430	28	179	2.188	1.016	0.875	1.062	1.104	1.903	0.039	
35	Granda ( <i>Carissa carandas</i> )	Murree	578	23	164	1.687	1.332	0.175	0.257	0.948	0.132	0.119	
36	Bari ( <i>Ziziphus mauritiana</i> )	Talagang	335	24	136	2.179	1.325	0.347	0.498	1.163	2.608	0.024	
37	Orange ( <i>Citrus xsinensis</i> )	Sargodha	596	14	196	1.72	1.595	0.493	0.299	1.463	0.44	0.137	
38	Bari ( <i>Ziziphus mauritiana</i> )	Kasur	301	17	158	2.018	0.756	0.706	0.178	0.331	0.771	0.442	
39	Clover ( <i>Trifolium repens</i> )	Chakwal	173	29	220	2.119	0.254	1.235	0.03	0.962	0.85	0.208	
40	Berseem ( <i>Trifolium alexandrinum</i> )	Kasur	885	22	237	2.266	0.374	1.089	0.028	0.792	1.36	0.049	
41	Orange ( <i>Citrus xsinensis</i> )	Sargodha	326	24	142	1.894	0.668	1.2	0.339	0.772	0.929	1.032	



Table 2. Continued...

Sr. No.	Dominant Flora	Locations	Concentration									
			Potassium (K) ppm	Calcium (Ca) ppm	Sodium (Na) ppm	Cobalt (Co) ppm	Chromium (Cr) ppm	Manganese (Mn) ppm	Molybdenum (Mo) ppm	Nickel (Ni) ppm	Lead (Pb) ppm	Selenium (Se) ppm
42	Bari ( <i>Ziziphus mauritiana</i> )	Talagang	1198	18	233	2.374	1.1	1.269	0.23	0.547	0.823	1.346
43	Orange ( <i>Citrus xsinensis</i> )	Sargodha	636	21	204	0.975	1.099	0.87	1.818	0.248	0.211	0.158
44	Bari ( <i>Ziziphus mauritiana</i> )	Fateh Jang	315	23	228	0.833	1.584	0.582	1.761	0.275	0.023	1.503
45	Bhaiker ( <i>Justicia adhatoda</i> )	Kallar Kahar	358	26	172	0.971	1.191	0.417	1.482	0.234	0.493	1.629
46	Bari ( <i>Ziziphus mauritiana</i> )	Fateh Jang	398	31	197	0.964	0.824	0.512	1.415	0.145	0.537	1.505
47	Granda ( <i>Carissa carandas</i> )	Murree	486	27	372	1.573	0.72	0.804	1.051	0.01	0.182	1.423
48	Phulai ( <i>Acacia modesta</i> )	Hafizabad	509	20	313	1.181	0.352	0.554	1.269	0.255	0.496	0.433
49	Phulai ( <i>Acacia modesta</i> )	Hafizabad	647	35	418	1.318	0.427	0.749	1.159	0.26	0.477	0.623
50	Orange ( <i>Citrus xsinensis</i> )	Shahkot	384	38	367	1.457	0.413	0.263	1.046	0.08	0.367	0.069
51	Berseem ( <i>Trifolium alexandrinum</i> )	Narowal	785	53	240	0.985	0.359	0.633	0.287	0.487	0.409	0.424

Table 2. Continued...

