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Characterization of Chemical Composition of Bee Pollen in China

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ABSTRACT: Bee pollen has been praised for its good nutrition and therapeutic values. China is the largest producer in the world. Twelve common varieties of monofloral bee pollen collected from China's main producing regions were selected for nutritional composition analysis, including proximate contents, dietary fibers, amino acid distribution, fatty acid composition, and mineral elements. The proximate compositions mostly met the specifications regulating pollen load quality of China. Proline and glutamic acids were found to be the predominant amino acids in the form of both total amino and free amino acids. Lysine was the relative limiting amino acid. The percentage of total essential amino acids (TEAA) to total amino acids (TAA) reached the nutrition recommendation of the Food and Agricultural Organization (FAO). The major fatty acids, presented as mean values, were C18:3 (25.1%), C16:0 (19.6%), C18:1 (17.3%), C18:2 (8.78%), C22:0 (4.07%), and C18:0 (2.96%) acids. The proportions of C18:3 were generally higher than those of C18:2, and the ratio of total unsaturated fatty acids (TUS) to total saturated fatty acids (TS) was >1.0, except for *Nelumbo nucifera* Gaertn. pollen for the characteristic absence of C18:3 acids. High levels of beneficial elements such as K, Ca, Mg, Zn, Fe, Mn. and Cu were observed in pollen samples. The contents of detrimental trace elements of Cd, Pb, and Hg were primarily lower or not detected. However, more attention should be paid to a large amount of Al, with a concentration of >100 mg/kg DW in most samples. There were some significant differences between samples. On the whole, the Chinese bee pollen was evaluated as a good complement to diet.

KEYWORDS: *Chinese bee pollen, chemical composition analysis, nutrition, amino acids, fatty acids, minerals*

INTRODUCTION

Bee pollen is an apicultural product of male reproductive cells collected by the honeybee *Apis mellifera* from flower stamens of gymnosperm and angiosperm used for feeding its larvae in the early stage of development and producing royal jelly.[1] Bee pollen contains all nutrients that are necessary for plant growth and development, and it is rich in sugars, proteins, amino acids, lipids, vitamins, minerals, nucleic acids, enzymes, and phenolics.[2,3] Praised for its excellent nutrition and medical values, pollen loads have been daily consumed throughout the world since ancient times and gained increased attention in current years owing to a tendency for natural diet supplementation. Recent research has also shown that bee pollen possesses the therapeutic benefits of improving cardiovascular system, stimulating body immunity, promoting antitumor effects, delaying aging, scavenging free radicals, regulating intestinal functions, and treating prostate disease,[4−8] with different pollens showing different specificities.

Each pollen pellet has a characteristic color, size, morphology, flavor, and composition, specific to the floral species or cultivars.[9] The monofloral pollen pellet maintains organoleptic and biochemical properties similar to those of the original plant, whereas the multifloral pollen has variable properties of more than two original plants. The major variant of composition in bee pollens is the species, which may be affected by differences in gathering area or time.[1] Other variations could be introduced through different processes or storage treatments in commercial production,[10] such as heat-drying, age-related oxidation, ultraviolet (UV) exposure, or irradiation sterilization. In addition, differences in chemical properties might result from methods of extraction in conjunction with corresponding analysis.

With its extensive territory, diversified flora, favorable climate throughout the year, and about 300,000 beekeepers from south to north, China currently possesses around 8.2 million bee hives, being the largest apiculture producer, consumer, and exporter in the world (including bee pollen),[11] followed by Spain, Australia, Argentina, and Brazil.[12] Switzerland (Swiss Food Manual: Pollen Bienenprodukte, BAG-Swiss Federal Office for Public Health), Argentina, Brazil (Instrucao Normativa n.3, de 19 de Janeiro de 2001), Bulgaria (Bulgarian standard 2567111-91), Poland (PN-R-78893 "Obnoza pylkowe"-Polish legislation for bee-pollen),[13] and China (NY 5137-2002 and GB/T 19330-2008) are countries that have already established national quality standards concerning bee pollen.[14,15]

The chemical compositions of pollen have gained worldwide research interest covering broad areas, ranging from plant physiology to biochemistry and even material science.[16] The generic pollen compositions of diverse bee pollens were studied in Spain,[17,18] Australia,[19−21] Portugal,[12] Brazil,[2,22,23] and South Africa.[24] The results indicated that it had appreciable differences among compositions of the pollen from various samples, regions, or countries. On the basis of the experience and studies from different countries (except China), a general global quality criterion for pollen was proposed by Campos et al.[13] To date, only a few studies on partial chemical constituents of certain Chinese pollen have been conducted, usually as an example of *Brassica napus* L.[25,26] There was little information related to the

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Figure 1. Collection locations of bee pollen samples in the map of China.

