Heavy metal in honey bees, honey and pollen produced in different locations of Konya province in Turkey

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Abstract

We investigated the levels of heavy metals in honey bee, honey and pollen samples obtained from different locations of Konya City in Turkey. Five honey-bee colonies were placed in eight different locations, four of them around the city center and four in rural areas, in the province of Konya City in Turkey. Heavy metal (Cd, Cr, Cu, Fe, Mn, Ni, Pb, and Zn) levels were determined in honey-bee, honey, and pollen samples taken from these colonies, with comparison between samples from urban and rural areas. The values of Cd and Pb in honey samples and those of Cd in pollen samples did not differ significantly among the locations. All heavy metal values of honey bee samples were lower in rural areas than in urban areas ($P < 0.05$). Significant statistical differences were determined for Cr, Cu, Fe, Mn, Ni, and Zn values of honey and pollen samples among the locations ($P < 0.05$). Heavy metal values of honey and pollen samples obtained from different locations were in agreement with the International Food Standard values.

Introduction

Beekeeping is sometimes practiced in places close to or within residential and industrial areas. Apiaries that are not suitable for bee colonies and mistakes made in production deteriorate the natural characteristics and quality of bee products.

Environmental problems that arise with population growth, urbanization, industrialization, and changing consumption habits are increasingly common. For example, heavy metals released into the air in the form of dust and ash from various sources accumulate on plants (Taha et al. 2017) and cause the pollution of soil and water resources by precipitation or by sedimentation. In addition, heavy metals in wastewater, chemicals used in agriculture, and fertilizers can also affect animals and humans via accumulation on the food chain (Duruibe et al. 2007; Squadrone et al. 2020; Türközü and Şanlıer 2014; Yılmaz 1996).

Heavy metals enter the bodies of animals and humans via ingestion (food and water), respiration, and the skin; they can be classified as essential and non-essential (Özbolat and Abdullah 2016). For example, some elements such as copper, zinc, iron, manganese, and selenium are essential for body development and proper functioning. On the other hand, non-essential elements, such as lead, cadmium, and mercury can cause various disorders by accumulating in the body and affecting the biological structure, even at low concentrations (Tuzen et al. 2007; Uluozlu et al. 2007).

To determine the environmental pollution of a region, living organisms (bioindicators or biomonitors) that show different sensitivities to various pollutants can be used (Yılmaz 1996). In this sense, honey-bees and bee products can contain residues of pollutants, making them important indicators of environmental pollution (Ahmida et al. 2012; Bogdanov 2008; Porrini et al. 2003; Taha et al. 2017; Zhelyazkova 2012).

Honey-bees can easily live in different environmental conditions, can be kept easily, have a short life cycle and a high reproductive rate, and as a colony, they can be transported to any place. They move around the apiary within an area of approximately 7 km^2 in search of food, and samples for analysis can be taken during flight activity (Conti and Botrè 2001; Leita et al. 1996; Perugini et al. 2011).

Bee and bee products can be contaminated with pollutants from different sources, and the contamination may differ according to the environmental conditions of the beekeeping area (Costa-Silva et al. 2011; Hennessy et al. 2010; Pohl et al. 2011). Demirezen and Aksoy (2005) and Bogdanov (2008) reported that heavy metal accumulation is higher in bee products obtained from industrial regions and areas with heavy vehicle traffic, especially those close to large settlements and garbage incinerators.

In this context, this study compared the heavy metal contents of honey bees, honey, and pollen obtained from places close to residential and industrial areas with those obtained from rural areas and determined whether honey and pollen taken from these areas pose a problem in terms of food safety.

Materials And Methods

Sampling Sites (Locations)

The study was carried out in Konya region, Turkey, in 2018. Five honey bee colonies (40 in total) were placed in eight different multifloral locations, four of which were around the city center (L1 to L4) and four in the rural area (L5 to L8) (Fig. 1).

