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PALYNOLOGICAL ANALYSES, CHEMICAL and MINERAL COMPOSITION OF SOME HONEYBEE POLLEN PELLETS

Bazı Bal Arısı Polenlerinin Palinolojik Analizleri, Kimyasal Ve Mineral Madde İçerikleri

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ABSTRACT

Bee products have gained popularity in recent years as food, dietary supplements and adjuvant products due to their extraordinary health properties. Globally, bee pollen is the second most consumed product after honey, has a special importance as a male reproductive unit of flowers, as well as a rich nutrient material, as it contains the oral secretions of the honeybee. In this study, palynological identification of bee pollen collected from apiaries from different regions was made. The bee pollen content was found to be consisting of dry matter between 71.47-81.38%, protein between 17.5-26.0%, fat 5.84-10.95%, and ash content 2.02-2.44%. Moreover, the most common mineral elements in pollen were calcium, potassium, magnesium, silicon, sodium and iron. Besides, heavy metals such as arsenic, cadmium and lead have been detected in bee pollen samples that is a result of the increased environmental pollution and have negative effects on health. Therefore, in bee pollen production, identification, determination of nutritional quality and standardization of pollen are very important for producers and consumers.

Keywords: Bee pollen, palynological analysis, protein and mineral content chemical composition

ÖZ

Arı ürünleri sağlık açısından olağanüstü özellikleri nedeniyle son yıllarda gıda, gıda takviyesi ve destekleyici ürünler olarak ilgi görmektedir. Dünyada baldan sonra tüketimi hızla artan arı poleni, çiçeklerin erkek üreme birimi olarak zengin bir besin maddesi yanında, bal arısının ağız salgıları içermesiyle ayrı bir öneme sahiptir. Bu çalışmada farklı bölgelerdeki arılıklardan toplanan arı polenlerinin olarak palinolojik tanımlaması yapılmıştır. Polen içeriğinin belirlenmesine yönelik yapılan analizlerde kuru madde %71,47-81,38 aralığında, protein %17,5-26,0 aralığında, yağ %5,84-10,95 ve kül miktarı ise %2,02-2,44 arasında bulunmuştur. Mineral madde kompozisyonuna bakıldığında en çok bulunanlar; kalsiyum, potasyum, magnezyum, silisyum, sodyum ve demir elementidir. Çevre kirliliğinin artmasının bir sonucu olan ve sağlık üzerine olumsuz etkileri bulunan ağır metallerden

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arsenik, kadmiyum ve kurşun arı poleni örneklerinde tespit edilmiştir. Polen üretiminde özellikle ticari olarak satışı yapılan polenlerin tanımlanması, besinsel kalitesinin belirlenmesi ve standardizasyonu üretici ve tüketici açısından oldukça önemlidir.

Anahtar Kelimeler: Arı poleni, palinolojik analiz, protein, mineral madde

GENİŞLETİLMİŞ ÖZET

Amaç: Türkiye koloni varlığı ve ürettiği bal ile dünyanın önde gelen ülkelerinden biridir. Bal üretimi haricinde üretimi yaygınlaşan ikinci bir ürün de arı polenidir. Arı ürünlerinin doğal ürün olması ve tüketicilerin sağlıklı beslenme için bu ürünlere karşı olan talebi her gün artmaktadır. Bulunduğu topraklar ve sahip olduğu iklimler nedeniyle bitki çeşitliliği yönünden zengin olan Türkiye, arı poleni üretiminin çeşitlendirerek tüketiciye alternatifler sunması gerekmektedir. Bu çalışmada özellikle arıcılık üretimi açısından önemli bitkilerden elde edilen arı polenlerinin kimyasal ve mineral madde özelliklerinin ortaya konulması amaçlanmıştır.

Gereç-Yöntem: Örnekler İzmir, Balıkesir ve Afyon illerinden haftalık olarak Langstroth tip çekmeceli kovanlardan alınmış ve analizleri yapılmaya kadar -20°C'de muhafaza edilmiştir. Örneklerin palinolojik inceleme ve tayinleri Bilisik v.d. (2008) metoduna göre ışık mikroskobu ile yapılmıştır. Polen örneklerinde (%) kuru madde, kül miktarı, protein ve yağ tayini yapılmıştır (AOAC 2000, Almeida-Muradian v.d., 2005, Commission 2009). Son olarak mineral madde analizi (NMKL-186, 2007) yapılarak arı poleni örneklerinin analiz sonuçları değerlendirilmiştir. İstatistiksel analizler JMP Pro 13 (SAS) programı kullanılarak tekrarlı ANOVA yöntemiyle yapılmıştır.