Table 1. Species, Producing Region, Time of Collection, Purity, and Color of Pollen Samples

species	abbrev	producing region	time of collection	purity (%)	color
Brassica napus L.	BNL	Menyuan, Qinhai	July	95.6	yellow
Citrullus lanatus L.	CLL	Fuzhou, Jiangxi	May	92.5	bright yellow
Camellia japonica L.	CJL	Kunming, Yunnan	March	91.6	bright yellow
Dendranthema indicum L.	DIL	Liuan, Anhui	October	90.2	bright yellow
Fagopyrum esculentum L.	FEL	Dingbian, Shanxi	September	92.1	dark yellow
Helianthus annuus L.	HAL	Gannan, Heilongjiang	August	94.6	orange
Nelumbo nucifera Gaertn.	NNG	Jinzhou, Hubei	July	93.8	light yellow
Papaver rhoeas L.	PRL	Wuwei, Gansu	May	93.4	yellowish brown
Rosa rugosa	RR	Xiangfan, Hubei	May	90.6	faint yellow
Schisandra chinensis	SC	Linhe, Neimenggu	May	91.6	deep yellow
Vicia faba L.	VFL	Wushan, Gansu	April	92.7	blackish green
Zea mays L.	ZML	Changzhi, Shanxi	August	90.5	yellowish brown

comprehensive composition of bee pollen samples from various species in China.

Because one of the fundamental aspects of quality that affects its commercial value is the botanical and geographical declaration of pollen origin, the full characterization of pollen loads of diverse origins still appears to be a sound area of research. The present research oriented toward the chemical characterization of 12 common species of Chinese monofloral bee pollen might clarify the basic features of their chemical components. The results would shed some light on the influence of the floral origin on the chemical composition of pollen pellets, increase the commercial value of products, contribute to the choice of criteria or establishment of a global quality standard of pollen, and provide clues to the pollen botanical taxa.

■ MATERIALS AND METHODS

Samples. Twelve samples of pollen pellets collected by *A. mellifera* L., weighing 1.0 kg each and vacuum packed in polyethylene bags with desiccants, were purchased from Zhenzhou Kerun Apiculture Co., Ltd. They were labeled with botanical origins, collected areas in China's main producing regions (marked in Figure 1), and flower season of 2010 (sample information as listed in Table 1). Prior to commercialization, samples were dried by the company to a moisture content of <8% by a ventilation oven (46 °C, 3−4 h) and were preserved at 4 °C by producers. On arrival at our laboratory, the samples were refined manually on the basis of color and morpha according to the literature.[27] Finally, the samples were automatically pulverized for 30 s, sieved through a 40 mesh (425 μm) sieve, repacked, and frozen at −18 °C for further chemical analysis in 3 months.

Standards, Chemicals, and Reagents. Heat stable α-amylase (A 3306), protease (P 3910), amyloglucosidase (A 9913), acid-washed Celite (C 8656), all L-amino acid standards, and fatty acid methyl ester (FAME) reference standard mixture (fatty acids from C8 to C22) were purchased from Sigma (St. Louis, MO, USA). All single-element stock solutions with a concentration of 1000 mg/L were purchased from the national standard center of China. Methanol, *n*-hexane, acetone, citrate buffer (pure for analysis) were purchased from

E. Merck (Darmstadt, Germany). Water was purified by a Milli-Qplus system from Millipore (Milford, MA, USA). All other reagents of analytical grade were obtained from Sinopharm Chemical Regent Co., Ltd. (Shanghai, China).

Melissopalynological Analysis. One gram of each hand-sorted sample was considered to be representative for botanical origin, which was dispersed in 25 mL of water and gently sonicated for 5 min. Then a little of the solution was submitted to the analysis of pollen grain identification and pollen grain counts under an optical microscope (BX 40, Olympus, Japan) with 450× magnification, according to the ref 30. The pollen grain counts were done by calculating its mean percentage in the main sample. The purities of all sorted samples were demonstrated at a rate >98%.

Chemical Composition Analysis. *Proximate and Fiber Composition Analysis.* Proximate analysis, including crude fat, crude protein, and ash, was performed according to Association of Official Analytical Chemists (AOAC) methods 991.36, 928.08, and 920.153, respectively.[28] Moisture content was determined by drying at 60 °C for 48 h using a vacuum oven, cooling in a desiccator, and weighing until a constant weight was obtained. Carbohydrates were calculated by difference: carbohydrates = 100 − (g fat + g protein + g ash). Total dietary fiber (TDF), soluble dietary fiber (SDF), and insoluble dietary fiber (IDF) were evaluated by AOAC method 991.43. The results were expressed in grams per 100 g of dry weight except for moisture (expressed in grams per 100 g of fresh weight). Total energy was calculated according to the following equation: energy (kcal) = 4 × (g of protein + g of carbohydrate) + 9 × (g of lipid).[29]

Amino Acid Analysis. For total amino acid analysis, sample (50−100 mg) defatted with petroleum ether (40−60 °C) was hydrolyzed with 10 mL of 6 M HCl in 20 mL vacuum sealed ampules at 110 °C for 24 h according to the method described by AOAC,[30] except that tryptophan was determined by hydrolyzing the sample with 4 M LiOH.[31] The cooled and filtered hydrolysate was dried in a TVE-1000 vacuum desiccator (EYELA, Japan) at 50 °C and redissolved in citrate buffer (pH 4.3). Aliquots of the solution were analyzed by an amino acid analyzer (model S-433D, Sykam, German). Amino acids (AA) were quantified by comparing retention times and peak areas with those of the standard curves. The results were reported as grams of amino acid per 100 g of dry sample.