The locations had the following characteristics:

L1: 38°02'05" N, 32°30'10" E, 1,180 m. On the northern side of the city and in the prevailing wind direction of the city, 1,210 m away from and west of the highway. There is very little agricultural activity in the vicinity, and there are no industrial facilities.

L2: 37°55'12" N, 32°26'10" E, 1,140 m. On the northwestern side of the city and in the prevailing wind direction of the city, 4,300 m away from and north of the highway. There is very little agricultural activity in the vicinity, and there are no industrial facilities.

L3: 37°51'07" N, 32°33'33" E, 1,010 m. Southeast of the city, southwest of the industrial regions; the prevailing wind direction is from the industrial regions. This site is 1,800 m away from and south of one highway and 1,300 m away from and east of another highway. Agricultural activities are carried out in the surrounding area.

L4: 37°49'12" N, 32°28'45" E, 1,027 m. On the southern side of the city; the prevailing wind direction is from the city side. It is 1,000 m away from and south of one highway and 3,400 m away from and west of another highway. Although not nearby, there are industrial facilities in the prevailing wind direction, and agricultural activities are carried out around it.

L5: 37°28'16" N, 31°48'56" E, 1,131 m. This site is 4,600 m away from and northwest of the highway. There is only one aluminum-processing plant at a distance of 3,750 m in the southeast, and there is little agricultural activity in its vicinity.

L6: 37°42'05" N, 33°31'20" E, 1,003 m. This site is 1,400 m away from and south of the highway. There are no industrial facilities in the vicinity; agricultural activities are carried out sparsely.

L7: 38°05'07" N, 32°16'45" E, 1,630 m. In the vicinity of this site, there is very little agricultural activity, and there are no highways, industrial facilities, etc. nearby.

L8: 36°58'38" N, 32°22'51" E, 1,775 m. In the vicinity of this site, there is very little agricultural activity, and there are no highways, industrial facilities, etc. nearby.

The prevailing wind direction in Konya is north. In the months of May to August 2018, which were the sampling months, the area experienced 67% northerly winds (39% north, 13% north east, and 15% northwest).

Beehive and Colony Characteristics

We used Langstroth type beehives with a plastic bottom and pollen trap. The colonies were arranged with a newly raised honeycomb, eight frame hives, and 1-year-old queen bee colonies were used, without additional feeding.

Collection and Conservation of Samples

Pollen: Pollen was collected three times every 15 days in May and June and dried in the dark. Subsequently, 25 g (total 75 g) pollen collected and dried from each colony at once was taken, mixed, placed into glass jars, and stored at -18°C until analysis.

Honey: The honey of each colony was harvested separately between 15 July and 15 August without using a smoker. Approximately 500 g of honey from each colony was placed in glass jars, and the honeys were kept at room temperature and in the dark until analysis.

Honey bee: After August 15, the entrance hole of each colony was closed before noon (around 09:00 – 10:00), and 30 worker bees returning from the field were caught at the hive entrance using plastic gloves. Samples were placed in glass jars and stored at -18°C until analysis.

Preparation of samples and heavy metal analysis

Dried pollen was ground to obtain a homogeneous sample (Kacar and İnal 2008). Approximately 2 g of ground pollen samples and bee samples kept at -18°C were taken and dried in an oven at 70°C until constant weight before being used in the analysis.

In the analysis of heavy metals, 0.2 g of honey, pollen and bee (whole bee) samples obtained from all locations were weighed into heat-resistant Teflon containers. Subsequently, 5 ml of concentrated HNO_3 and 2 ml of H_2O_2 (30% w/v) were added to the weighed samples, and the samples were thawed in a microwave device under high temperature (210°C) and pressure (200 PSI). To ensure the reliability of the analysis, one control (blank) and one certified reference sample (Peach Leaves, NIST, SRM 1547) were added into the 40-cell microwave set. The volumes of the thawed samples were made up to 20 ml with deionized water, filtered with blue-banded filter paper, and stored in the refrigerator at +4°C until readings were taken.

