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Diversity and botanical characterization of polyfloral honey from Bosnia and Herzegovina

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Key words: Melisopalynology, pollen, Shannon-Weaver, Shannon uniformity index.

Ključne besede: melisopalinologija, pelod, Shannon-Weaver, Shannonov indeks enotnosti.

Abstract

As part of the research, 50 polyfloral honey samples from Bosnia and Herzegovina (B&H) were collected and analyzed. The study aimed to determine the botanical specificity and degree of diversity of polyfloral honey originating from B & H. Melisopalynological preparations were prepared and interpreted by the Regulation on Methods for the Control of Honey and Other Bee Products of Bosnia and Herzegovina and methods proposed by the ICBB. Melissopalynological analysis revealed 15,000 pollen grains from 50 polyfloral honey samples and from 24 plant families. The most prominent number of pollen grains was from Fabaceae, Rosaceae and Asteraceae. Through the analysis of micromorphological characteristics, 51 different pollen types were identified. The Shannon-Weaver (H) index value of the analyzed samples were from 1.65 to 2.72, while the Shannon uniformity index (J') was from 0.7 to 1. Each analyzed palynological profile represents a unique combination of pollen from honey plants and reflects the floristic diversity of the apiflora in B&H.

Izvleček

V raziskavi smo analizirali 50 cvetličnih medov iz Bosne in Hercegovine (B & H). Namen študije je ugotoviti posebnosti in raznolikost cvetličnih medov. Melisopalinološke preparate smo naredili v skladu z Uredbo o metodah za nadzor medu in drugih čebeljih proizvodov Bosne in Hercegovine in metodami, ki jih predlaga ICBB. Z melisopalinološko metodo smo ugotovili 15.000 pelodnih zrn iz 50 vzorcev ki pripadajo 24 družinam. Najpogostejša so pelodna zrna iz družin Fabaceae, Rosaceae in Asteraceae. V analizi mikromorfoloških značilnosti smo ugotovili 51 različnih tipov pelodnih zrn. Vrednost Shannon-Weaverjevega (H) indeksa je bila med 1,65 in 2,72, Shannonovega indeksa enotnosti pa med 0,7 in 1. Vsak palinološki vzorec predstavlja posebno kombinacijo peloda medovitih rastlin in odraža floristično raznolikost apiflore Bosne in Hercegovine.

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Introduction

Honey plants include all plant species whose natural products (pollen, nectar and honeydew) are the main source of food for honey bees (Dujmović Prugar & Hulina, 2007; Ljevnaić-Mašić et al., 2019; Khan et al., 2024). The distribution of honey plants and the chemical properties of their products are the results of a complex interaction of ecological, geographic, and anthropogenic factors in the corresponding area (Chiş & Purcarea, 2011; Tomczyk et al., 2019; Ivanović et al., 2021). The botanical origin of honey is defined by the floristic composition of the honey pasture, the number of honey plants and the percentage of pollen in the melissopalynological profile, and it is declared as monofloral or polyfloral (RBB&H, 2009; Haidamus et al., 2019; Hailu & Belay, 2020; Homrani et al., 2020). According to the same Rulebook (RBB&H, 2009), the palynological composition of polyfloral honey samples is richer in the number of plant species than monofloral ones, but the percentage share of each species in the palynological spectrum is below 45%. Honey has an apitherapeutic effect, and it is characterized by numerous biological activities such as: antimicrobial, antioxidant, anti-inflammatory, immunomodulatory and antitumor (Almasaudi et al., 2017; Kolayli et al., 2020; Hailu & Belay, 2020; Horčinová Sedláčková et al., 2021; Güneş, 2021; Celebioglu et al., 2021; McLoone et al., 2021; Balázs et al., 2021; Bhatta, 2022; Hulea et al., 2022; Pătruică et al., 2022; Rojo et al., 2023; Ramić et al., 2023; Shakoori et al., 2024). The biological activities of honey depend on the presence of numerous complex phytochemicals, as well as bioactive compounds that are conditioned by the botanical and geographical origin of honey and contribute to the health benefits of honey (Đogo Mračević et al., 2020; Tasić et al., 2024). Specifically, polyfloral honey samples have up to twice the total phenol content and higher antioxidant capacity than monofloral honey samples (Atanacković Krstonošić et al., 2019).

This research aimed to determine a) specific botanical characteristics and b) the degree of diversity of polyfloral honey samples from different areas of B&H.

Materials and Methods

Material

As part of the research, 50 polyfloral honey samples from B&H were collected and analyzed. The collection of honey samples covered the entire territory of Bosnia and Herzegovina. However, the availability of samples was conditioned by the distribution of apiaries and the willingness of honey producers to cooperate, which is why the sampling did not proportionally follow the current geographical regionalization and ecological differentiation of Bosnia and Herzegovina (Drešković et al., 2011; Gekić et al., 2022).

The geographical distribution of the samples is presented by numerical values in Figure 1.

Methods

Melisopalynological preparations and analysis were made by the RBB&H (2009) as well as the methods proposed by the International Commission for Bee Botany (ICBB) (Von Der Ohe et al., 2004). All melissopalynological preparations were analyzed using a Wild M20 phase-contrast microscope. Plant species identification was performed based on micromorphological features of pollen grains (Erdtman, 1943, 1952; Hesse et al., 2009) and qualitative-quantitative analysis of each sample. Qualitative analysis included a list of determined honey plants, while quantitative analysis involved recording the exact number of pollen grains in the sample. Based on the results of these analysis melissopalynological profiles were created for each sample.