For free amino acid analysis, the free amino acid extraction was performed with a 3% 5-sulfosalicylic acid (SSA) solution (1 g/20 mL) on previously defatted powdered samples, according to the method of ref 32 with slight modifications, filtered with 0.22 μM membranes, and then analyzed by an amino acid analyzer.

Fatty Acid Analysis. Determination of fatty acid was accomplished through the quantification of their methyl esters (FAMEs) by GC in the extracted fat from pollens. Fat was obtained by Soxhlet extracting 10 g of dry sample with *n*-hexane for 8 h in triplicate. The solvent was removed in a rotary evaporator at 40 °C. Afterward, the FAMEs were prepared by diluting an aliquot of oil in *n*-hexane (1:10, w/w) and adding 50 μL of methanolic 2 M KOH.[33] After 30 min at room temperature, the upper layer was collected for GC analysis. An Agilent Technologies 6890 series gas chromatograph was used, which was equipped with a split−splitless injector, a flame ionization detection (FID), a 7693 autosampler, and an Agilent ChemStation software system (version A.10.02). Separation was performed on a CP-Sil 88 fused capillary column (100 m × 0.25 mm i.d., Varian Inc., Mississauga, ON, Canada). The injector and detector temperatures were both set at 250 °C; H_2 served as the carrier gas at a flow of 1 mL/min. The column was initially operated at 45 °C for 4 min and then temperature-programmed at 13 °C/min to 175 °C, held for 27 min, increased at 4 °C/min to 215 °C, and finally held for 35 min. The split ratio was 1:50, and the injected volume was 1.0 μL.[34] Identification and quantification of the FAMEs were achieved by comparing the relative retention times and peak areas with those of the standard FAME mixture, and the results were expressed as percent of total FAMEs.

Element Analysis. Twenty elements were determined by inductively coupled argon plasma-atomic emission spectrometry (ICP-AES) after wet mineralization. Each variety (1.0 g) was digested with 10 mL of concentrate nitric acid (65%) and 1 mL of hydrogen peroxide (30%) in a closed polytetrafluoroethylene-stoppered vessel (PDS-6; Loftfields Analytische Lösungen LAL, Neu Eichenberg, Germany) for 8 h at 170 °C.[35] After cooling, the extract was quantitatively transferred into 50 mL volumetric flasks and made up to the mark. It was then analyzed by ICP-AES (model IRIS Intrepid, Thermo Jarrell Ash, USA) with the following conditions: plasma gas flow rate (Ar), 16 L/min; auxiliary flux, 1.0 L/min; nebulized pressure, 25 psi; sample flush time, 30 s; delay time, 30 s; solution uptake rate, 1.60 mL/min; radio frequency, 27.12 MHz; power, 1.05 kW. The absorbance data for each element was recorded at the following wavelengths: Al (λ = 308.2 nm), As (λ = 189.0 nm), Ca (λ = 317.9 nm), Cd (λ = 228.8 nm), Co (λ = 228.6 nm), Cr (λ = 283.5 nm), Cu (λ = 324.7 nm), Fe (λ = 259.9 nm), Ge (λ = 265.1 nm), Hg (λ = 184.9 nm), K (λ = 766.4 nm), Mg (λ = 279.5 nm), Mn (λ = 257.6 nm), Mo (λ = 202.0 nm), Na (λ = 589.5 nm), Ni (λ = 231.6 nm), P (λ = 213.6 nm), Pb (λ = 220.3 nm), Se (λ = 196.0 nm), and Zn (λ = 213.8 nm). The results were expressed in micrograms per gram on a dry weight basis.

Statistical Analysis. All analyses were carried out in triplicate. Results were expressed as the mean value ± standard deviation (SD). Differences were tested with analysis of variance (ANOVA) followed by multiple-comparison test (Tukey HSD). Whenever necessary, data were transformed to satisfy normal distribution and homoscedasticity requirements. If transformed data could not meet these assumptions, differences were analyzed with nonparametric analysis of variance (Kruskall−Wallis) followed by a nonparametric multiple-comparison test (Mann−Whitney). All statistical analyses were tested at the 0.05 level of probability with the software SPSS 16.0.

RESULTS AND DISCUSSION

The results of proximate and fiber composition of samples are shown in Table 2. The differences observed in the contents of proximate components seemed to be related to pollen varieties. The moisture content in the samples was in a range of 1.82−7.33% and fell into the range of GB/T 19330-2003 (≤10%)[16] and NY 5137-2002 (≤8%) standards.[17] All other data were expressed in dry weight (DW). Carbohydrates, calculated by difference, were the predominant macronutrient and ranged from 59.43% in CJL to 75.65% in ZML. The values were 13−55% higher than those references summarized by Campos et al.[15] Protein was the second most abundant component, and it varied between 14.26% in FEL and 28.95% in CJL with a mean value of 21.56%, in accordance with standard NY 5137-2002 (≥15%), except for DIL (14.86%) and FEL (14.26%). It could be observed that the contents of protein were >20%, higher than pollen samples collected in first half year, except BNL. Fat fell within a range of 0.66% in VFL to 10.79% in HAL. Ash content of the pollen ranged from 1.70% in HAL to 5.01% in FEL. Only FE (5.01%) and NNG (4.18%) slightly exceeded the criterion of NY 5137−2002 (≤4%). The energy values of the pollen, which were calculated by using Atwater's constant, ranged from 380 to 486 kcal/100 g with slight difference. Therefore, pollen can be used in low-calorie diets for due to its low fat and energy contents.