The heavy metal contents of the samples (total Pb, Cd, Cr, Zn, Cu, Ni, Mn, and Fe) were determined using an ICP-AES (Inductively Coupled Plasma Atomic Emission Spectrometry, Varian-Vista Model, Axial) (USDA 2004). Elemental amounts of the samples are given in µg/kg for Cd and Cr and in mg/kg for the other elements.

Statistical Analysis

Heavy metal amounts were determined in five honey bee, honey, and pollen samples taken from each of the eight different locations. The study was carried out in a randomized plot design. The data were subjected to one-way analysis of variance, and Tukey´s test was used to determine the differences among groups. Data are presented as mean and standard error.

Results And Discussion

Heavy metal values in honey samples

Heavy metal (Cd, Cr, Cu, Fe, Mn, Ni, Pb, and Zn) mean values and standard errors of honey samples are shown in Table 1. In honey samples, significant statistical differences were found among the locations in terms of Cr, Cu, Fe, Mn, Ni and Zn, except Cd and Pb, ($P<0.05$).

The Cd value of honey samples ranged from 4.301–6.898 µg/kg, and the difference among locations was statistically insignificant. Bosancic et al. (2020) stated that no statistically significant differences were found between conventional and organic honey production systems. On the other hand, Arslan and Arıkan (2013) found the highest amount of Cd in honey samples taken from stations close to the highway. Adugna et al. (2020) found Cd contents of honey samples from different regions in the range of 17–35 μg/kg,which was higher compared with our results.

The maximum Cu content of honey samples was 0.950 mg/kg at L2, whereas the minimum values was 0.395 mg/kg at (L7) ($P<0.05$). Demirezen and Aksoy (2005) found that the Cu content of honey samples taken from sites close to residential areas was higher than those from rural areas (P�0.01). On the other hand, Silici et al. (2016) stated that there was no statistical difference among the Cu values of honey samples taken from different distances to a thermal power plant.

The Cr, Fe, Mn, Ni, and Zn values of honey samples were highest around the city (L3, 67.01 μg/kg; L4, 67.01 μg/kg; L1, 1,270 mg/kg; L3, 0.216 mg/kg; L4, 1.635 mg/kg) and lowest in the rural sites (L8, 27.70 μg/kg; L6, 6.266 mg/kg; L6, 0.520 mg/kg; L8, 0.106 mg/kg; L6, 1.039 mg/kg).

Leblebici (2006) reported that the Cr content of honey samples taken from sites close to residential areas was higher than that from rural areas ($P<0.05$). Gurel et al. (1998) found significant differences among honey samples obtained from different locations in terms of Fe concentration ($P<0.01$). Leblebici (2006) found the Fe content of honey samples taken from locations close to the city center was higher than that of locations far from the city center ($P<0.05$). Arslan and Arıkan (2013) did not detect a significant difference among the Mn values of honey samples obtained from colonies placed at different distances from the highway. Taha et al. (2017) found that the Ni content in honey samples taken from colonies placed at different distances to the cement factory was higher in samples close to the factory ($P<0.05$). Demirezen and Aksoy (2005) stated that the Ni content of honey samples taken from sites close to residential areas was higher than those from rural areas (P�0.01). Taha et al. (2017) found the Zn content of honey samples taken from colonies placed at different distances from a cement factory was higher in samples close to the factory ($P<0.05$). The Zn values (1.039-1.635 mg/kg) obtained in our study were lower than the value (4.814 mg kg) determined by Aliu et al. (2020) in honey.

The Pb content of honey samples ranged between 0.073 and 0.118 mg/kg, and the differences among the locations were statistically insignificant. On the other hand, Roman (2010) determined that the Pb content of honey samples obtained from settlements was higher than the values obtained from agriculture and forest areas ($P<0.05$). The Pb value (0.073-0.118 mg/kg) obtained in our study was similar to the value (0.02-0.098 mg/kg) determined by Purcarea et al. (2017) in Polish honeys.