To accurately assess the biodiversity of the samples, both diversity and evenness indices were calculated. The Shannon-Weaver diversity index (H') (Shannon, 2001) was used to evaluate the diversity of the honey samples, calculated using the formula:

$$H' = -(\Sigma (P_i * \ln P_i))$$

To provide a more comprehensive interpretation of sample diversity, the Shannon evenness index (J'), also known as the equitability index, was employed (Magurran, 2013), and is calculated as:

$$J' = H' / H_{max} = H' / \ln S.$$

Results and Discussion

A total of 15,000 pollen grains were identified by melissopalynological analysis of 50 polyfloral honey samples. Based on micropalynological characteristics, 51 pollen types originating from 24 plant families were determined (Figure 2). The plant families with the highest share in melissopalynological profiles were as follows: Fabaceae with 3,480 pollen grains (23.2%), Rosaceae with 2,650 grains (17.67%) and Asteraceae with 2,312 grains (15.41%). Figure 2 does not show the families Chenopodiaceae, Juglandaceae, Cupressaceae, Corylaceae, Caryophyllaceae, Amaryllidaceae, Ranunculaceae and Cornaceae, since their share in the total sample was only 1.8%.

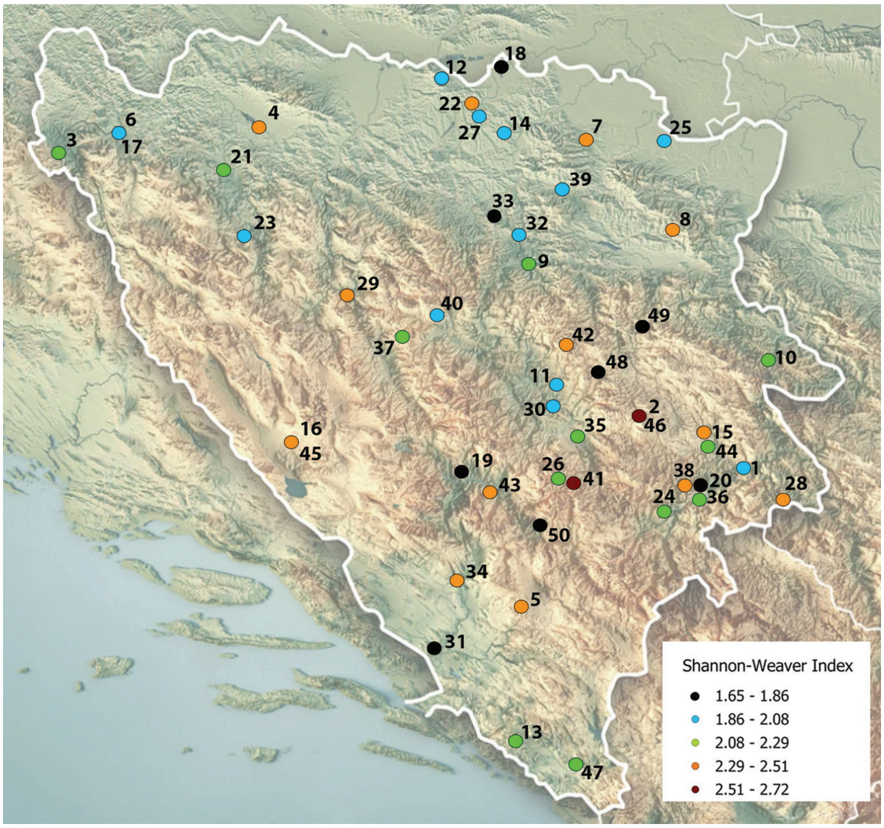

Figure 1: Geographical distribution of analyzed honey samples.

Slika 1: Geografska razporeditev analiziranih vzorcev medu.

1–Ustiprača, Radič, 2–Romanija, 3–Bihać, 4–Prijedor, Petrov gaj, 5–Nevesinje, 6–Bosanska Krupa, 7–Gradačac, 8–Majevica, 9–Zavidovići, 10–Srebrenica, Osmanovići, 11–Breza, 12–Bosanski Kobaš, 13–Trebinje, Bobani, 14–Brezići, 15–Rogatica, 16–Livno, 17–Bosanska Krupa, 18–Bosanski brod, Zborišta, 19– Buturović polje, 20–Goražde, 21–Sanski most, 22–Bosanski Dubačac, 23–Ključ, 24–Ustikolina, 25–Brčko, 26–Trnovo, 27–Derventa, 28–Rudo, 29–Jajce, 30–Ilijaš, 31–Čapljina, 32–Maglaj, 33–Tešanj, 34–Mostar, 35–Hrasno, 36–Goražde, 37–Travnik, Goleš, 38–Goražde, 39–Gračanica, 40–Vlašić, Suho polje, 41–Bjelašnica, Dejčići, 42–Vareš, 43–Konjic, 44–Rogatica, 45–Livno, 46–Romanija, 47–Trebinje, Zubci, 48–Olovo, 49–Kladanj, 50–Crvanj

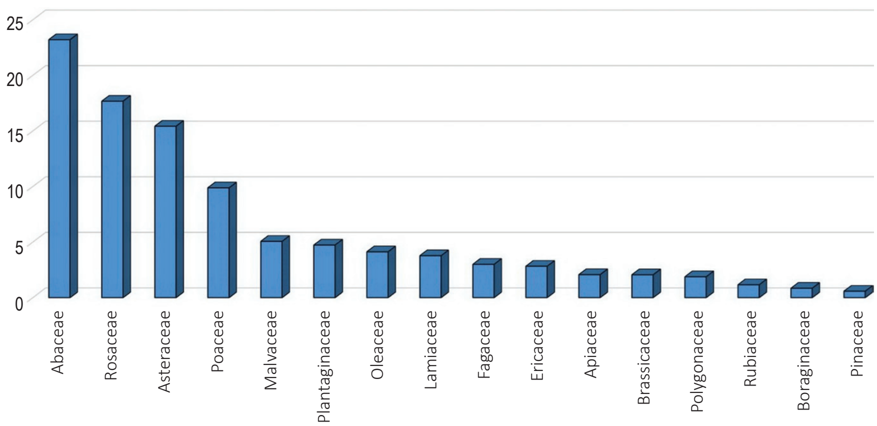

Figure 2: Families with a percentage share of pollen grains above 0.5% in the total sample.