Sr. No.	Dominant Flora	Locations	Concentration										
			Potassium (K) ppm	Calcium (Ca) ppm	Sodium (Na) ppm	Cobalt (Co) ppm	Chromium (Cr) ppm	Manganese (Mn) ppm	Molybdenum (Mo) ppm	Nickel (Ni) ppm	Lead (Pb) ppm	Selenium (Se) ppm	
52	Berseem ( <i>Trifolium alexandrinum</i> )	Narowal	1732	90	337	1.397	0.401	0.812	0.341	0.326	0.423	0.499	
53	Orange ( <i>Citrus xsinensis</i> )	Okara	731	34	278	1.325	0.377	0.202	0.119	1.318	0.318	0.027	
54	Bhaiker ( <i>Justicia adhatoda</i> )	Kallar Kahar	526	78	245	1.589	0.286	0.453	0.362	0.843	0.141	0.715	
55	Bhaiker ( <i>Justicia adhatoda</i> )	Chakwal	763	22	333	1.362	0.593	0.491	0.828	0.795	0.034	0.474	
56	Berseem ( <i>Trifolium alexandrinum</i> )	Narowal	1088	19	164	2.921	0.394	0.919	0.492	0.463	0.439	0.235	
57	Bhaiker ( <i>Justicia adhatoda</i> )	Choa Saidan Shah	703	27	144	1.581	0.697	0.509	0.149	0.976	0.468	1.62	
58	Bari ( <i>Ziziphus mauritiana</i> )	Chakwal	510	24	346	2.174	1.821	0.635	1.008	0.183	1.905	0.497	
59	Bhaiker ( <i>Justicia adhatoda</i> )	Okara	738	16	338	1.093	0.248	0.532	0.139	0.873	0.363	0.306	
60	Sheesham ( <i>Dalbergia sissoo</i> )	Hasilpur	896	55	306	1.185	1.167	0.379	0.253	1.231	1.032	0.248	
61	Phulai ( <i>Acacia modesta</i> )	Okara	604	34	324	2.658	0.095	0.836	1.359	0.549	0.949	0.871	

Table 2. Continued...

Sr. No.	Dominant Flora	Locations	Concentration										
			Potassium (K) ppm	Calcium (Ca) ppm	Sodium (Na) ppm	Cobalt (Co) ppm	Chromium (Cr) ppm	Manganese (Mn) ppm	Molybdenum (Mo) ppm	Nickel (Ni) ppm	Lead (Pb) ppm	Selenium (Se) ppm	
62	Bhaiker ( <i>Justicia adhatoda</i> )	Okara	585	32	273	2.053	0.484	0.614	0.156	0.612	0.781	0.395	
63	Bhaiker ( <i>Justicia adhatoda</i> )	Chakwal	878	30	392	1.711	0.583	0.108	0.987	0.764	0.054	0.559	
64	Bhaiker ( <i>Justicia adhatoda</i> )	Chakwal	812	25	112	1.894	0.632	0.672	0.872	0.801	0.126	0.612	
65	Sarson ( <i>Brassica campestris</i> )	Sargodha	1258	52	216	2.189	0.107	0.856	0.933	0.799	0.864	0.532	
66	Bhaiker ( <i>Justicia adhatoda</i> )	Chakwal	634	14	187	0.912	0.285	0.908	0.779	0.739	0.183	0.464	
67	Bhaiker ( <i>Justicia adhatoda</i> )	Murree	745	17	255	0.784	0.331	0.525	0.687	0.898	0.733	0.982	
68	Sarson ( <i>Brassica campestris</i> )	Okara	638	42	282	2.079	0.296	0.76	0.818	0.712	0.894	0.419	
69	Berseem ( <i>Trifolium alexandrinum</i> )	Narowal	644	38	210	2.651	0.424	0.763	0.423	0.206	0.375	0.689	
70	Berseem ( <i>Trifolium alexandrinum</i> )	Narowal	1166	33	241	1.417	0.651	0.603	0.441	0.313	0.431	0.383	

Table 2. Continued...