Bulgular: Polen örneklerinde kuru madde %71,47-81,38, protein %17,50-26,00, yağ %5,84-10,95 arasında ve kül miktarı %2,02-2,44 arasında tespit edilmiştir. Polen örneklerinde en çok bulunan mineraller potasyum, kalsiyum, magnezyum, silisyum, demir ve sodyumdur. *Scabiosa* L. ve *Salix* L. polenlerinin magnezyum yönünden, *Castanea sativa* Mill. polenlerinin ise demir, manganez ve sodyum minerali yönünden zengin olduğu belirlenmiştir. Ayrıca arı poleni örneklerinde ağır metallere arsenik, kurşun ve kadmiyum elementleri tespit edilmiştir. En çok arsenik ve kurşun ağır metalleri *Scabiosa* L. poleninde tespit edilmiştir.

Sonuç: Polen örneklerinin kimyasal analiz sonuçları daha önceki yapılmış çalışmalar ile uyumlu gözükmektedir. Türkiye'de üretilen arı polenleri özellikle Ca, K, Mg, Fe, Si ve eser elementleri açısından iyi bir mineral kaynağı olarak değerlendirilebilir. En önemli konu artan çevre kirliliğine karşı polende oluşabilecek sağlık risklerinin (ağır metal gibi) tespit edilmesine yöneliktir. Arı poleninin riskli gruplar (yaşlılar, hamileler, hastalar) tarafından tüketilebileceği düşünüldüğünde, hem üreticilerin hem de tüketicilerin ortaya çıkabilecek bu soruna karşı dikkatli olmaları gerekmektedir. Arı poleni hakkında bilimsel çalışmalar yapıldıkça polen kullanımının sağlık için önemi artmaktadır.

Özellikle ticari bir ürün olan arı poleninin piyasada doğru tanıtılması önemlidir. Bu nedenle ticari firmaların büyük çaplı polen stoklarının palinolojik analizinin yapılması ve kalitelerinin ortaya konulması üretici ve tüketici için oldukça önemlidir.

INTRODUCTION

Proteins, sugars, lipids, amino acids, vitamins and mineral elements are the main components in bee pollen (BP) (Szczęsna 2007a, Hassan 2011, Liolios et al. 2016, Velásquez et al. 2017, Lilek et al. 2021). BP is important for human health and nutrition. The chemical components of BP differ based on plant species. There is a correlation between chemical composition and botanical origin of the pollen (Taha 2015). BP consists of 20.0-30.4% protein, 2.8-50.2 mg/kg carotenoids and 22.8-918.4 mg/kg phenolic components (Velásquez et al. 2017). BP has high sugar and protein content and relatively low lipid content. Furthermore, pollen is a rich source of vitamins and other bioactive compounds. It was reported that 8 important minerals (calcium, iron, copper, chromium, manganese, molybdenum, phosphorus, and zinc) were found and six of them were high enough to meet dietary requirements (Sattler et al. 2016, Mărgăoan et al. 2014). Most of the daily consumption requirements of these minerals can be met with the consumption of 20-30 g of pollen (Ialomiteanu 1978). According to a

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systematic review of more than 100 studies, BP consists of an average of 54.22% (18.50–84.25%) carbohydrates, 21.30% (4.50–40.70%) protein, 5.31% (0.41–13.50%) lipids, 8.75% (0.15–31.26%) fiber, and 2.91% (0.50–7.75%) ash. Besides, the presence of mineral content in BP was reported as: 4951.61 mg/kg (3.06–13366.60 mg/kg) potassium, 4157.86 mg/kg (234.40–9587.00 mg/kg) phosphorus, 1751.22 mg/kg (1.09–5752.19 mg/kg) calcium, 1246.99 mg/kg (44.00–4680.53 mg/kg) magnesium, 46.97 mg/kg (0.10–105.80 mg/kg) zinc, and 197.41 mg/kg (2.60–1180.00 mg/kg) iron (Thakur and Nanda 2020).

BP consumption has increased in recent years, particularly due to its nutritional value and therapeutic applications. Quantification of mineral components is of great importance to evaluate both toxicity and beneficial effects of essential elements (Costa et al. 2019). It shows that bee products contain essential macro elements (K, P and S) and micro elements (Zn and Fe) in concentrations depending on the bee product type. However, the presence of toxic heavy metals makes it necessary to investigate the quality of bee products before using them as nutritional supplements. Since bee products are quite heterogeneous, they also differ in element content depending on environmental factors. Therefore, it is necessary to develop standards that regulate acceptable inorganic pollutant levels. Moreover, since bees and their products are considered an effective biological monitoring tool, analysis results can reflect the state of the environment that affects the health and well-being of both humans and bees (Matuszewska et al. 2021). In the present study, mineral content, protein, fat, dry matter, and ash content in some monofloral BP and mixed BP produced in Turkey were investigated.