Slika 2: Družine, ki imajo v celotnem vzorcu več kot 0,5 % delež pelodnih zrn

The species from the Asteraceae family were present in 94% of melissopalynological profiles (47 samples). This was followed by the Fabaceae and Poaceae families, which appeared in 88% of the samples (44 samples), and the Rosaceae family, which was present in 82% (41 samples). The Asteraceae family had the highest number of plant species, with seven taxa detected. It was followed by the Fabaceae family with six species, and the Rosaceae family with five species (Table 1). The sample from the Romanija locality (sample number 46) had the highest number of plant species (17), followed by the Livno locality (sample number 45) with 16 species and Vareš with 14 (Table 1). The average number of species per sample was 11.

An overview of the 15 most abundant taxa identified across all analyzed samples is presented in Figure 3. Among these, a total of 1,406 pollen grains were attributed to the genus *Prunus*, followed by *Poa* with 1,357 grains, and *Lotus corniculatus* with 838 grains. *Castanea sativa*, *Cichorium intybus*, and the genus *Potentilla* exhibited the highest mean values across the full dataset, while

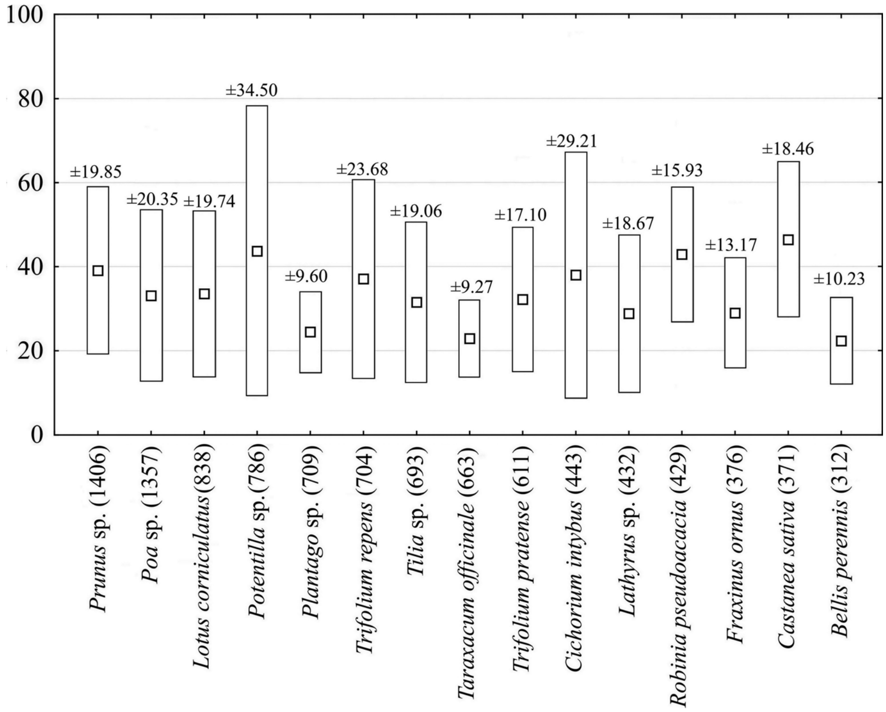

Figure 3: The top 15 most abundant taxa identified in all analyzed samples (for each taxon, minimal, maximal and mean values are presented along with the total number of pollen grains in parentheses. Standard deviations at the top of each bar).

Slika 3: Petnajst najbolj pogostih taksonov v vseh analiziranih vzorcih (za vsak takson so predstavljene minimalna, maksimalna in povprečna vrednost s skupnim številom pelodnih zrn v oklepaju). Na vrhu stolpcev je prikazan standardni odklon.

the greatest standard deviation values were observed for *Potentilla* and *Cichorium*. The highest percentage share in an individual sample was that of *Potentilla* sp. with 121 pollen grains (40.3%) at the Bosanski Brod site (sample number 18 in Table 1), and the lowest was that of *Gallium* sp. with only two pollen grains (0.7%) at the Trebinje, Bobani site (sample number 13 in Table 1).

Pollen grains of ragweed (*Ambrosia artemisiifolia* L.) were detected in melissopalynological profiles from the Bihać and Livno sites. Given that ragweed pollen is characterized by a combination of specific antigens, lactone compounds, flavonoids, monoterpenes and bitter flavonoids and has a very high allergenic potential (Kanter et al., 2013; Chen et al., 2018), according to the European Food Safety Authority, it may pose a serious health problem (EFSA, 2010). It is noteworthy that the actual distribution of ragweed in Bosnia and Herzegovina does not align with the obtained melissopalinological profiles since the time of bee grazing usually does not coincide with its flowering period.