Sr. No.	Dominant Flora	Locations	Concentration									
			Potassium (K) ppm	Calcium (Ca) ppm	Sodium (Na) ppm	Cobalt (Co) ppm	Chromium (Cr) ppm	Manganese (Mn) ppm	Molybdenum (Mo) ppm	Nickel (Ni) ppm	Lead (Pb) ppm	Selenium (Se) ppm
71	Berseem ( <i>Trifolium alexandrinum</i> )	Narowal	827	23	309	1.943	0.439	0.442	0.586	0.228	0.573	0.397
72	Berseem ( <i>Trifolium alexandrinum</i> )	Narowal	1502	26	226	2.875	0.465	0.517	0.455	0.096	0.382	0.586
73	Orange ( <i>Citrus xsinensis</i> )	Murree	793	19	195	0.358	0.213	0.279	0.281	0.787	0.457	0.217
74	Berseem ( <i>Trifolium alexandrinum</i> )	Murree	684	24	274	0.592	0.829	0.174	0.254	0.281	0.118	0.085
75	Phulai ( <i>Acacia modesta</i> )	Hafizabad	673	25	188	1.864	0.037	0.731	1.392	0.268	0.396	0.651
76	Orange ( <i>Citrus xsinensis</i> )	Chichawatni	794	28	215	1.749	0.328	0.856	0.498	0.527	0.685	0.046
77	Bhaiiker ( <i>Justicia adhatoda</i> )	Okara	886	27	296	2.121	0.601	0.621	0.451	0.741	0.722	0.311
78	Phulai ( <i>Acacia modesta</i> )	Kallar Kahar	839	29	260	1.207	0.132	0.814	1.167	0.118	0.049	0.732
79	Bhaiiker ( <i>Justicia adhatoda</i> )	Kallar Kahar	565	21	181	0.418	0.573	0.637	0.364	0.952	0.158	0.658
80	Sarson ( <i>Brassica campestris</i> )	Faisalabad	756	30	147	0.769	0.291	0.838	0.836	0.847	0.914	0.591

56, 57, 59, 61, 62, 63, 64, 65, 66, 67, 68, 69, 70, 71, 72, 73, 74, 75, 76, 77, 78, 79 and 80 had Pb as a micro element within the International limit (<1 ppm) (Table 2).

### 3.2.7. Selenium (Se)

The concentration of Selenium (Se) was found maximum (1.968 ppm) than International Honey limit (<1 ppm) in sample No. 9 was collected from Chakwal. Whereas Se in samples No. 9, 11, 13, 14, 16, 17, 32, 41, 42, 44, 45, 46, 47 and 57 had Se in the range of 1-2 ppm (Table 2).

The composition of minerals in fresh 80 honey samples were analyzed as shown in Table 3. In macro elements, the maximum range of potassium, calcium and sodium was found in Berseem (*Trifolium alexandrinum*), Sarson (*Brassica campestris*) and Sheesham (*Dalbergia sissoo*) samples *i.e.*, 810.50, 640.63 and 238.62 ppm, respectively. Whereas lowest concentration of these three elements was present in Orange (*Citrus xsinensis*) ranged 396, 17.50 and 169.88 ppm, respectively.

In micro elements, the optimum concentration of Cobalt is 201 ppm in Berseem while lowest in Clover sample 1.29 ppm. Maximum range of Cr was 1.27 in Beri, its minimum range was present in Sheesham which was 0.22 ppm. Manganese highest value (0.71 ppm) in Sheesham and lowest value (0.37 ppm) in orange. Molybdenum and nikel containing maximal range in Beri and Sarson samples *i.e.*, 0.92 and 0.88 respectively, however its minimal range present in Berseem (0.34 and 0.35 ppm). Samples of orange containing the highest concentration of Lead and Selenium (1.52 and 0.85, respectively) whereas least value (0.39 ppm) of lead was observed in Black locust and minimum selenium concentration (0.38) was found in sarson (Table 3).