As seen in Table 2, pollen is a good source of total dietary fiber (TDF), but similarly to the research of Bonvehí et al.,[17] most of the TDF in pollen was insoluble dietary fiber (IDF). The levels of TDF varied between 17.60% in CLL and 31.26% in NNG, a little higher than other species having a range of 10.6−15.9%,[17] and the SDF values ranged from 0.86% in CLL to 5.92% in BNL. The values of SDF/IDF in all samples were all lower than an appropriate ratio of 1:2 needed for a healthy diet.[36] Major constituents of IDF also showed that polysaccharides, that is, cellulose and callose, were probably the most important fraction of the DF.

Table 2. Proximate and Fiber Composition of Pollen Samples (Grams per 100 g)[a]

component	BNL	CLL	CJL	DIL	FEL	HAL	NNG	PR	RR	SCL	VFL	ZML
	pollen sample											
proximate												
moisture	6.59 ± 0.13 c	2.76 ± 0.02 h	6.92 ± 0.14 g	7.52 ± 0.36 a	4.22 ± 0.26 g	4.57 ± 0.19 g	5.98 ± 0.26 d	7.33 ± 0.31 a	1.82 ± 0.10 i	7.17 ± 0.22 ab	5.22 ± 0.15 f	5.59 ± 0.23 e
ash	3.53 ± 0.10 c	2.81 ± 0.08 d	3.37 ± 0.10 c	1.67 ± 0.06 g	5.01 ± 0.12 a	1.70 ± 0.11 g	4.18 ± 0.21 b	2.69 ± 0.09 de	2.57 ± 0.05 ef	3.41 ± 0.10 c	4.33 ± 0.11 b	2.48 ± 0.10 f
fat	6.56 ± 0.32 a	3.60 ± 0.20 d	5.25 ± 0.27 b	4.01 ± 0.18 c	5.15 ± 0.30 b	1.52 ± 0.24 f	5.22 ± 0.21 b	2.17 ± 0.10 e	1.36 ± 0.09 f	2.36 ± 0.13 e	0.66 ± 0.23 g	4.04 ± 0.11 c
protein	27.27 ± 0.72 b	20.68 ± 1.09 e	28.95 ± 0.97 a	14.86 ± 0.42 gh	14.26 ± 0.55 h	15.34 ± 0.38 g	16.96 ± 0.58 f	24.99 ± 0.53 c	22.94 ± 0.61 d	27.40 ± 0.65 b	27.85 ± 0.41 ab	17.89 ± 0.41 f
carbohydrates	60.34 ± 0.45 e	72.21 ± 0.72 bc	59.43 ± 0.68 e	77.82 ± 0.37 a	74.51 ± 0.42 ab	72.02 ± 0.28 c	71.59 ± 0.26 c	70.27 ± 0.26 cd	73.05 ± 0.38 ab	67.01 ± 0.39 d	67.24 ± 0.37 d	75.65 ± 0.25 a
energy (kcal/100 g)	410 ± 7 b	406 ± 5 bc	401 ± 5 bc	406 ± 6 bc	401 ± 4 bc	446 ± 8 a	400 ± 5 bc	398 ± 6 bc	397 ± 5 bc	397 ± 4 bc	380 ± 5 c	411 ± 6 b
fiber												
IDF	13.39 ± 1.67 e	16.28 ± 0.88 d	17.06 ± 1.29 d	18.04 ± 1.85 c	18.07 ± 1.24 c	17.33 ± 0.75 d	30.12 ± 2.23 a	16.38 ± 0.96 d	19.82 ± 1.66 b	19.89 ± 0.95 b	16.86 ± 0.86 d	16.25 ± 0.88 d
SDF	5.92 ± 0.46 a	0.86 ± 0.10 i	2.84 ± 0.17 c	4.03 ± 0.31 b	1.17 ± 0.14 h	3.06 ± 0.14 c	0.98 ± 0.25 hi	2.38 ± 0.16 de	1.99 ± 0.09 f	2.03 ± 0.11 ef	1.62 ± 0.09 g	2.46 ± 0.18 d
TDF	18.95 ± 1.92 c	17.60 ± 0.79 d	19.37 ± 1.31 c	21.72 ± 1.75 b	19.00 ± 1.20 c	20.11 ± 1.06 bc	31.26 ± 2.54 a	18.46 ± 1.16 d	21.63 ± 1.58 b	21.50 ± 1.21 b	18.27 ± 0.79 cd	19.01 ± 0.93 c
SDF/IDF	1:2.3	1:18.9	1:6.0	1:4.5	1:15.4	1:5.7	1:30.7	1:6.9	1:10.0	1:9.8	1:10.0	1:6.6

[a]Results are expressed on a dry basis, except for moisture. Means with different letters within the same row are significantly different ($p < 0.05$).