MATERIALS AND METHODS

The 7 different BP samples were collected between April and July 2020. Samples 3 and 4 were taken from Izmir, sample 2 was taken from Balıkesir and the other samples (1,5,6,7) were taken from Afyon. All pollen samples were harvested weekly from Langstroth type drawer hives under similar conditions and stored at -20°C until analysis.

Palynological Identification of Pollen Samples

To determine the plant sources used by honeybees in the region, fresh pollen granules were prepared as

specimens by separating them according to their colors (Wodehouse 1935). Palynological identifications were made using a light microscope (Olympus BX41) and the percentages of pollen belonging to each taxon in each sample were determined (Almeida-Muradian et al. 2005, Bilisik et al. 2008, Mărgăoan et al. 2013).

Chemical Analysis of Pollens

Moisture Analysis: Moisture content of pollen samples was determined by Radwag 50/NH moisture analyzer (103°C, 3-4 hour) by drying the samples until they reached a constant weight (Commission 2009).

Ash Analysis: Pollen samples were weighed into porcelain crucibles (2 g) and burned in a muffle furnace at 600°C until no black spots remained in the sample. Then, after cooling in the desiccator, weighing was made and the results were calculated as % (AOAC 2000).

Protein Assay: The amount of protein was determined according to the Dumas method. Leco FP-528 nitrogen/protein analyzer is used to determine the protein amount of the samples (AOAC 2000). Pollen samples were burned in the protein analyzer. The amount of nitrogen obtained as a result of combustion was multiplied by 5.60 and the amount of protein was calculated (Rabie et al. 1983, Sorkun et al. 2010; Çakıcı et al. 2018).

Fat Assay: The amount of fat was determined using diethyl ether with the Soxhlet extraction method in a semi-continuous solvent extraction system (Almeida-Muradian et al. 2005). The results were calculated as % of weight loss (Yetim and Keskin 2008).

Mineral analysis: Pollen samples were burned in a microwave system at 200 °C with nitric acid and hydrogen peroxide. Then, the minerals were determined with the ICP-MS device (NMKL-186 2007).

Statistical Analysis

Statistical analyzes were performed in JMP Pro 13 (SAS) software using the repeated ANOVA method. Chemical analysis of the samples was done in duplicate, and mineral analyzes were performed in triplicate. Results were expressed as mean ± standard deviation. The heatmap and principal components plots were prepared using the Clustvis webtool (<https://biit.cs.ut.ee/clustvis/>).

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RESULTS

From the total samples used in the study, one of the pollen types was found to be more than 90%. Thus, sample 1 was found to be predominant in *Salix* (94.17%), sample 2 in *Castanea sativa* (99.50%) and sample 3 in *Scabiosa* pollen (99.40%). In addition to these samples, which can be considered

monotypic, no dominant pollen was observed in other samples, and they were accepted as polytypic. Majority of the pollen was represented by *Brassicaceae* (38.10%) and *Echium* sp. (34.01%) in the 4th sample, while *Brassicaceae* (32.69%) and *Apiaceae* (30.77%) families in the 5th sample *Salix* (51.33%) and *Rosaceae* (30.51%) in the 6th sample, and *Papaver* (44.50%) in the 7th sample (Table 1).

Table 1. Percentage of pollen types represented in studied pollen pellets.

Çizelge 1. Polen örneklerini temsil eden polen türleri (%)

| Taxa / Sample | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|------------------------|-------|------|-------|-------|-------|-------|-------|
| <i>Acer</i> | | | | | | 10,90 | |
| <i>Apiaceae</i> | | | | | 30,77 | | 9,09 |
| <i>Asteraceae</i> | | | | | | | 1,21 |
| <i>Brassicaceae</i> | 0,82 | | | 38,10 | 32,69 | | 5,45 |
| <i>Castanea sativa</i> | | 99,5 | | | | | |
| <i>Centaurea</i> | | 0 | | 3,40 | | | |
| <i>Cistus</i> | | 0,5 | | 2,72 | 6,73 | | 9,09 |
| <i>Echium</i> | | | | 34,01 | | | 1,82 |
| <i>Eucalyptus</i> | | | | 9,52 | | | 7,88 |
| <i>Fabaceae</i> | | | | | 0,96 | | |
| <i>Juglans</i> | | | | | | 3,63 | |
| <i>Lamiaceae</i> | | | | 1,36 | 2,88 | | |
| <i>Onobrychis</i> | | | | | 0,96 | | 1,82 |
| <i>Papaver</i> | | | | 2,04 | 16,35 | | 44,85 |
| <i>Plantago</i> | | | | | 0,96 | | |
| <i>Poaceae</i> | | | | | 4,81 | | |
| <i>Portulaca</i> | | | 0,60 | | | | |
| <i>Ranunculaceae</i> | | | | | | 2,18 | |
| <i>Roemeria</i> | | | | | 1,92 | | 3,64 |
| <i>Rosaceae</i> | 5,01 | | | 4,76 | | 30,51 | 15,15 |
| <i>Salix</i> | 94,17 | | | | | 51,33 | |
| <i>Sarco/Poterium</i> | | | | | 0,96 | | |
| <i>Scabiosa</i> | | | 99,40 | | | | |
| <i>Symphytum</i> | | | | | | 1,45 | |
| <i>Trifolium</i> | | | | 4,08 | | | |