Although the sampling did not follow to the geographical and biogeographic regionalization of Bosnia and Herzegovina (B&H) as outlined in the previous chapter, we can still draw some conclusions. The samples from the Continental region predominantly featured pollen grains from the Asteraceae, Fabaceae, and Rosaceae families (see Table 1, Figure 1). This finding aligns with the results reported for the continental region of neighboring Croatia by Rašić et al. (2018) and Štefanić et al. (2012). In the Mediterranean region of B&H, the dominant pollen grains come from the Fabaceae and Lamiaceae families, a pattern also observed in Croatia (Zima, 2007; Britvec et al., 2013). In the melissopalynological profiles of honey samples originating from the Alpine region of B&H, pollen grains from the Fabaceae and Asteraceae families were notably dominant. It is important to note that this paper analyzed only polyfloral honey types; therefore, their melissopalynological profiles do not accurately reflect the phytocoenological characteristics of the Continental, Mediterranean, and Alpine regions of B&H. An analysis of the botanical characterization of polyfloral honey samples from other countries in the region reveals that, in Serbia, the Asteraceae family shows great potential among plant species (Mačukanović-Jocić et Jarić, 2015). In contrast, Albania demonstrates a pronounced botanical potential within the Ericaceae family (Pupuleku et al., 2016). Melisopalynological studies indicate that the Fabaceae family comprises the most abundant honey plants across different regions of Turkey (Altay et al., 2018; Topal et al., 2023). Meanwhile, in Bulgaria, the families Fabaceae, Brassicaceae, and Rosaceae exhibit the highest botanical potential (Balkanska et al., 2023).

Representation of pollen types in the analyzed samples: dominant pollen grains (≥45%), accessory (from ≥15% to <45%), important (from ≥3% to <15%) and minor (<3%) (Louveaux et al., 1978; Villalpando-Aguilar et al., 2022) (Table 1).

Table 1: Melisopalynological characterization in analyzed profiles of honey samples and values of diversity and evenness indices.
Tabela 1: Melisopalinološke značilnosti analiziranih vzorcev medu in vrednosti indeksov diverzitete in vrstne poravnanosti.

Location	H'	J'	n	Honey plants and proportion of their pollen types in bracket Dominant species in black (≥45%); accessory (from ≥15% to <45%) in red; important in blue (from ≥3% to <15%) and minor (<3%) in green (Louveaux et al., 1978; Villalpando-Aguilar et al., 2022)
Ustiprača, Radič	2.00	0.91	9	*Prunus* sp. (25.33%); *Robinia pseudoacacia* L. (18%); *Castanea sativa* Mill. (14%); Fabaceae (12.33%); *Poa* sp. (11%); Asteraceae (6.67%); *Tilia* sp. (5.67%); *Trifolium repens* L. (5.67%); *Artemisia* sp. (1.33%)
Romanija	2.07	1.00	8	Fabaceae (14.33%); *Calluna vulgaris* (L.) Hull (14%); *Poa* sp. (14%); *Prunus* sp. (13.33%); Apiaceae (12.33%); Asteraceae (10.67%); *Robinia pseudoacacia* L. (10.67%); *Plantago* sp. (10.67%)
Bihać	2.13	0.89	11	*Cichorium intybus* L. (33.67%); *Poa* sp. (9.67%); *Plantago* sp. (9%); *Trifolium repens* L. (7.33%); Asteraceae (7.33%); *Lotus corniculatus* L. (6.67%); Fabaceae (6.67%); Apiaceae (6.67%); *Prunus* sp. (6%); *Tilia* sp. (5%); *Ambrosia artemisiifolia* L. (2%)
Prijedor, Petrov gaj	2.49	0.97	13	*Robinia pseudoacacia* L. (15.67%); *Poa* sp. (11%); *Cichorium intybus* L. (9.67%); *Bellis perennis* L. (9.33%); *Prunus* sp. (8%); Rosaceae (7.33%); *Plantago* sp. (7.33%); *Taraxacum officinale* F. H. Wigg. (6.67%); *Fraxinus ornus* L. (6.33%); Asteraceae (6%); *Rumex* sp. (5.67%); Apiaceae (4%); *Tilia* sp. (3.01%)
Nevesinje	2.30	0.90	13	*Trifolium pratense* L. (22.33%); *Cichorium intybus* L. (14.33%); *Prunus* sp. (12.67%); *Poa* sp. (10.67%); *Lamium* sp. (8.67%); Brassicaceae (8%); *Onobrychis* sp. (5%); *Rumex* sp. (5%); *Myosotis* sp. (4.33%); *Tilia* sp. (3.33%); *Plantago* sp. (2.67%); *Plantago* sp. (2%); Pinaceae (1.67%); *Artemisia* sp. (1.33%)
Bosanska Krupa	1.89	0.91	8	*Poa* sp. (31.33%); *Tilia* sp. (20%); *Cichorium intybus* L. (10.33%); *Prunus* sp. (10%); *Lamium* sp. (9.67%); *Lathyrus* sp. (7.33%); Asteraceae (7%); *Plantago* sp. (4.33%)
Gradačac	2.36	0.95	12	*Poa* sp. (15%); *Tilia* sp. (14%); *Robinia pseudoacacia* L. (13.33%); *Plantago* sp. (10.33%); *Potentilla* sp. (9.67%); *Bellis perennis* L. (8.33%); Brassicaceae (6.33%); *Rumex* sp. (6%); *Prunus* sp. (5.33%); *Fraxinus ornus* L. (5.33%); Apiaceae (5%); *Cirsium* sp. (1.33%)
Majevica	2.50	0.97	13	*Tilia* sp. (13.33%); *Taraxacum officinale* F. H. Wigg. (11.67%); Asteraceae (10.67%); *Trifolium repens* L. (9%); *Lathyrus* sp. (8.67%); *Trifolium pratense* L. (7.33%); *Robinia pseudoacacia* L. (7%); *Potentilla* sp. (7%); *Plantago* sp. (6.33%); Brassicaceae (6.33%); *Echium vulgare* L. (5%); *Chenopodium* sp. (4%); *Lamium* sp. (3.67%)
Zavidovići	2.28	0.95	11	*Robinia pseudoacacia* L. (19.33%); *Plantago* sp. (12%); *Tilia* sp. (11.67%); *Poa* sp. (11%); *Prunus* sp. (10.33%); *Taraxacum officinale* F. H. Wigg. (8.33%); Asteraceae (7.67%); *Potentilla* sp. (7%); *Rumex* sp. (6.33%); *Lathyrus* sp. (4.33%); Pinaceae (2%)
Srebrenica, Osmanovići	2.27	0.91	12	Brassicaceae (20%); *Tilia* sp. (15%); Asteraceae (13%); *Poa* sp. (11%); *Plantago* sp. (10.67%); *Trifolium pratense* L. (8.33%); *Malva* sp. (7.33%); *Fraxinus ornus* L. (4%); *Artemisia* (3.33%); *Cirsium* sp. (2.67%); Lamiaceae (2.33%); *Chenopodium* sp. (2.33%)
Breza, Bukovik	2.02	0.84	11	*Sanguisorba minor* Scop. (33.33%); *Robinia pseudoacacia* L. (15.67%); *Prunus* sp. (15%); *Plantago* sp. (7.67%); *Taraxacum officinale* F. H. Wigg. (7.33%); *Poa* sp. (5%); *Rubus* sp. (5%); *Rumex* sp. (3.33%); Asteraceae (3.33%); Pinaceae (2.33%); Apiaceae (2%)
Bosanski Kobaš	1.92	0.84	10	*Potentilla* sp. (36%); *Tilia* sp. (14.33%); *Prunus* sp. (11.67%); *Cichorium intybus* L. (11.33%); *Lotus corniculatus* L. (9.33%); *Zea mays* L. (6%); Pinaceae (5.33%); *Taraxacum officinale* F. H. Wigg. (2.67%); *Rubus* sp. (1.67%); *Lamium* sp. (1.67%)
Trebinje, Bobani	2.17	0.91	11	Ericaceae (19.33%); Fabaceae (16.67%); *Cichorium intybus* L. (12.67%); *Lathyrus* sp. (12.67%); Apiaceae (9%); Lamiaceae (9%); *Plantago* sp. (8.33%); *Lotus corniculatus* L. (7%); *Lamium* sp. (2.67%); *Salvia officinalis* L. (2%); *Gallium* sp. (0.67%)
Brezići	1.94	0.89	9	Ericaceae (31.67%); *Prunus* sp. (15.33%); Brassicaceae (15%); *Trifolium pratense* L. (11.67%); *Poa* sp. (7.33%); Lamiaceae (5.67%); *Fraxinus ornus* L. (5.67%); *Cornus mas* L. (5.67%); Pinaceae (2%)