Amino Acid. The profiles of total amino acids and free amino acids are presented in Table 3. The recovery of protein contents ranged from 63.30% (RR) to 95.36% (PRL), with a mean value of 85.68%, in accordance with proximate analysis of crude protein. Proline (mean of 2.33g/100 g DW), glutamic acid (mean of 2.00 g/100 g DW), and aspartic acid (mean of 1.91g/100 g DW) were the predominant amino acids in the pollen. Especially high contents of proline were detected in VFL (5.95 g/100 g DW), SC (4.95 g/100 g DW), and CJL (3.09 g/100 g DW), and the lowest was found in NNG (0.74 g/100 g DW). The contents of total essential amino acids (TEAA) varied between 4.62% (HA) and 11.60% (CJ). The percentages of TEAA to total amino acids (TAA) ranged from 35.18% in SC to 42.78% in NNG, with a mean of 39.40%. It was higher than the 33.9% essential amino acids in the FAO reference protein.[37]

It was observed that lysine was the relatively restrictive amino acid in pollen compared favorably with the FAO/WHO reference pattern (Table 4).[38] The scores of tryptophan all exceeded the recommendation of FAO/WHO except PRL, whereas it appeared to be the first limiting amino acid in Australian pollen from eucalyptus species.[39] In terms of essential amino acid scores, NNG had a better nutritional value, and PRL had a relatively worse value among pollen samples. On the whole, pollen was a good source of high-quality proteins. The results were in accordance with the same species of BNL, FEL, HAL, and ZML,[20] but wide variation was observed in BNL compared with the consequence of HPLC–fluorescence.[25]

The investigation of the free amino acid of the pollen samples is given in Table 5. It was revealed that free cystathionine was absent in all pollen samples. Methionine was only detected in FEL with an average of 8.30 mg/100 g DW. Threonine was found only in BNL, NNG, and VFL. Serine was present in almost all pollen samples except for FEL. Isoleucine was present in almost all pollen samples except for PRL and RR. Proline and glutamic acid were the predominant free amino acids in the pollen, similar to total amino acids. The contents of total free amino acids (TFAA) ranged from 1007 mg/100 g DW in NNG to 6925 mg/100 g DW in SC, less than the average value of 3.2 g/100 g DW in Spanish pollen of 20 cultivars, except of DIL, SC, and VFL. The proportion of proline against TFAA was also less than the mean value of 63.1% in Spanish pollen except of VFL (69.40%).[17] The total free amino acid content and the average of proline against the total free amino acid could be the index of drying process, according to the proposal that a minimum quantity of 2 g/100 g of free amino acid content was suggested to standardize the commercial honeybee-collected pollen in the European market,[17] but our results did not coincide with those inferences.

Fatty Acids. Fatty acids play an essential role in the reproduction, development, and growth of honeybees. Certain fatty acids, such as linoleic, linolenic, myristic, and lauric acids, have bactericidal and antifungal properties that are important for colony hygiene.[40] The fatty acid proportions determined in 12 cultivars of pollen are listed in Table 6. The dominant fatty acids presented as mean values in pollen samples were C18:3 (25.1%), C16:0 (19.6%), C18:1 (17.3%), C18:2 (8.78%), C22:0 (4.07%), and C18:0 (2.96%), together with small amounts of C8:0, C10:0, C12:0, and C14:0. Palmitic acid (C16:0), oleic acid (C18:1), linoleic acid (C18:2), and arachic acid (C20:0) were detected in all specimens, and the concentrations of individual acid varied greatly between

Table 3. Amino Acid Composition of the Pollen Samples (Grams per 100 g Dry Weight)[a]