In this study, dry matter was between 71.47-81.38%, protein 17.5-26.0%, fat 5.8-10.95% and ash content between 2-2.44% (Table 2). Chestnut pollen was the

monofloral pollen with the highest protein content (23.5%). It can be said that the fat composition of mixed pollen is higher than monofloral pollen.

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Table 2. Chemical analysis pollen samples (%)

Çizelge 2. Çalışılan örneklerin kimyasal analiz ortalama ve standart sapma sonuçları (%)

| Sample | Dry matter | Protein | Fat | Ash |
|--------|--------------|--------------|--------------|-------------|
| 1 | 71.470±0.028 | 19.000±1.414 | 5.847±0.953 | 2.163±0.185 |
| 2 | 78.480±0.212 | 23.500±0.707 | 8.596±1.063 | 2.209±0.004 |
| 3 | 79.480±0.000 | 18.500±0.707 | 6.420±0.364 | 2.287±0.022 |
| 4 | 76.595±0.162 | 20.500±0.707 | 10.148±0.062 | 2.448±0.112 |
| 5 | 72.685±0.063 | 17.500±0.707 | 7.486±0.487 | 2.025±0.134 |
| 6 | 81.380±0.430 | 17.500±0.707 | 10.951±1.330 | 2.287±0.358 |
| 7 | 73.065±0.374 | 26.000±0.000 | 7.937±0.929 | 2.129±0.038 |

mean ± standard deviation

The most abundant minerals in the pollen samples in our study are potassium, calcium, magnesium, silicon, iron and sodium (Table 3). It was

determined that *Scabiosa* and *Salix* pollen were rich in magnesium, while *Castanea sativa* pollen was rich in iron, manganese and sodium elements (Table 3).

Table 3. Mineral content profile of honeybee pollen pellets (mg/kg)

Çizelge 3. Bal Arısı Polenlerinin Mineral Madde Profili (mg/kg)

| Mineral composition | Pollen samples | | |
|---------------------|-------------------|------------------|-------------------|
| | 1 | 2 | 3 |
| Calcium (Ca) | 413.871 ± 3.725 | 363.390 ± 7.926 | 358.020 ± 10.382 |
| Potassium (K) | 4470.637 ± 40.075 | 4644.076 ± 0.0 | 4751.461 ± 86.225 |
| Magnesium (Mg) | 910.500 ± 21.642 | 440.501 ± 16.346 | 990.769 ± 8.459 |
| Sodium (Na) | 17.435 ± 0.571 | 41.017 ± 1.179 | 23.432 ± 0.396 |
| Boron (B) | 9.141 ± 0.176 | 8.359 ± 0.176 | 18.909 ± 0.418 |
| Iron (Fe) | 50.877 ± 17.234 | 74.818 ± 1.918 | 55.843 ± 2.484 |
| Aluminium (Al) | 32.490 ± 1.395 | 48.425 ± 2.111 | 79.690 ± 0.709 |
| Silicium (Si) | 288.192 ± 11.987 | 388.754 ± 13.186 | 501.398 ± 23.981 |
| Vanadium (V) | 0.136 ± 0.054 | 0.329 ± 0.024 | 0.664 ± 0.093 |
| Chrome (Cr) | 1.049 ± 0.048 | 1.553 ± 0.070 | 2.360 ± 0.103 |
| Manganese (Mn) | 19.568 ± 0.812 | 79.122 ± 5.693 | 8.883 ± 0.029 |
| Cobalt (Co) | 0.160 ± 0.077 | 0.279 ± 0.074 | 0.300 ± 0.074 |
| Nickel (Ni) | 0.184 ± 0.059 | 4.277 ± 0.115 | 1.560 ± 0.060 |
| Copper (Cu) | 5.698 ± 0.070 | 8.943 ± 0.409 | 10.416 ± 0.139 |
| Zinc (Zn) | 23.882 ± 0.603 | 31.260 ± 1.098 | 20.236 ± 1.041 |
| Selenium (Se) | 0.019 ± 0.030 | 0.021 ± 0.029 | 0.033 ± 0.025 |
| Strontium (Sr) | 2.670 ± 0.054 | 0.475 ± 0.014 | 0.646 ± 0.271 |
| Molybdenum (Mo) | 0.085 ± 0.034 | 0.077 ± 0.082 | 0.125 ± 0.030 |
| Silver (Ag) | 0.038 ± 0.007 | 0.019 ± 0.002 | 0.043 ± 0.003 |
| Thallium (Tl) | 0.003 ± 0.000 | 0.016 ± 0.000 | 0.014 ± 0.000 |
| Antimony (Sb) | 0.016 ± 0.001 | 0.051 ± 0.002 | 0.017 ± 0.010 |
| Barium (Ba) | 1.247 ± 0.040 | 2.222 ± 0.873 | 1.095 ± 0.048 |
| Beryllium (Be) | 0.012 ± 0.003 | 0.019 ± 0.005 | 0.027 ± 0.007 |
| Arsenic (As) | 0.020 ± 0.016 | 0.029 ± 0.024 | 0.108 ± 0.031 |
| Lead (Pb) | 0.034 ± 0.011 | 0.050 ± 0.009 | 0.455 ± 0.019 |
| Cadmium (Cd) | 0.050 ± 0.017 | 0.017 ± 0.003 | 0.012 ± 0.009 |