The physicochemical parameters of the 80 honey samples were analyzed. The lowest moisture content was present in both Phulai and Sheesham samples which were 15.00%, whereas 31.23% highest moisture contents were measured in Clover. In the present study, variation of color was observed in different honey samples collected from various flora. The value of color variation was observed 19.5, 25.89 and 32.01 mmpfund (white) in Clover, Bhaiker and Black locust, respectively, 34.67 to 50.00 mm pfund (extra light amber) was observed in granda, serson, orange, sheesham, barseem, and phulai samples while bari samples contained 80.00 mm pfund (light amber). pH value of Black locust and Sheesham were neutral *i.e.*, 7.01 and 7.2, respectively, serson samples have basic pH (8.23) while rest of the samples contained acidic pH values. The acidity range vary between 32.29 to 72.02 meq/kg in the observed honey samples in the similar way EC values of given samples was observed between 0.21 to 0.83 ms/cm. The bari samples have highest (0.83%) and sheesham (0.20%) lowest ash contents (Table 4).

## 4. Discussion

The concentration of macro and microelements in bees' bodies varied widely and based on various circumstances, including soil and nectariferous plant varieties, beekeeping procedures, and the physiological and health statuses of

bee workers (Bogdanov, 2006). Naturally, two kinds of minerals are found in honey *i.e.*, macro and micro minerals. In the present study, ten minerals, including three macro minerals (K, Ca and Na) and seven micro minerals (Co, Cr, Mn, Mo, Ni, Pb and Se) were analyzed.

The most abundant elements identified in the present research were K, Ca and Na with concentrations ranging from 166-1732, 7-99 and 107-418 ppm, respectively. And the presence of trace elements (Co, Cr, Mn, Mo, Ni, Pb and Se) in honey samples indicates a nutritional value and potential component for table sweeteners. These three minerals are also discovered in the highest concentrations in honey from the Canary Islands, Spain (Fernández-Torres et al., 2005). (Cantarelli et al., 2008; Pisani et al., 2008; Kiliç Altun et al., 2017; Altunatmaz et al., 2019) found similar results with honey samples from Turkey, Argentina and Italy. According to the results, K was the most prevalent mineral in all local and exotic honey varieties, followed by Na and Ca. (Mesallam and El-Shaarawy, 1987) determined that K and Na were the most abundant elements in the honey. The Dietary Guidelines for Americans 2010 Advisory Committee recognized K as the first significant insufficient nutrient. Its advantages for organic anions are connected and occur in foods such as honey. The values of K vary between 1.18 and 268 ppm. According to our results, greater levels of K in honey samples have already been recorded in Eastern Slovakia (Kováčik et al., 2016). Na is required for appropriate cell activity, plasma volume maintenance, acid-base balance, and nerve impulse transmission (Kiliç Altun et al., 2017). Ca is a necessary vitamin for bone health because it promotes bone calcification (Tuyen et al., 2016).

The trace minerals are also common in honey, and they are present in smaller quantities than major minerals. The trace mineral elements of honey were obtained in order of decreasing Se > Co > Ni > Pb > Cr > Mo > Mn. According to European Union (2002) and Sobhanardakani and Kianpour (2016), the quantities of trace elements Co, Cr, Mn, Mo, Ni, Pb, and Se found in several honey samples in this study are relatively low and fall below acceptable international limits. Although necessary in minute amounts in the body, these trace elements are essential to the normal biological activities of the human body but may be harmful when present in excessive concentrations.

Normally, the most abundant elements in the given honey samples were potassium, calcium, and sodium, ranging between 396.00-810.50, 17.50-640.63 and 169.88-238.62 ppm honey, respectively. The findings of this study revealed that the honey sample was highly rich in macro minerals. (Boussaid et al., 2015) determined the sodium range from 497.54 to 362.55 ppm and 251.34 to 521.22 ppm, almost equal to the current research. All of the fresh honey samples had Ca<sup>+</sup> levels lower than the international range (200-2300 ppm). (Abdulkhalik and Swaileh, 2017) investigated potassium and sodium concentrations ranging from 183.86 ppm to 104.66 ppm, which are lower than the previous study. The findings agreed with those of (Agbagwa et al., 2011) and (Joel, 2014), who found potassium dominance in the honey. On the other hand, microelements such as Cobalt, Chromium, Manganese, Molybdenum, Nickel, Lead and Selenium were found in the normal range. Bari samples had the greatest levels

**Table 3.** Comparison of means  $\pm$  SE of different macro and micro elements on the behalf of different plants resources (n = samples).