amino acid	pollen sample BNL	CLL	CJL	DIL	FEL	HAL	NNG	PR	RR	SCL	VFL	ZML
EAA[b]												
threonine	0.97 ± 0.06 a	0.75 ± 0.05 bc	0.99 ± 0.06 a	0.45 ± 0.02 e	0.46 ± 0.01 e	0.43 ± 0.02 e	0.66 ± 0.03 c	0.91 ± 0.05 a	0.55 ± 0.02 d	0.82 ± 0.05 b	0.92 ± 0.03 a	0.57 ± 0.04 cd
valine	1.29 ± 0.08 b	1.19 ± 0.06 b	1.57 ± 0.07 a	0.73 ± 0.04 e	0.70 ± 0.04 e	0.64 ± 0.02 f	1.02 ± 0.05 c	1.41 ± 0.05 a	0.84 ± 0.05 d	1.26 ± 0.06 b	1.37 ± 0.11 ab	0.84 ± 0.04 d
methionine	0.42 ± 0.01 b	0.32 ± 0.02 c	0.62 ± 0.03 a	0.29 ± 0.00 d	0.20 ± 0.01 g	0.12 ± 0.00 h	0.25 ± 0.01 e	0.43 ± 0.01 b	0.25 ± 0.00 e	0.25 ± 0.01 e	0.40 ± 0.03 b	0.22 ± 0.01 f
isoleucine	1.11 ± 0.07 b	1.01 ± 0.03 c	1.33 ± 0.08 a	0.60 ± 0.02 e	0.56 ± 0.02 f	0.51 ± 0.02 g	0.86 ± 0.05 d	1.18 ± 0.06 ab	0.68 ± 0.03 d	1.04 ± 0.05 bc	1.19 ± 0.06 ab	0.65 ± 0.02 d
leucine	1.70 ± 0.11 cd	1.55 ± 0.10 e	2.06 ± 0.09 a	0.89 ± 0.07 i	0.88 ± 0.05 i	0.79 ± 0.05 i	1.25 ± 0.06 f	1.84 ± 0.06 b	1.10 ± 0.06 g	1.62 ± 0.04 de	1.79 ± 0.11 c	1.01 ± 0.06 h
phenylalanine	1.08 ± 0.06 b	0.97 ± 0.04 c	1.30 ± 0.05 a	0.53 ± 0.02 ef	0.53 ± 0.02 ef	0.46 ± 0.02 f	0.79 ± 0.05 d	1.12 ± 0.06 b	0.67 ± 0.01 e	1.11 ± 0.06 b	1.12 ± 0.12 b	0.60 ± 0.04 e
lysine	1.53 ± 0.08 a	1.12 ± 0.05 c	1.55 ± 0.06 a	0.86 ± 0.03 ef	0.72 ± 0.06 f	0.70 ± 0.03 k	0.95 ± 0.04 d	1.35 ± 0.03 b	0.80 ± 0.04 f	1.42 ± 0.05 b	1.59 ± 0.11 a	0.92 ± 0.04 e
tryptophan	1.28 ± 0.04 de	1.82 ± 0.05 b	14.80 ± 0.07 a	1.17 ± 0.08 f	0.77 ± 0.03 jk	0.70 ± 0.02 k	0.94 ± 0.06 hi	1.38 ± 0.01 c	1.03 ± 0.04 gh	1.05 ± 0.00 g	1.25 ± 0.13 ef	0.86 ± 0.05 ij
total EAA	9.38 ± 0.14 b	8.73 ± 0.08 c	11.62 ± 0.15 a	5.52 ± 0.22 f	4.82 ± 0.21 g	4.35 ± 0.08 h	6.72 ± 0.21 d	9.62 ± 0.09 b	5.92 ± 0.15 e	8.57 ± 0.18 c	9.63 ± 0.15 b	5.67 ± 0.20 ef
NEAA[c]												
aspartic acid	2.21 ± 0.06 b	2.57 ± 0.11 a	2.58 ± 0.09 a	1.24 ± 0.06 e	1.24 ± 0.08 e	1.13 ± 0.06 e	2.02 ± 0.09 c	2.36 ± 0.06 b	1.99 ± 0.08 c	2.20 ± 0.08 b	2.30 ± 0.20 b	1.42 ± 0.05 d
serine	1.23 ± 0.07 ab	1.05 ± 0.01 c	1.25 ± 0.00 a	0.63 ± 0.02 e	0.58 ± 0.06 ef	0.51 ± 0.01 f	0.81 ± 0.02 d	1.16 ± 0.06 b	0.73 ± 0.05 d	1.05 ± 0.05 c	1.18 ± 0.09 ab	0.76 ± 0.04 d
glutamic acid	2.50 ± 0.03 c	2.20 ± 0.01 d	2.87 ± 0.10 a	1.29 ± 0.03 h	1.81 ± 0.06 e	1.11 ± 0.03 i	1.77 ± 0.02 d	2.68 ± 0.07 b	1.64 ± 0.05 f	2.50 ± 0.06 c	2.55 ± 0.05 c	1.41 ± 0.03 g
proline	1.57 ± 0.08 f	1.12 ± 0.06 g	3.09 ± 0.16 c	2.01 ± 0.11 e	1.48 ± 0.05 f	0.95 ± 0.04 gh	0.74 ± 0.04 h	2.72 ± 0.10 d	1.18 ± 0.04 g	4.95 ± 0.21 b	5.95 ± 0.35 a	2.21 ± 0.13 e
glycine	1.17 ± 0.05 b	0.95 ± 0.02 e	1.25 ± 0.01 a	0.63 ± 0.03 h	0.55 ± 0.02 i	0.64 ± 0.02 h	0.80 ± 0.04 f	1.15 ± 0.03 b	0.72 ± 0.02 g	1.05 ± 0.02 d	1.10 ± 0.03 c	0.72 ± 0.01 g
alanine	1.25 ± 0.06 bc	1.12 ± 0.07 d	1.45 ± 0.05 a	0.75 ± 0.03 g	0.73 ± 0.04 g	0.70 ± 0.02 g	0.96 ± 0.04 e	1.29 ± 0.05 b	0.87 ± 0.04 f	1.18 ± 0.04 cd	1.29 ± 0.08 b	0.91 ± 0.02 ef
cystine	0.25 ± 0.00 cd	0.27 ± 0.01 b	0.30 ± 0.01 a	0.25 ± 0.00 cd	0.15 ± 0.01 h	0.24 ± 0.01 de	0.23 ± 0.01 ef	0.26 ± 0.01 bc	0.20 ± 0.01 g	0.29 ± 0.02 a	0.22 ± 0.01 f	0.22 ± 0.00 f
tyrosine	0.68 ± 0.04 bc	0.63 ± 0.02 c	0.85 ± 0.03 a	0.35 ± 0.01 f	0.36 ± 0.01 f	0.28 ± 0.01 g	0.51 ± 0.02 d	0.74 ± 0.02 b	0.46 ± 0.01 de	0.69 ± 0.03 bc	0.72 ± 0.05 b	0.42 ± 0.02 e
histidine	0.81 ± 0.05 b	0.70 ± 0.04 c	0.93 ± 0.04 a	0.55 ± 0.03 d	0.42 ± 0.02 e	0.69 ± 0.04 c	0.65 ± 0.02 c	0.91 ± 0.04 a	0.53 ± 0.01 d	0.70 ± 0.05 c	0.72 ± 0.09 c	0.44 ± 0.03 e
arginine	1.40 ± 0.09 b	1.32 ± 0.09 b	2.58 ± 0.17 a	0.83 ± 0.04 ef	0.72 ± 0.05 f	1.05 ± 0.04 c	1.02 ± 0.06 cd	1.34 ± 0.05 b	0.98 ± 0.04 cd	1.27 ± 0.05 b	1.38 ± 0.10 b	0.90 ± 0.05 de
total NEAA	13.07 ± 0.18 d	11.93 ± 0.24 e	17.15 ± 0.30 a	8.53 ± 0.17 g	8.04 ± 0.16 h	7.30 ± 0.17 i	9.52 ± 0.20 f	14.61 ± 0.23 c	9.30 ± 0.08 f	15.88 ± 0.27 b	17.41 ± 0.23 a	9.41 ± 0.25 f
TAA[d]	22.45 ± 1.85 c	20.66 ± 1.65 d	28.77 ± 1.32 a	14.05 ± 1.23 fg	12.86 ± 0.03 gh	11.65 ± 0.03 h	16.24 ± 1.32 e	24.23 ± 0.03 b	15.22 ± 0.03 ef	24.45 ± 0.03 b	27.04 ± 0.03 a	15.08 ± 0.03 ef
TEAA/TAA (%)	41.78	42.26	40.39	39.29	37.48	37.34	41.38	39.70	38.90	35.05	35.61	37.60