Location	H'	J'	n	Honey plants and proportion of their pollen types in bracket Dominant species in black (≥45%); accessory (from ≥15% to <45%) in red; important in blue (from ≥3% to <15%) and minor (<3%) in green (Louveaux et al., 1978; Villalpando-Aguilar et al., 2022)
Rogatica	2.36	0.95	12	*Rumex* sp. (18.67%); *Prunus* sp. (10.33%); *Poa* sp. (10%); *Bellis perennis* L. (9.67%); *Lamium* sp. (9%); *Lotus corniculatus* L. (8.33%); *Trifolium pratense* L. (8.33%); Brassicaceae (7.33%); *Plantago* sp. (7%); *Artemisia* sp. (6%); Apiaceae (2.33%); Asteraceae (1.67%)
Livno	1.82	0.88	8	*Lotus corniculatus* L. (27.33%); *Gallium* sp. (22.67%); *Poa* sp. (20%); *Cichorium intybus* L. (11.67%); Apiaceae (6.67%); *Juglans regia* L. (5%); Brassicaceae (3.33%); *Rumex* sp. (3.33%)
Bosanska krupa	2.00	0.96	8	*Tilia* sp. (24%); *Castanea sativa* Mill. (16%); *Potentilla* sp. (13.33%); *Poa* sp. (11.67%); *Sanguisorba minor* Scop (11%); *Bellis perennis* L. (10.67%); *Plantago* sp. (6.67%); *Prunus* sp. (6.67%)
Bosanski brod, Zborišta	1.85	0.84	9	*Potentilla* sp. (40.33%); *Prunus* sp. (15.67%); *Poa* sp. (8.67%); *Tilia* sp. (8.33%); *Taraxacum officinale* F. H. Wigg. (8%); *Lathyrus* sp. (6.67%); *Plantago* sp. (5%); Apiaceae (4%); *Zea mays* L. (3.33%)
Buturović polje	1.84	0.89	8	*Potentilla* sp. (34.67%); *Castanea sativa* Mill. (20%); *Lathyrus* sp. (10.67%); *Tilia* sp. (9%); *Taraxacum officinale* F. H. Wigg. (8.33%); *Poa* sp. (7.33%); *Calluna vulgaris* (L.) Hull (6%); Lamiaceae (4%)
Goražde	1.71	0.78	9	*Trifolium repens* L. (38.33%); *Lotus corniculatus* L. (26%); *Plantago* sp. (11.67%); *Bellis perennis* L. (7.67%); *Trifolium pratense* L. (5%); Lamiaceae (4.67%); *Artemisia* sp. (3.33%); *Taraxacum officinale* F. H. Wigg (1.67%); *Chenopodium* sp. (1.67%)
Sanski most	2.19	1.00	9	*Prunus* sp. (14%); *Trifolium repens* L. (12.67%); *Lotus corniculatus* L. (12.67%); *Lamium* sp. (11%); *Plantago* sp. (11%); *Taraxacum officinale* F. H. Wigg (10%); *Tilia* sp. (10%); *Zea mays* (9.67%); Asteraceae (9%)
Bosanski dubačac	2.50	0.95	14	*Lotus corniculatus* L. (13%); *Tilia* sp. (12.33%); *Poa* sp. (12%); *Bellis perennis* L. (10.33%); *Prunus* sp. (10.33%); *Lamium* sp. (7%); *Trifolium pratense* L. (6.33%); *Potentilla* sp. (6%); *Plantago* sp. (6%); *Gallium* sp. (5.33%); *Artemisia* sp. (3.67%); *Zea mays* L. (3%); *Onobrychis* sp. (3%); *Ranunculus* sp. (1.67%)
Ključ	1.87	0.90	8	*Cichorium intybus* L. (36.67%); *Poa* sp. (12.67%); *Lotus corniculatus* L. (11%); *Onobrychis* sp. (9%); *Prunus* sp. (8.67%); *Taraxacum officinale* F. H. Wigg. (8.33%); *Trifolium repens* L. (7%); *Tilia* sp. (6.67%)
Ustikolina, Goražde	2.21	0.96	10	*Prunus* sp. (21.11%); *Rubus* sp. (14.33%); *Potentilla* sp. (11.33%); *Fraxinus ornus* L. (11.33%); *Poa* sp. (9.67%); *Sanguisorba minor* Scop (7.67%); *Carpinus* sp. (6.33%); Asteraceae (6.33%); Cupressaceae (6%); *Rumex* sp (5.67%)
Brčko	1.97	0.90	9	*Prunus* sp. (29.33%); *Trifolium pratense* L. (19.33%); *Lotus corniculatus* L. (10.33%); *Echium vulgare* L. (9.67%); *Rumex* sp. (9.33%); Asteraceae (9.33%); *Poa* sp. (5.67%); *Plantago* sp. (4.67%); *Ranunculus* sp. (2.33%)
Trnovo, Sarajevo	2.11	0.92	10	Asteraceae (29.67%); *Echium vulgare* L. (9.67%); *Lotus corniculatus* L. (8.67%); *Taraxacum officinale* F. H. Wigg. (8.67%); *Tilia* sp. (7%); *Poa* sp. (7.33%); *Artemisia* sp. (7.67%); *Prunus* sp. (12.33%); *Potentilla* sp. (6.33%); *Zea mays* L. (2.67%)
Derventa	1.94	0.93	8	*Robinia pseudoacacia* L. (19.33%); Asteraceae (19.33%); *Potentilla* sp. (18.67%); *Taraxacum officinale* F. H. Wigg (13.33%); *Trifolium pratense* L. (12.67%); *Prunus* sp. (7.67%); *Plantago* sp. (6.67%); *Zea mays* L. (2.33%)
Rudo	2.41	0.97	12	*Lotus corniculatus* L. (15%); *Onobrychis* sp. (12.67%); *Plantago* sp. (11.33%); Ericaceae (9.33%); *Trifolium pratense* L. (9.33%); *Taraxacum officinale* F. H. Wigg. (8.33%); *Chenopodium* sp. (8%); *Poa* sp. (6.67%); Lamiaceae (6%); *Lamium* sp. (5%); *Trifolium repens* L. (5%); Asteraceae (3.33%)
Jajce	2.50	0.97	13	*Tilia* sp. (12%); *Plantago* sp. (11%); Asteraceae (10%); *Poa* sp. (9.33%); Brassicaceae (9%); *Prunus* sp. (9%); *Sanguisorba minor* Scop (7.67%); *Taraxacum officinale* F. H. Wigg. (7.67%); *Castanea sativa* Mill. (6.33%); *Lotus corniculatus* L. (6.33%); *Potentilla* sp. (5.67%); *Bellis perennis* L. (3.33%); *Zea mays* L. (2.67%)