Floral sources	Macro Elements				Micro Elements						
	Potassium (ppm) or mg/kg	Calcium (ppm)	Sodium (ppm)	Cobalt (ppm)	Chromium (ppm)	Manganese (ppm)	Molybdenum (ppm)	Nickel (ppm)	Lead (ppm)	Selenium (ppm)	
<b>Bari</b> ( <i>Ziziphus mauritiana</i> ) n=8	480.88 $\pm$ 36.56b	25.875 $\pm$ 36.56b	203.13 $\pm$ 33.46b	1.83e $\pm$ 46.56c	1.2763 $\pm$ 86.56e	0.6637e $\pm$ 76.36cd	0.9291 $\pm$ 76.56c	0.5755 $\pm$ 36.56bc	1.3775 $\pm$ 96.56e	0.7566 $\pm$ 46.56c	
<b>Berseem</b> ( <i>Trifolium alexandrinum</i> ) n=12	810.50 $\pm$ 88.26e	36.500 $\pm$ 68.16c	201.00 $\pm$ 38.26b	201.00a $\pm$ 88.26e	0.5876 $\pm$ 81.26a	0.6912a $\pm$ 84.36c	0.3425 $\pm$ 48.16a	0.3513 $\pm$ 28.26a	0.4415 $\pm$ 58.26ab	0.4850 $\pm$ 38.26ab	
<b>Bhaiker</b> ( <i>Justicia adhatoda</i> ) n=19	675.13 $\pm$ 81.17d	23.875 $\pm$ 31.17b	206.38 $\pm$ 51.27b	131.70c $\pm$ 71.37d	0.4829 $\pm$ 73.17c	0.4466c $\pm$ 51.67ab	0.4350 $\pm$ 51.07ab	0.5213 $\pm$ 31.17b	0.4550 $\pm$ 61.17ab	0.6414 $\pm$ 61.17b	
<b>Black locust</b> ( <i>Robinia pseudoacacia</i> ) n=2	595.13 $\pm$ 123.80c	29.500 $\pm$ 43.70b	212.00 $\pm$ 73.60bc	1.43d $\pm$ 23.80b	0.4850 $\pm$ 68.70d	0.4375d $\pm$ 43.90ab	0.5713 $\pm$ 63.60b	0.6138 $\pm$ 43.80bc	0.3925 $\pm$ 23.80a	0.6803 $\pm$ 53.80b	
<b>Clover</b> ( <i>Trifolium repens</i> ) n=2	693.88 $\pm$ 117.70d	22.125 $\pm$ 26.70b	231.12 $\pm$ 87.70c	1.29c $\pm$ 18.70a	0.5001 $\pm$ 76.50c	0.5225c $\pm$ 117.70b	0.6650 $\pm$ 77.80bc	0.7989 $\pm$ 87.70d	0.4253 $\pm$ 47.70ab	0.7388 $\pm$ 77.70c	
<b>Granda</b> ( <i>Carissa carandas</i> ) n=4	606.38 $\pm$ 96.30d	22.375 $\pm$ 27.30b	190.25 $\pm$ 28.30a	1.32c $\pm$ 21.30ab	0.5663 $\pm$ 96.30c	0.6675c $\pm$ 79.40cd	0.4404 $\pm$ 86.30ab	0.4655 $\pm$ 96.30ab	0.4757 $\pm$ 76.30ab	0.5083 $\pm$ 56.30bc	
<b>Orange</b> ( <i>Citrus xsinensis</i> ) n=15	396.00 $\pm$ 138.98a	17.500 $\pm$ 18.88a	169.88 $\pm$ 22.98a	1.47f $\pm$ 28.98b	0.4528 $\pm$ 59.88f	0.3717f $\pm$ 38.78a	0.4881 $\pm$ 38.98ab	1.0308 $\pm$ 98.98a	1.5424 $\pm$ 108.98b	0.8514 $\pm$ 88.98d	
<b>Phulai</b> ( <i>Acacia modesta</i> ) n=11	738.00 $\pm$ 84.28e	25.000 $\pm$ 34.3b	213.50 $\pm$ 79.28bc	1.33b $\pm$ 24.28ab	0.3985 $\pm$ 53.28b	0.5759b $\pm$ 67.48bc	0.6389 $\pm$ 54.28bc	0.6602 $\pm$ 84.28c	0.5203 $\pm$ 84.28b	0.4635 $\pm$ 64.28ab	
<b>Sheesham</b> ( <i>Dalbergia sissoo</i> ) n=4	520.75 $\pm$ 85.45c	32.625 $\pm$ 55.45c	238.62 $\pm$ 95.45c	1.73d $\pm$ 35.45bc	0.2263 $\pm$ 45.45d	0.7150d $\pm$ 86.55d	0.8360 $\pm$ 85.55c	0.3598 $\pm$ 45.45a	0.6130 $\pm$ 95.45c	0.5510 $\pm$ 75.45bc	