[a]Means with different letters within the same row are significantly different ($p < 0.05$). [b]EAA, essential amino acids. [c]NEAA, nonessential amino acids. [d]TAA, total amino acids.

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Table 4. Essential Amino Acid Score of Pollen Protein (Milligrams per Gram Protein)[a]

essential amino acid	FAO/WHO (1991)	pollen sample											
		BNL	CLL	CJL	DIL	FEL	HAL	NNG	PR	RR	SCL	VFL	ZML
threonine	34	35.57 (104.6)	36.27 (106.7)	34.19 (100.6)	30.28 (89.1)	32.26 (94.9)	28.03 (82.4)	38.91 (114.4)	36.42 (107.1)	23.97 (70.5)	29.93 (88.0)	33.04 (97.2)	31.86 (93.7)
valine	35	47.30 (135.1)	57.55 (164.4)	54.23 (154.9)	49.12 (140.3)	49.09 (140.3)	41.72 (119.2)	60.14 (171.8)	56.43 (161.2)	36.62 (104.6)	45.99 (127.8)	49.20 (140.6)	46.96 (134.2)
methionine + cystine	25	24.57 (98.3)	28.54 (114.2)	31.77 (127.1)	36.33 (145.3)	24.55 (98.2)	23.47 (93.9)	28.30 (113.2)	27.62 (110.5)	19.62 (78.5)	19.72 (78.9)	22.26 (89.0)	24.60 (98.4)
isoleucine	28	40.70 (145.4)	48.84 (174.4)	45.94 (164.1)	40.37 (144.2)	39.27 (140.3)	33.25 (118.8)	50.71 (181.1)	47.22 (168.6)	29.64 (105.9)	37.96 (135.6)	42.73 (152.6)	36.34 (129.8)
leucine	66	62.34 (94.5)	74.96 (113.6)	71.15 (107.8)	59.89 (90.7)	61.71 (93.5)	51.50 (78.0)	73.70 (111.7)	73.64 (111.6)	47.95 (72.7)	59.13 (89.6)	64.28 (97.4)	56.46 (85.5)
phenylalanine + tyrosine	63	64.54 (102.4)	77.38 (122.8)	74.26 (117.9)	59.21 (94.0)	62.42 (99.1)	48.24 (76.6)	76.65 (121.7)	74.43 (118.1)	49.26 (78.2)	65.71 (104.3)	66.08 (104.9)	57.02 (90.5)
lysine	58	56.11 (96.7)	54.16 (93.4)	53.54 (92.3)	57.87 (99.8)	50.49 (87.1)	45.63 (78.7)	56.01 (96.6)	54.03 (93.2)	34.87 (60.1)	51.83 (89.4)	57.10 (98.4)	51.43 (88.7)
tryptophan	11	18.78 (170.7)	35.21 (320.1)	20.45 (185.9)	31.49 (286.3)	21.60 (196.4)	18.25 (165.9)	22.17 (201.5)	8.84 (80.4)	17.96 (163.3)	15.33 (139.4)	17.96 (163.3)	19.23 (174.8)

[a]Data in parentheses show the essential amino acid (EAA) score, which is the result of dividing mean milligrams of EEA in 1 g of pollen protein by milligrams of EEA in 1 g of FAO/WHO (1990) reference pattern and multiplying by 100.