In our study, the Cd and Pb values of honey samples were within the International Food Standard values (Alimentarius 2015).

Heavy metal values in pollen samples

The mean values and standard errors of Cd, Cr, Cu, Fe, Mn, Ni, Pb, and Zn in pollen samples are given in Table 2. In pollen samples, the differences among the locations were statistically significant in terms of Cr, Cu, Fe, Mn, Ni, Pb, and Zn, except Cd, ($P<0.05$).

The Cd contents of the pollen samples ranged between 6.535 and 11.304 μg/kg, and the difference among the locations was statistically insignificant. Similarly, Arslan and Arıkan (2013) stated that there was no significant difference among the Cd values of pollen samples taken from colonies placed at different distances from the highway.

The Cd value (6.535 - 11.304 μg/kg) obtained in our study was lower than the value (26.1- 92.0 μg/kg) determined by Formicki et al. (2013) in Poland pollen.

The Cr, Cu, Fe, Mn, Ni, and Pb values of pollen samples were highest at sites around the city (L3, 79.36 μg/kg; L4, 7.275 mg/kg; L4, 96.95 mg/kg; L1, 19.149 mg/kg; L4, 0.384 mg/kg; L4, 0.180 mg/kg) and lowest in the rural sites (L8, 46.10 μg/kg; L6, 3.915 mg/kg; L7, 61.40 mg/kg; L6, 5.356 mg/kg; L7, 0.218 mg/kg; L8, 0.059 mg/kg).

Conti and Botrè (2001) found higher Cr contents in pollen samples obtained from inner-city locations compared to urban locations ($P<0.01$). Taha et al. (2017) found that the Cu content of pollen samples taken from colonies placed at different distances from a cement factory was higher in samples close to the factory ($P<0.05$). Taha (2015) found significant statistical differences among pollen obtained from different plants in terms of Fe content ($P<0.05$). Arslan and Arıkan (2013) did not find a significant statistical difference among the Fe values of pollen samples obtained from colonies placed at different distances from the highway. Fakhimzadeh and Lodenius (2000) found no significant statistical difference among industrial, urban, and rural areas in terms of Mn values in pollen samples. Taha et al. (2017) found that the Ni content in the pollen samples taken from the colonies placed at different distances to a cement factory was higher in the samples close to the factory ($P<0.05$). Conti and Botrè (2001) found higher Pb contents of pollen samples taken from the colonies in the city center compared to those from colonies around the city ($P<0.01$).

The Zn content of the pollen samples was highest in the urban location (L3), with 20.27 mg/kg and lowest in the rural location (L7), with 10.25 mg/kg ($P<0.05$). On the other hand, Arslan and Arıkan (2013) did not detect significant statistical differences among the Zn values of pollen samples obtained from colonies placed at different distances from the highway.

The Zn value (10.25-20.27 mg/kg) obtained in our study was lower than the value (75.2- 159.3 μg/g) determined by Formicki et al. (2013) from Poland pollen. Altunatmaz et al. (2017) stated that mineral levels are related to plant type rather than to the soil and geographical situation.

Heavy metal results in honey bee samples

Heavy metals (Cd, Cr, Cu, Fe, Mn, Ni, Pb and Zn; mean values and standard errors) of honey bee samples are summarized in Table 3.

The Cd content of honey bee samples was highest in the site around the city (L4), with 20.78 μg/kg, and lowest at the rural site (L8), with 9.52 μg/kg ($P<0.05$). Similar results were found by Conti and Botrè (2001), where the Cd content of bee samples obtained from the inner-city location was higher than that from the surrounding locations ($P<0.01$). Similarly, Fakhimzadeh and Lodenius (2000) found a higher Cd content of bee samples taken from urban and industrial areas compared to samples from rural areas ($P<0.05$). The Cd values (9.52-20.78 μg/kg) obtained in our study were lower than the range (0.03-0.30 mg/kg) determined by Goretti et al. (2020).