*mean ± standard deviation (n:3)

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Table 3- Continuation Mineral content profile of honeybee pollen pellets (mg/kg)

Çizelge 3-Devamı. Bal Arısı Polenlerinin Mineral Madde Profili (mg/kg)

| Mineral composition | Pollen samples | | | |
|------------------------|------------------|-------------------|-------------------|------------------|
| | 4 | 5 | 6 | 7 |
| <i>Calcium (Ca)</i> | 540.314 ± 11.359 | 241.489 ± 6.053 | 343.457 ± 8.752 | 431.849 ± 9.089 |
| <i>Potassium (K)</i> | 4690.504 ± 0.890 | 4633.516 ± 27.419 | 4769.771 ± 22.986 | 4599.277 ± 0.0 |
| <i>Magnesium (Mg)</i> | 906.868 ± 14.109 | 498.734 ± 10.497 | 566.264 ± 21.951 | 849.189 ± 8.752 |
| <i>Sodium (Na)</i> | 64.653 ± 1.212 | 22.443 ± 0.200 | 16.925 ± 0.563 | 15.830 ± 0.433 |
| <i>Boron (B)</i> | 32.269 ± 1.117 | 14.431 ± 0.345 | 12.148 ± 0.247 | 10.967 ± 0.193 |
| <i>Iron (Fe)</i> | 39.263 ± 1.105 | 47.036 ± 0.634 | 42.773 ± 1.353 | 50.877 ± 1.859 |
| <i>Aluminium (Al)</i> | 33.281 ± 0.957 | 21.254 ± 1.381 | 56.729 ± 7.101 | 8.993 ± 0.215 |
| <i>Silicium (Si)</i> | 447.230 ± 23.279 | 360.060 ± 7.221 | 546.316 ± 33.621 | 355.795 ± 19.982 |
| <i>Vanadium (V)</i> | 0.287 ± 0.010 | 0.241 ± 0.005 | 0.268 ± 0.018 | 0.045 ± 0.001 |
| <i>Chrome (Cr)</i> | 1.681 ± 0.064 | 1.314 ± 0.069 | 1.761 ± 0.078 | 1.605 ± 0.043 |
| <i>Manganese (Mn)</i> | 14.802 ± 0.228 | 12.201 ± 0.288 | 70.357 ± 0.715 | 16.984 ± 0.274 |
| <i>Cobalt (Co)</i> | 0.111 ± 0.010 | 0.343 ± 0.035 | 0.168 ± 0.018 | 0.165 ± 0.054 |
| <i>Nickel (Ni)</i> | 0.556 ± 0.225 | 1.350 ± 0.027 | 0.409 ± 0.018 | 0.515 ± 0.013 |
| <i>Copper (Cu)</i> | 7.665 ± 0.141 | 7.032 ± 3.858 | 6.460 ± 0.095 | 10.577 ± 0.818 |
| <i>Zinc (Zn)</i> | 20.875 ± 1.156 | 16.850 ± 0.020 | 20.013 ± 1,415 | 23.132 ± 0.610 |
| <i>Selenium (Se)</i> | 0.024 ± 0.008 | 0.016 ± 0.035 | 0.014 ± 0.009 | 0.020 ± 0.060 |
| <i>Strontium (Sr)</i> | 0.965 ± 0.031 | 0.602 ± 0.025 | 0.486 ± 0.020 | 0.526 ± 0.279 |
| <i>Molybdenum (Mo)</i> | 0.460 ± 0.015 | 0.261 ± 0.128 | 0.166 ± 0.053 | 0.218 ± 0.077 |
| <i>Silver (Ag)</i> | 0.030 ± 0.006 | 0.025 ± 0.006 | 0.051 ± 0.004 | 0.028 ± 0.005 |
| <i>Thallium (Tl)</i> | 0.062 ± 0.002 | 0.009 ± 0.000 | 0.010 ± 0.000 | 0.006 ± 0.000 |
| <i>Antimony (Sb)</i> | 0.084 ± 0.003 | 0.030 ± 0.002 | 0.0243 ± 0.001 | 0.007 ± 0.001 |
| <i>Barium (Ba)</i> | 1.173 ± 0.160 | 0.677 ± 0.220 | 1.509 ± 0.051 | 0.416 ± 0.010 |
| <i>Beryllium (Be)</i> | 0.018 ± 0.004 | 0.016 ± 0.001 | 0.023 ± 0.007 | 0.010 ± 0.003 |
| Arsenic (As) | 0.087 ± 0.059 | 0.039 ± 0.023 | 0.025 ± 0.003 | 0.015 ± 0.016 |
| Lead (Pb) | 0.119 ± 0.034 | 0.046 ± 0.013 | 0.050 ± 0.012 | 0.015 ± 0.003 |
| Cadmium (Cd) | 0.010 ± 0.005 | 0.027 ± 0.011 | 0.017 ± 0.003 | 0.042 ± 0.002 |