Location	H'	J'	n	Honey plants and proportion of their pollen types in bracket Dominant species in black (≥45%); accessory (from ≥15% to <45%) in red; important in blue (from ≥3% to <15%) and minor (<3%) in green (Louveaux et al., 1978; Villalpando-Aguilar et al., 2022)
Ilijaš, Sarajevo	1.97	0.90	9	*Prunus* sp. (25%); *Fraxinus ornus* L. (19%); *Sanguisorba minor* Scop (16.67%); *Potentilla* sp. (13.33%); *Taraxacum officinale* F. H. Wigg. (9.33%); *Zea mays* L. (6%); *Robinia pseudoacacia* L. (4.67%); Brassicaceae (4%); *Fagus* sp. (2%)
Čapljina	1.77	0.85	8	Asteraceae (32.33%); Lamiaceae (22.67%); Rosaceae (15%); *Poa* sp. (10.67%); Fabaceae (10%); Apiaceae (4.67%); *Satureja* sp. (3.67%); *Salvia officinalis* L. (1%)
Maglaj	2.01	0.97	8	*Poa* sp. (19.33%); *Robinia pseudoacacia* L. (19.33%); *Plantago* sp. (14.33%); *Trifolium pratense* L. (13.33%); *Taraxacum officinale* F. H. Wigg. (11.33%); *Fagus* sp. (7.67%); Asteraceae (7.67%); *Tilia* sp. (7%)
Tešanj	1.77	0.85	8	Oleaceae (36.67%); Ericaceae (19%); *Prunus* sp. (13.33%); *Fraxinus ornus* L. (12%); *Taraxacum officinale* F. H. Wigg. (7.67%); *Bellis perennis* L. (5%); *Poa* sp. (3.33%); Caryophyllaceae (3%)
Mostar	2.31	0.90	13	*Prunus* sp. (20%); Oleaceae (19%); *Fraxinus ornus* L. (13.33%); *Lotus corniculatus* L. (8.33%); Cupressaceae (7%); Lamiaceae (6.67%); Brassicaceae (5%); *Trifolium pratense* L. (5%); *Onobrychis* sp. (5%); *Taraxacum officinale* F. H. Wigg. (3.33%); *Lathyrus* sp. (3.33%); *Carpinus* sp. (2.67%); *Potentilla* sp. (1.33%)
Hrasno	2.08	0.95	9	*Castanea sativa* Mill. (23.33%); *Fraxinus ornus* L. (13.67%); *Trifolium pratense* L. (13%); *Poa* sp. (12.67%); *Lathyrus* sp. (11%); *Plantago* sp. (10%); *Lotus corniculatus* L. (7%); Rosaceae (5%); Apiaceae (4.33%)
Goražde	2.24	0.94	11	*Lathyrus* sp. (24.67%); *Trifolium repens* L. (13.67%); *Prunus* sp. (9.67%); *Lotus corniculatus* L. (9.33%); *Poa* sp. (9.33%); *Taraxacum officinale* F. H. Wigg. (7.67%); *Onobrychis* sp. (6%); *Trifolium pratense* L. (5.67%); *Fraxinus ornus* L. (5%); *Lamium* sp. (5%); Pinaceae (4%)
Travnik, Goleš	2.25	0.94	11	*Prunus* sp. (21%); *Trifolium repens* L. (16.33%); *Trifolium pratense* L. (11.67%); *Castanea sativa* Mill. (9%); Asteraceae (8.33%); *Fraxinus ornus* L. (8%); *Lathyrus* sp. (7%); *Lamium* sp. (7%); *Artemisia* sp. (4.33%); *Gallium* sp. (4%); *Poa* sp. (3.33%)
Goražde	2.31	0.93	12	*Trifolium repens* L. (15.33%); *Lotus corniculatus* L. (15%); *Poa* sp. (14%); *Taraxacum officinale* F. H. Wigg. (11%); *Trifolium pratense* L. (10%); *Rumex* sp. (9.33%); Pinaceae (8.67%); *Gallium* sp. (4.67%); Apiaceae (4%); *Cichorium intybus* L. (3.33%); *Bellis perennis* L. (3%); *Onobrychis* sp. (1.67%)
Gračanica	2.02	0.88	10	*Poa* sp. (26.33%); *Tilia sp.* (24%); *Plantago* sp. (11.33%); *Taraxacum officinale* F. H. Wigg. (11%); Asteraceae (6%); *Rumex* sp. (6%); *Gallium* sp. (5.33%); *Prunus* sp. (4%); *Lotus corniculatus* L. (3.67%); *Trifolium repens* L. (2.33%)
Vlašić, Suho polje	2.02	0.92	9	*Lotus corniculatus* L. (25.67%); *Prunus* sp. (14.33%); *Trifolium repens* L. (13.33%); *Potentilla* sp. (12.67%); *Plantago* sp. (12%); *Taraxacum officinale* F. H. Wigg. (11%); *Poa* sp. (4%); *Gallium* sp. (3.67%); *Trifolium pratense* L. (3.33%)
Bjelašnica, Dejčići	2.52	0.98	13	*Poa* sp. (12%); *Onobrychis* sp. (10.67%); *Allium* sp. (9.33%); *Lotus corniculatus* L. (8.67%); *Trifolium repens* L. (8.33%); *Lamium* sp. (8%); *Trifolium pratense* L. (7.67%); Lamiaceae (7%); *Rumex* sp. (7%); *Bellis perennis* L. (7%); *Prunus* sp. (6.67%); *Gallium* sp. (4.33%); Apiaceae (3.33%)
Vareš	2.46	0.93	14	Apiaceae (15%); *Trifolium repens* L. (15%); *Fraxinus ornus* L. (11.67%); *Poa* sp. (11%); *Lathyrus* sp. (7.33%); *Prunus* sp. (7.33%); *Lotus corniculatus* L. (7%); *Fagus* sp. (5.67%); *Onobrychis* sp. (5%); Lamiaceae (4.67%); *Bellis perennis* L. (3.67%); *Tilia* sp. (2.33%); *Malva* sp. (2.33%); *Taraxacum officinale* F. H. Wigg. (2%)
Konjic	2.29	0.92	12	*Trifolium pratense* L. (23.33%); *Castanea sativa* Mill. (12.67%); *Fagus* sp. (10.67%); *Taraxacum officinale* F. H. Wigg. (10%); *Poa* sp. (9.33%); Lathyrus sp. (8.33%); *Bellis perennis* L. (6.33%); *Prunus* sp. (6%); *Juglans regia* L. (4.33%); *Onobrychis* sp. (3%); Caryophyllaceae (3%); *Rumex* sp. (3%)