The Cr content of honey bee samples was highest, at 99.24 μg/kg, in the urban site (L3) and lowest, at 58.24 μg/kg, in the rural site (L7) ($P<0.05$). Gutiérrez et al. (2015) found the highest Cr contents in honey bee samples taken from different locations, including urban, industrial, agricultural, and forested areas (P�0.01).

In this study, the Cu content of honey bee samples was highest in the site around the city (L3), with 17.18 mg/kg, and lowest at the rural site (L6), with 12.18 mg/kg ($P<0.05$). Roman (2010) found that the Cu contents of honey bee samples obtained from residential areas were higher than those obtained from agricultural and forest areas ($P<0.05$). Silici et al. (2016), on the other hand, did not detect a statistical difference among the Cu values of honey bee samples taken at different distances to a thermal power plant.

Te Fe contents of honey bee samples were highest at the site around the city (L3), with 101.81 mg/kg, and lowest at the rural site (L6), with 82.46 mg/kg ($P<0.05$). Nisbet et al. (2013) reported that the Fe content of honey bee samples taken from locations with different environmental and flora characteristics varied from region to region ($P<0.05$). Taha et al. (2017) found that the Fe contents in honey bee samples taken from colonies placed at different distances to a cement factory were higher in samples close to the factory ($P<0.05$).

The Mn contents of honey bee samples were highest at the site around the city (L1), with 35.55 mg/kg, and lowest at the rural location (L6), with 15.63 mg/kg ($P<0.05$). On the other hand, Silici et al. (2016) did not detect a statistical difference among the Mn values of honey bee samples taken at different distances to a thermal power plant.

The Ni values of honey bee samples were highest at the site around the city (L3), with 0.432 mg/kg, and the lowest at the rural location (L7), with 0.201 mg/kg ($P<0.05$). Nisbet et al. (2013) found a statistical difference among the Ni contents of honey bee samples obtained from locations with different environmental and floral characteristics ($P<0.05$).

The Pb contents of honey bee samples were highest at the site around the city (L3), with 0.358 mg/kg, and lowest at the rural site (L8), with 0.192 mg/kg ($P<0.05$). Roman (2010) found higher Pb levels of honey bee samples in residential areas compared to agricultural and forest areas ($P<0.05$). The Pb values (0.192-0.358 mg/kg) obtained in our study were lower than those (4-27 μg/g) determined by Leita et al. (1996). On the other hand, our values were similar to those (0.14-0.52 mg/kg) determined by Perugini et al. (2011).

The Zn contents of honey bee samples were highest at the site around the city (L4), with 44.58 mg/kg, and lowest at the rural location (L8), with 29.93 mg/kg ($P<0.05$). Nisbet et al. (2013) reported that the Zn values of honey bee samples taken from locations with different environmental and floral characteristics varied from region to region ($P<0.05$), In a similar study, Taha et al. (2017), analyzing honey bee samples from colonies placed at different distances to a cement factory, found higher Zn values in the samples closer to the factory ($P<0.05$).

Conclusions

There were no statistical differences among the locations in terms of Cd and Pb in honey samples. However, the Cr, Cu, Fe, Mn, Ni, and Zn values were lower in samples from rural areas.

Regarding the pollen samples, we observed significant differences among the locations in terms of Cr, Cu, Fe, Mn, Ni, Pb, and Zn, with values being lower in rural than in urban areas. The Cd Cr, Cu, Fe, Mn, Ni, Pb, and Zn values of honey bee samples were also lower in rural than in urban areas.

The Cd and Pb values of honey and pollen samples were in agreement with the International Food Standard values (Alimentarius, 2015). To obtain healthy bees and bee products, it is recommended that the colonies are placed away from residential areas, industrial areas, highways, polluted water sources, and areas where bee products are likely to be subjected to contamination.