*mean±standard deviation (n:3)

In the pollen samples of our study, the highest amount of heavy metals arsenic (0.108 mg/kg) and lead (0.455 mg/kg) were detected in the 3rd sample which consists of 99.40% of *Scabiosa* pollens, and the highest amount of cadmium (0.050 mg/kg) was detected in the 1st sample which consists of 94.17% *Salix* pollens.

DISCUSSION

In our study, moisture content varied between 19-27%, protein 17.5-26.0%, fat between 5.8-10.95% and ash content between 2-2.44% (Table 2). Similarly, moisture content was found to be 16.9-31%, ash 1.34-2.81%, protein 13.16-24.14% and fat 1.33%-5.47% in BP samples from Romania (Spulber

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et al. 2018). In another study conducted in Brazil, it was determined that the moisture content was between 3.45-4.85%, protein was between 18.55-22.6%, and fat was between 4.80-5.07% (Carpes et al. 2009). In the pollen samples collected throughout the year in eastern Saudi Arabia, the composition was as follows; 20.16% spring pollen, 19.27% summer pollen, 18.02% autumn pollen, and 20.14% winter pollen (Al Kahtani et al., 2020). In Brazil physico-chemical analyzes of two pollen samples were done. For both pollen, protein content (24.8 ± 2.4 g 100 g⁻¹), total sugar content (36.2 ± 1.1 g 100 g⁻¹), lipid content (4.0 ± 0.3 g 100 g⁻¹), and ash (2.6 ± 0.05 g 100 g⁻¹) were determined within the limits, however, it was determined that moisture levels (6.6 ± 2.2 g 100 g⁻¹) of both samples were not in accordance with the values recommended by the laws (<4 g 100 g⁻¹) (Lorini et al., 2020). In another study, eight monofloral BP samples were collected from different apiaries in Morocco. Botanical origins of BP samples were determined using scanning electron microscopy (SEM), and physicochemical parameters (pH, moisture, ash and mineral

contents) were investigated. It was shown that pH was between 4.19 ± 0.17 and 4.82 ± 0.36 , humidity was between 10.7 ± 0.04 and 26.8 ± 0.01 , and ash content was between $1.81\% \pm 0.10$ and 4.22 ± 0.08 for the samples. The protein content has been reported to be between 19.86 ± 0.36 mg/100 g BP, and 30.32 ± 0.12 mg/100 g BP (Asmae et al., 2021).

It was demonstrated that BP samples were rich in mineral elements. Potassium, calcium, magnesium, silicon, iron, and sodium were the most abundant mineral elements. Pollen sources in the present study consist of a significant amount of silicium mineral distinct from those reported in other studies (Szczesna 2007b; Carpes et al. 2009). The samples with monotypic *Salix* and *Scabiosa* pollens were found to be magnesium rich, whereas the ones with monotypic *Castanea sativa* pollen were rich in iron, manganese, sodium, and zinc composition. It can be foreseen that all pollen samples have characteristic mineral composition which could be seen in heatmap (Figure 1) and PCA plots (Figure 2).