Location	H'	J'	n	Honey plants and proportion of their pollen types in bracket Dominant species in black (≥45%); accessory (from ≥15% to <45%) in red; important in blue (from ≥3% to <15%) and minor (<3%) in green (Louveaux et al., 1978; Villalpando-Aguilar et al., 2022)
Rogatica	2.25	0.94	11	*Potentilla* sp. (21.33%); *Bellis perennis* L. (15%); Brassicaceae (10.67%); *Artemisia* sp. (9.33%); *Poa* sp. (8.67%); Apiaceae (8%); *Taraxacum officinale* F. H. Wigg. (7.33%); *Helianthus annuus* L. (6.67%); *Zea mays* L. (5.67%); *Plantago* sp. (5.67%); *Malva* sp. (1.67%)
Livno	2.38	0.86	16	Fabaceae (34.33%); *Lotus corniculatus* L. (9.33%); *Taraxacum officinale* F. H. Wigg. (6%); *Poa* sp. (5%); *Rumex* sp. (5%); *Lathyrus* sp. (4.67%); *Bellis perennis* L. (4.67%); *Echium vulgare* L. (4.33%); *Poa* sp. (4.33%); *Carpinus* sp. (4%); *Cichorium intybus* L. (4%); *Malva* sp. (4%); *Trifolium repens* L. (4%); *Ambrosia artemisiifolia* L. (3.67%); Boraginaceae (3.33%); Asteraceae (2.67%); *Chenopodium* sp. (1.67%)
Romanija	2.72	0.96	17	*Trifolium repens* L. (13.33%); *Potentilla* sp. (13%); *Lotus corniculatus* L. (8.33%); *Lamium* sp. (6.67%); *Plantago* sp. (6.67%); *Poa* sp. (6%); *Echium vulgare* L. (5.67%); *Taraxacum officinale* F. H. Wigg. (5.33%); *Lathyrus* sp. (5%); *Malva* sp. (4.67%); *Cirsium* sp. (4.67%); Asteraceae (4%); *Gallium* sp. (3.67%); *Ranunculs* sp. (3.33%); Apiaceae (3.33%); *Prunus* sp. (3.33%); Lamiaceae (3%)
Trebinje, Zubci	2.12	0.88	11	Oleaceae (25%); *Prunus* sp. (16.67%); *Lotus corniculatus* L. (14.33%); *Trifolium repens* L. (13.67%); Lamiaceae (7.33%); Brassicaceae (6.33%); Apiaceae (4.67%); *Juglans regia* L. (4%); *Taraxacum officinale* F. H. Wigg. (3.67%); *Gallium* sp. (2.33%); *Malva* sp. (2%)
Olovo	1.85	0.84	9	*Poa* sp. (33.33%); *Lathyrus* sp. (22.33%); *Trifolium repens* L. (14.67%); Asteraceae (10%); *Plantago* sp. (4.67%); *Potentilla* sp. (4.33%); Lamiaceae (4%); Caryophyllaceae (3.33%); *Taraxacum officinale* F. H. Wigg. (3.33%)
Kladanj	1.75	0.70	12	Ericaceae (41.67%); *Prunus* sp. (25.67%); *Fraxinus ornus* L. (10%); Asteraceae (4.67%); Apiaceae (4%); *Poa* sp. (3.67%); *Tilia* sp. (3%); Pinaceae (2.67%); *Plantago* sp. (1.33%); *Chenopodium* sp. (1.33%); *Gallium* sp. (1%); *Lotus corniculatus* L. (1%)
Crvanj	1.65	0.92	6	Rosaceae (28%); *Castanea sativa* Mill. (22.33%); *Trifolium repens* L. (19.67%); *Prunus* sp. (17.33%); Lamiaceae (9.67%); *Poa* sp. (3%)