Declarations

Ethical Approval

Not applicable

Consent to Participate

Not applicable

Consent to Publish

Not applicable

Author Contributions

Study conception, study design, material preparation, data collection, analysis and writing were performed by Huseyin Bayir.

Study conception, study design, analysis, writing and editting were performed by Ali Aygun.

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Competing Interests

The authors have no competing interests to declare that are relevant to the content of this article

Availability of data and materials

The datasets used and/or analyzed during the present study are available from the corresponding author on reasonable request.

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Tables

Table 1 Heavy metalcontents of honey samples

Locations	Cd (µg/kg)	Cr (µg/kg)	Cu (mg/kg)	Fe (mg/kg)	Mn (mg/kg)	Ni (mg/kg)	Pb (mg/kg)	Zn (mg/kg)
L1	6.509±0.642	43.63±3.634^{bc}	0.676±0.073^{abc}	6.880±0.711^{b}	1.270±0.116^{a}	0.185±0.010^{abc}	0.102±0.007	1.289±0.071^{ab}
L2	5.849±0.853	42.77±1.679^{bc}	0.950±0.073^{a}	6.753±0.580^{b}	0.712±0.030^{c}	0.203±0.012^{ab}	0.115±0.007	1.378±0.069^{ab}
L3	6.625±0.701	67.01±6.194^{a}	0.612±0.037^{bc}	10.000±1.076^{ab}	0.784±0.050^{bc}	0.216±0.016^{a}	0.118±0.010	1.606±0.114^{a}
L4	6.898±0.447	60.40±4.943^{ab}	0.821±0.050^{ab}	14.500±1.577^{a}	1.152±0.110^{ab}	0.210±0.021^{ab}	0.118±0.014	1.635±0.116^{a}
L5	5.803±0.574	39.13±1.864^{c}	0.541±0.054^{bc}	7.915±0.922^{b}	0.694±0.058^{c}	0.137±0.015^{bcd}	0.079±0.010	1.159±0.054^{b}
L6	4.861±0.645	42.57±4.007^{bc}	0.446±0.034^{c}	6.266±0.514^{b}	0.520±0.058^{c}	0.137±0.014^{bcd}	0.092±0.007	1.039±0.070^{b}
L7	4.301±0.682	36.85±3.217^{c}	0.395±0.031^{c}	6.744±0.656^{b}	0.564±0.041^{c}	0.124±0.010^{cd}	0.079±0.005	1.148±0.075^{b}
L8	4.816±0.339	27.70±2.187^{c}	0.533±0.055^{bc}	6.326±0.819^{b}	0.710±0.071^{c}	0.106±0.011^{d}	0.073±0.009	1.158±0.025^{b}
P value	0.059	0.000	0.000	0.000	0.000	0.000	0.001	0.000

$^{a-d}$ Different letters in the same column indicate significant differences (P<0.05).