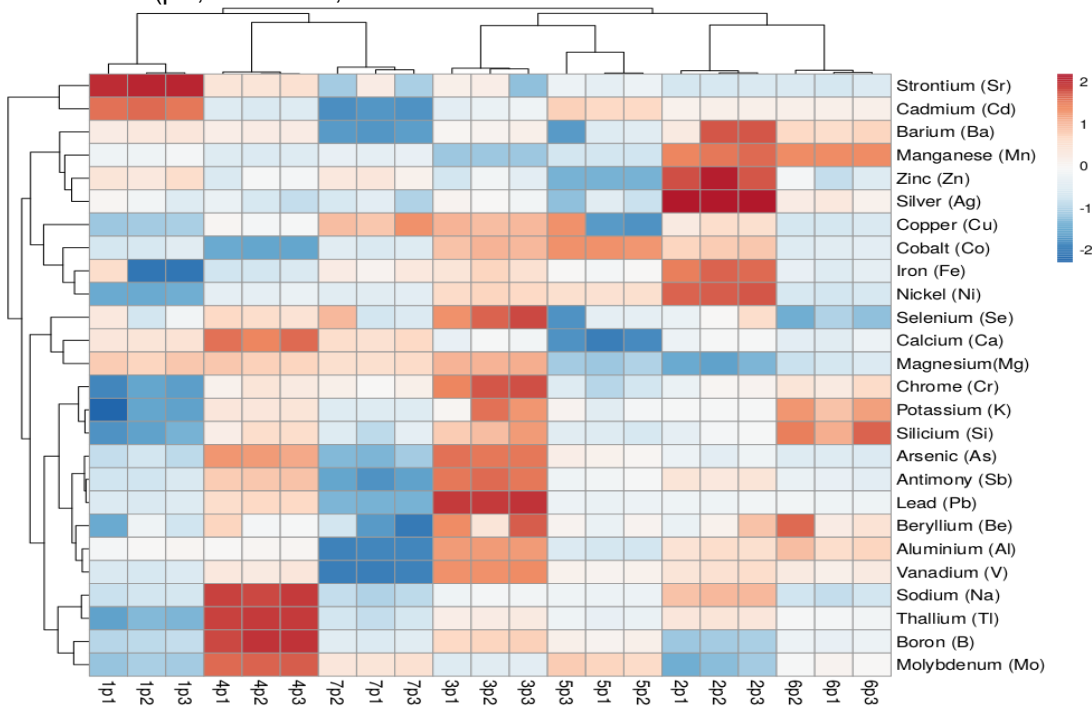


Figure 1. Heatmap plot of mineral content in pollen samples

Şekil 1. Polen örneklerindeki mineral maddelerinin Heatmap haritası

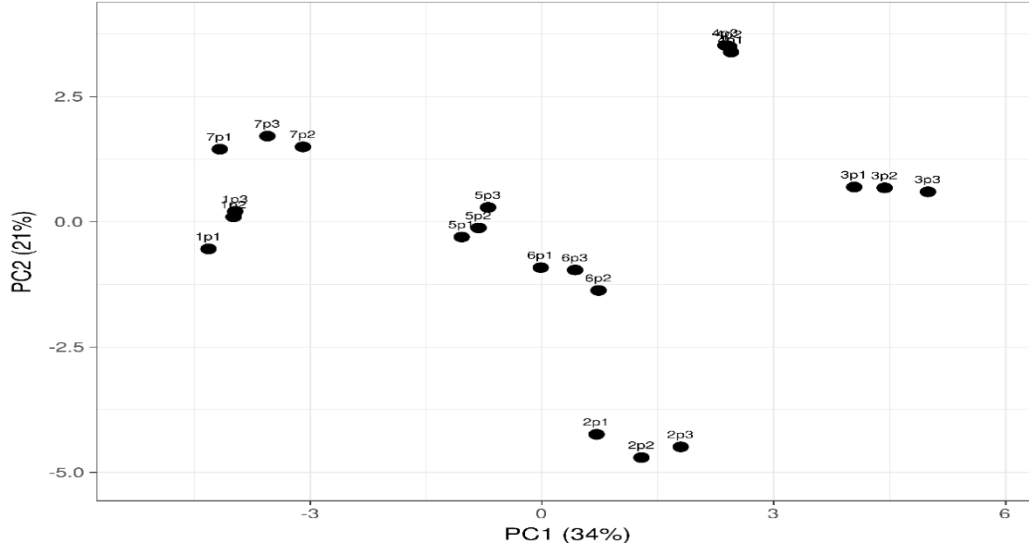


Figure 2. PCA plot according to the mineral content in pollen samples

Şekil 2. Polen örneklerindeki mineral maddelerinin temel bileşenler analizi (PCA)