H'- Shannon-Weaver index, J'- Shannon uniformity index, n- number of species in the sample

The degree of the sample's biodiversity was calculated by the Shannon-Weaver index (Table 1, Figure 1). The highest index values were at the locations of Romanija (2.72) and Bjelašnica (2.52), and the lowest values were at the locations of Goražde (1.71) and Crvanj (1.65). According to Pešić & Tomović (2011), the diversity index increases as the number of honey-bearing species in the sample rises because the proportion of individual species decreases, leading to a more negative value in the logarithm. The results of other studies show that the values of the Shannon-Weaver index for samples originating from Italy ranged from 0.01 to 2.37 (Conti et al., 2016), Mexico from 0.93 to 1.16 (Villalpando-Aguilar et al., 2022), while in China the values index from 1.79 to 2.21 (Song et al., 2012). In Ethiopia, the index values were higher than the previous ones and ranged from 2.377 to 2.541 (Dugda & Addi, 2024). By comparing the results, we can see that honey samples originating from B&H have similar or slightly higher diversity index values. High values of the Shannon-Weaver index and the botanical specificity of each melissopalynological profile (Table 1) reflect the diversity of honey plants and represent the extremely high honey and ecological potential of B&H in honey production.

The interpretation of the honey samples' diversity is complemented by the evenness or equitability index of the samples (Table 1). Comparing two or more samples with the same number of species, the one with species with an uniform number of individuals has a higher degree of diversity, and the one in which some species dominate while others are present with a small number of individuals in the community is less diverse (Herrmann et al., 2022). In our study, the values of the uniformity index (J') was from 0.70 to 1, showing that all samples had moderate to uniform numbers of pollen grains of different types in the sample.

The high values of the diversity index and uniformity of the analyzed melissopalynological profiles are the outcome of the floristic richness and uniqueness of the honey-bearing pastures of B&H. Dynamic changes in geomorphological, climatic and edaphic parameters have caused heterogeneous habitat conditions in a relatively small geographical area (Redžić, 2007; Redžić et al., 2008). The complex action of ecological factors has enabled the specific chorology and phenology of honey-bearing plants, which is directly reflected in the unique botanical and chemical properties of honey (Homrani et al., 2020; Yıldırım et al., 2021; Ivanović et al., 2021). Ecological heterogeneity has made B&H a biodiversity hotspot in Europe (Grimmett et al., 2017), and one of the most diverse countries in Europe with 4498 described Spermatophyta (Redžić et al., 2008). All this is reflected in the diversity of melissopalynological profiles where the combination of melliferous plants represents a unique imprint of the environment in which melliferous grazing was carried out (Table 1).

Conclusion

As part of the research, 50 polyfloral honey samples from Bosnia and Herzegovina were analyzed, with 15,000 pollen grains from 24 different plant families. Melisopalynological analysis established the botanical uniqueness of each investigated profile. Bosnia and Herzegovina is characterized by a high level of biodiversity (Redžić, 2007; Redžić et al., 2008) which is directly reflected in the floristic richness and diversity of the investigated melisopalynological profiles. The results showed the potential and biodiversity of honey plants and are crucial in expanding knowledge about the diversity of the apiflora of B&H, sustainable use of this important biological resource and increasing the competitiveness of local domestic honey producers on the international market.

Research data availability

The data analysed during this study are available from the corresponding author upon reasonable request.

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