Table 2 Heavy metalcontents of pollen samples

Locations	Cd (μg/kg)	Cr (μg/kg)	Cu (mg/kg)	Fe (mg/kg)	Mn (mg/kg)	Ni (mg/kg)	Pb (mg/kg)	Zn (mg/kg)
L1	10.426±0.981	69.71±3.475^{ab}	5.600±0.378^{abc}	73.18±5.362^{ab}	19.149±0.424^{a}	0.338±0.017^{ab}	0.108±0.004^{cd}	15.10±0.757^{b}
L2	9.848±1.514	75.51±5.626^{ab}	5.685±0.223^{ab}	80.99±6.902^{ab}	10.153±0.623^{c}	0.321±0.030^{ab}	0.123±0.010^{bc}	18.75±0.760^{a}
L3	11.304±1.331	79.36±6.060^{a}	6.355±0.288^{ab}	95.32±8.379^{ab}	18.603±0.846^{a}	0.370±0.036^{ab}	0.171±0.012^{ab}	20.27±0.539^{a}
L4	10.567±0.712	75.76±8.980^{ab}	7.275±0.473^{a}	96.95±4.494^{a}	13.312±0.494^{b}	0.384±0.032^{a}	0.180±0.009^{a}	19.52±0.376^{a}
L5	9.343±0.694	64.96±4.808^{ab}	5.430±0.298^{bc}	89.93±6.951^{ab}	9.789±0.716^{c}	0.247±0.027^{ab}	0.097±0.012^{cd}	17.89±0.388^{ab}
L6	9.328±1.127	58.08±3.582^{ab}	3.915±0.252^{c}	72.70±5.600^{ab}	5.356±0.299^{d}	0.284±0.037^{ab}	0.101±0.009^{cd}	11.59±0.338^{c}
L7	6.535±0.771	56.47±7.630^{ab}	5.260±0.318^{bc}	61.40±5.642^{b}	5.746±0.454^{d}	0.218±0.018^{b}	0.083±0.008^{cd}	10.25±0.197^{c}
L8	8.983±1.023	46.10±3.855^{b}	5.405±0.227^{bc}	71.45±6.923^{ab}	7.137±0.424^{cd}	0.280±0.035^{ab}	0.059±0.005^{d}	10.48±0.788^{c}
P value	0.111	0.003	0.000	0.003	0.000	0.000	0.000	0.000

$^{a\text{-}d}$ Different letters in the same column indicate significant differences (P<0.05).

Table 3 Heavy metalcontents of honey bee samples

Locations	Cd (μg/kg)	Cr (μg/kg)	Cu (mg/kg)	Fe (mg/kg)	Mn (mg/kg)	Ni (mg/kg)	Pb (mg/kg)	Zn (mg/kg)
L1	17.48±0.744^{ab}	66.30±2.138^{b}	13.42±1.059^{ab}	91.59±1.579^{ab}	35.55±1.759^{a}	0.362±0.022^{ab}	0.320±0.023^{ab}	42.01±2.279^{a}
L2	18.50±1.067^{a}	69.79±5.443^{b}	14.77±1.067^{ab}	100.39±3.757^{a}	24.31±1.492^{bc}	0.372±0.021^{ab}	0.330±0.021^{ab}	35.25±1.509^{ab}
L3	19.20±1.058^{a}	99.24±3.247^{a}	17.18±0.911^{a}	101.81±2.102^{a}	28.08±1.429^{ab}	0.432±0.021^{a}	0.358±0.019^{a}	43.83±2.529^{a}
L4	20.78±0.916^{a}	98.54±3.613^{a}	14.90±1.128^{ab}	99.72±3.318^{a}	34.71±2.140^{a}	0.417±0.021^{ab}	0.333±0.019^{ab}	44.58±2.998^{a}
L5	12.98±0.623^{bc}	68.42±3.421^{b}	13.93±0.795^{ab}	96.34±3.287^{ab}	25.67±1.257^{b}	0.315±0.017^{bc}	0.315±0.020^{ab}	33.68±1.433^{ab}
L6	13.25±0.706^{bc}	64.20±3.452^{b}	12.18±1.132^{b}	82.46±4.651^{b}	15.63±1.182^{c}	0.343±0.022^{abc}	0.251±0.016^{bc}	35.12±2.458^{ab}
L7	12.41±0.786^{c}	58.24±3.511^{b}	13.16±0.710^{ab}	85.43±2.737^{ab}	22.20±1.699^{bc}	0.201±0.017^{d}	0.231±0.016^{bc}	32.73±1.957^{ab}
L8	9.52±0.623^{c}	58.69±4.119^{b}	13.01±1.181^{ab}	85.31±1.494^{ab}	24.02±1.653^{bc}	0.238±0.017^{cd}	0.192±0.020^{c}	29.93±1.767^{b}
P value	0.000	0.000	0.047	0.000	0.000	0.000	0.000	0.000

$^{a\text{-}d}$ Different letters in the same column indicate significant differences (P<0.05).

Figures

Figure 1

Locations of the honey bee colonies (sampling sites).