Mean sharing similar letter in a column are statically non – significant (P&gt;0.05)

Table 3. Continued...

Floral sources	Macro Elements				Micro Elements					
	Potassium (ppm) or mg/kg	Calcium (ppm)	Sodium (ppm)	Cobalt (ppm)	Chromium (ppm)	Manganese (ppm)	Molybdenum (ppm)	Nickel (ppm)	Lead (ppm)	Selenium (ppm)
Sarson ( <i>Brassica campestris</i> ) n=3	640.63 ± 82.75d	640.63 ± 132.75cd	200.50 ± 32.75c	1.36c ± 26.75ab	0.4538 ± 60.95c	0.5576c ± 62.25b	0.5903 ± 81.57bc	0.8816 ± 89.75e	0.7229 ± 82.75d	0.3801 ± 52.75a

Mean sharing similar letter in a column are statically non – significant (P>0.05)

Table 4. Physicochemical parameters of different honey samples analyzed from various flora (mean ± SD).

Parameters	Bari <i>Zizipus jojoba</i> n=8	Barseem ( <i>Trifolium repens</i> ) n=12	Bhaiker ( <i>Jusdticia adhatoda</i> ) n=19	Black locust ( <i>Robinia pseudoacacia</i> ) n=2	Clover ( <i>Trifolium repens</i> ) n=2	Granda ( <i>Carissa caranads</i> ) n=4	Orange ( <i>Citrus xsinensis</i> ) n=15	Phulai ( <i>Acacia modesta</i> ) n=11	Sheesham ( <i>Dalbergia sisso</i> ) n=3	Serson ( <i>Brassica compestris</i> ) n=3	P value	Codex*
Moisture (%)	15.8 ± 2.07	18.45 ± 2.78	15.09 ± 1.07	22.07 ± 4.60	31.23 ± 6.07	20.13 ± 2.12	15.34 ± 2.78	15.00 ± 0.01	15.00 ± 0.78	15.78 ± 3.09	0.56	≤21%
Color (mm Pfund)	80 ± 22.12	45.41 ± 9.15	25.89 ± 11.12	32.01 ± 21.22	19.5 ± 5.89	34.67 ± 3.31	45 ± 5.79	50.00 ± 10.99	45 ± 3.25	42.1 ± 25.39	0.00	—
pH	5.98 ± 0.52	6.78 ± 0.76	4.75s ± 2.45	7.01 ± 3.09	5.21 ± 1.78	4.78 ± 3.14	6.43 ± 0.89	5.67 ± 1.05	7.2 ± 1.02	8.23 ± 0.78	0.00	—
Acidity (meq/kg)	39.2 ± 5.89	38.55 ± 5.87	32.66 ± 7.08	72.02 ± 5.11	43.34 ± 2.78	45.32 ± 1.02	56.02 ± 4.92	32.29 ± 2.02	39.03 ± 5.09	35.02 ± 4.76	0.05	≤40 meq/kg
EC (ms/cm)	0.51 ± 0.01	0.5 ± 0.03	0.85 ± 0.16	0.45 ± 0.02	0.21 ± 0.03	0.41 ± 0.07	0.35 ± 0.01	0.24 ± 0.94	0.21 ± 0.02	0.21 ± 0.06	0.06	≤0.7 ms/cm
Ash (%)	0.83 ± 0.09	0.79 ± 0.32	0.71 ± 0.52	0.29 ± 0.81	0.56 ± 0.71	0.51 ± 0.98	0.62 ± 0.20	0.29 ± 0.05	0.20 ± 0.08	0.21 ± 0.02	0.58	≤0.6 g/kg