In BP samples collected from the Northeast Anatolia Region the chemical composition was determined as 8.60 ± 0.43 mg/kg of copper, 35.30 ± 2.13 mg/kg of zinc, 1113.33 ± 61.00 mg/kg of magnesium, and 1034.00 ± 53.86 of calcium. These amounts were stated to be at a level that would contribute to the daily mineral need (Uçar et al. 2018). It has been reported that the most abundant minerals in BP samples from Morocco were potassium and magnesium, and heavy metals were not detected (Asmae et al., 2021). In the study carried out with the pollen samples of Turkey and Russia, it was determined that there was a statistical difference in the composition of the minerals between the regions where the pollen was collected. It has been reported that Russian pollen samples are richer in potassium, while Turkey's pollen samples are rich in potassium, phosphorus, magnesium, and iron elements (Özcan et al. 2019). In a study conducted in Romania, *Prunus spp.* pollen contained the highest Fe (150.9 ± 1.11 mg/kg), while the highest Mg level (1505 ± 1.43 mg/kg) was detected in *Brassica* pollen. The other pollen samples in the study were found to be consisting of 1980-4284 mg/kg of potassium, 474.3-1505 mg/kg of magnesium, 1155-4335 mg/kg of calcium, 21.7-58 mg/kg of iron, and 20.2-59.5 mg/kg of zinc (Spulber et al. 2018). In another study with pollen samples from China, South Korea, and

Poland, it is reported that there are differences in mineral compositions between countries. For example, 762 mg/kg of calcium, 26.3 mg/kg of manganese, 1305 mg/kg of magnesium, 36.8 mg/kg of zinc, 9.3 mg/kg of copper, 65.4 mg/kg of iron, 3903. mg/kg of potassium, and 739 mg/kg of sodium was reported in Polish samples (Szczesna 2007b). In BP collected in Brazil, phosphorus, potassium, calcium and magnesium minerals were found to be at the highest levels, respectively (Carpes et al. 2009).

In the research carried out on pollen samples from Turkey and Russia, Russian samples had 0.11-0.19 mg/kg of cadmium, and 0.18-0.64 mg/kg of lead, whereas 0.04-0.14 mg/kg of cadmium and 0.25-0.60 mg/kg of lead was found in Turkish samples (Özcan et al. 2019). In fresh BP from Bulgaria, lead was determined as 0.467-0.483 mg/kg and cadmium as 0.022-0.029 mg/kg (Dinko and Stratev 2016). In Serbia, cadmium was detected as 0.004-0.125 mg/kg in pollen samples (Kostić et al. 2015). According to Campos et al. (2008), the highest amount of Pb and Cd in fresh BP should not exceed 0.03 mg/kg and 0.5 mg/kg, respectively. In addition, the composition of other minerals should be as follows; 4000-20000 mg/kg of potassium, 200-3000 mg/kg of magnesium, 200-3000 mg/kg of calcium,

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11-170 mg/kg of iron, 30-250 mg/kg of zinc, 2-16 mg/kg of copper, and 20-110 mg/kg of manganese.

It is known that the chemical composition of pollen sources varies according to the type of plant from which the pollen is produced. Similar to our results, it is reported that BPs of different origins collected throughout Serbia contain potassium, calcium and magnesium as major elements, and are also extremely rich in iron and zinc, which are very important nutrients. It has been reported that the mineral composition of BP is more dependent on the type of pollen producing plant than its geographical origin (Kostić et al. 2015). Moreover, in the study conducted in Brazil, it was reported that the mineral composition changed according to the place and year of pollen production (Morgano et al. 2012).

Conclusion

The biochemical content of BP varies depending on the plant diversity, the period and place it was produced (Taha and Al-Kahtani 2020), and the effect of the season/period in which the pollen was taken on pollen diversity and yield should also be considered. It is important for the producer to define the contents of BP, which is sold as a commercial product, and to create a demand according to its quality.

BP produced in Turkey can be considered as a good mineral source, especially in terms of Ca, K, Mg, Fe, Si and trace elements. Recently, BP has become a prominent product due to the increasing interest and consumer demand for natural products. Both its components and its use in apitherapy make pollen an important product. However, due to the increase in environmental pollution in recent years, BP taken from apiaries close to residential areas, industrial facilities and highways should be controlled in terms of heavy metals. Considering that BP can be consumed by risky groups (the elderly, pregnant women, patients), both producers and consumers should be careful about health problems that may arise.

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