of chromium and lead. Cadmium was not found in any honey samples tested. (Mena et al., 1996) determined that cadmium and lead are non-essential components in plant nutrition and one of the most harmful chemicals that accumulate in biological systems. According to mineral analyses, fresh honey is high in nutritional components and low in harmful metals.

The range of moisture contents varied in the given honey sample between 15.00 to 31.23% (Table 3). It depends on various factors like the floral origin of honey, way of storage, and manufacturing techniques. Kumar et al. (2018) was determined 18.37 to 25% moisture contents, which is comparable to the present findings. The moisture contents in honey samples are based on various circumstances like environmental factors, degree of maturity and producing season (Acquarone et al., 2007). For classifying different flora of honey, color determination is an effective technique. The color of honey in the present samples varies 19.5 to 80 mm pfund of clover and bari samples, respectively. (Boussaid et al., 2015; Khalafi et al., 2016; Aazza et al., 2018) determined the range of color 71.72 mm pfund, 36.46-50.73 mm pfund and 19.01-46.7 mm pfund, which is very similar to the present research. pH is an essential parameter while extracting and storing honey. Terrab et al. (2002) determined that pH impacts the stability, texture and shelf life of honey. In the present study, all samples showed acidic pH except black locust, sheesham and serson samples, which showed neutral and slightly basic pH values. Ouchemoukh et al. (2007) found the pH value was comparable to the earlier reported studies in Spanish, Algerian and Portugal honeys, which ranged between pH 3.50 to 6.58.

The range of acidity varied between 32.29- 72.02 meq/kg in given honey samples. (Ajouluni and Sujirapinyokul, 2010) found that a high level of acidity causes yeast fermentation of honey sugar. High free acidity levels have been observed to indicate yeast fermentation of honey sugar. Glucose and fructose are generally converted into carbon dioxide and alcohol during fermentation. In the presence of oxygen, alcohol is more hydrolyzed and transformed into acetic acid. De La Fuente et al. (2011) observed that the EC value of honey is directly linked to the mineral's concentration or total ash, organic acid, salts, and protein. This parameter differs widely and depends upon the floral origin of the honey samples. After observing 80 samples of honey collected from different floral species, the EC values ranged between 0.21 to 0.83 ms/cm. Ash content is a parameter used to find out the floral origin (Boussaid et al., 2015). Parviz et al. (2015) and Sousa et al. (2016) found ash contents showed values 0.03 to 0.53 and 0.5- 0.8, respectively, which is very close to the present study.

## 5. Conclusion

The present study showed the findings concerned honey and their level of micro and macro elements. Some of the variation in the honey sample was seen deviated from the international standard. It has been noted from the honey freshness and purity that the flora affects the micro and macro elements level in the honey composition.

The presence of trace amounts of heavy metals such as Pb, Cr, and Mo in all fresh honey samples indicated a clean environment, while the presence of other critical metals indicated the high nutritional value of Pakistani honey. The values of physicochemical parameters of given honey samples were very close to the suggested limit of international standards.

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### Supplementary Material

Supplementary material accompanies this paper.

#### **Table S1: Operational Conditions of Atomic Absorption Spectrum**

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