samples. Among samples, RR contained all 12 kinds of fatty acids, and PRL contained only 6 of them, but ranked first with a TUS/TS value of 10.98. The proportions of C18:3 were generally higher than C18:2 when compared with other studies.[17,19,41] Linoleic acid is an essential fatty acid required by humans from external sources, which is implicated in decreasing the ratio of low-density lipoproteins to high-density lipoproteins.[42] The ratios of TUS/TS were >1.0, with the exception of NNG pollen, which characteristically lacked C18:3 but had the highest concentration of C16:0 (64.38%). The higher ratios of TUS/TS in pollen supported the use of bee pollen as a food supplement in the diet to reduce the total amount of fats and cholesterol, preventing some cardiovascular disease. There were considerable variations in the unsaturated/saturated fatty acid ratio, which might be contributed to the different botanical origins or the processing and storage conditions. The results were also consistent with the notion that bees collect pollen with a high level of unsaturated fatty acids.[17]

Minerals. The mineral contents are reported in Table 7. The mineral composition of pollen is dependent on the intrinsic (botanical source) and the extrinsic (soil, geographical origin) conditions. The mean concentrations (mg/kg DW) of main abundant macro and minor elements could be put in the following decreasing order: phosphorus (P), 5946; potassium (K), 5324; calcium (Ca), 2068; magnesium (Mg), 1449; sodium (Na), 483.4; aluminum (Al), 129.3; iron (Fe), 119.3; manganese (Mn), 70.23; zinc (Zn), 45.10; and copper (Cu), 17.35. The amounts of beneficial elements, that is, K, Ca, Mg, Zn, Fe, Mn, and Cu, were higher than those of Spanish pollen[17,43] and Australian pollen,[21] especially as far as Ca and Mg were concerned. However, other beneficial trace elements, such as germanium (Ge) and selenium (Se), were not determined in all samples. RR contained the highest amounts of Ca, followed by VFL, BNL, and DIL. FEL and VFL had relatively higher contents of Fe. Both K and Mg reached a maximum concentration in NNG and ranked second in VFL. CJL contained the highest contents of both Mn and Zn, and the concentration of Mn in CJL was several times higher than that in other pollen species. Due to high contents of valuable minerals, pollen could be used as a natural source of minerals for human beings. The higher proportions of Zn against Cu were presented in all pollen species, and they might balance the lower ratio of Zn to Cu in the serum of cancer patients.[44]

The contents of detrimental trace elements, such as chromium (Cd), lead (Pb), and mercury (Hg), were all lower or not detected in pollen samples. Only the concentration of Pb in VFL and DIL slightly exceeded the standard of NY5137-2002 (1 mg/kg DW), but our results were higher than the international standard suggested by Campos et al.[13] as Cd ≤ 0.1 mg/kg, Pb ≤ 0.5 mg/kg, As ≤ 0.5 mg/kg, and Hg ≤ 0.03 mg/kg. Finally, it is worth noting that similar to those reported by Morgano et al.,[45] the concentration of aluminum (Al) was >100 mg/kg DW in most samples, especially in the pollen of CLL, with the highest value of 218.2 mg/kg DW. The excessive ingestion of Al may induce neurotoxicity[46] or reproductive or developmental toxicity.[47] Therefore, the consumption of pollen should take Al into consideration, primarily for elderly people, pregnant women, and children, referring to the provisional tolerable weekly intake (PTWI) of 2 mg/kg BW proposed by the Jonint FAO/WHO Expert Committee on Food Additives (JECFA) in 2011.[48]

Table 5. Free Amino Acid Composition of Pollen Samples (Milligrams per 100 g Dry Weight)[a]

amino acid	BNL	CLL	CJL	DIL	FEL	HAL	NNG	PRL	RR	SC	VFL	ZML
	pollen sample											
EAA												
threonine	2.95 ± 0.12 c	ND[b]	ND	ND	ND	ND	4.81 ± 2.32 b	ND	ND	ND	11.08 ± 5.32 a	ND
valine	1.94 ± 0.09 g	12.64 ± 1.13 d	7.73 ± 0.35 e	22.74 ± 1.50 b	15.22 ± 1.27 c	15.78 ± 1.00 c	17.78 ± 0.98 c	3.01 ± 0.14 f	1.80 ± 0.08 g	27.61 ± 1.65 a	16.32 ± 0.41 c	15.41 ± 0.62 c
methionine	ND	ND	ND	ND	8.30 ± 0.31 a	ND	ND	ND	ND	ND	ND	ND
isoleucine	2.58 ± 0.09 e	2.41 ± 0.09 e	0.97 ± 0.04 f	2.57 ± 0.11 e	7.22 ± 0.25 a	1.08 ± 0.06 f	5.0 4 ± 0.23 c	ND	ND	6.10 ± 0.23 b	2.68 ± 0.11 de	2.91 ± 0.12 d
leucine	4.24 ± 0.20 f	6.00 ± 0.22 c	2.68 ± 0.15 h	5.21 ± 0.24 d	13.41 ± 0.42 a	3.03 ± 0.13 g	13.59 ± 0.38 a	1.99 ± 0.07 i	1.14 ± 0.05 j	4.65 ± 0.19 e	4.64 ± 0.22 e	7.45 ± 0.36 b
phenylalanine	8.58 ± 0.34 g	11.48 ± 0.38 d	9.70 ± 0.39 f	10.71 ± 0.42 e	10.13 ± 0.43 ef	18.08 ± 0.40 c	23.35 ± 0.97 b	7.66 ± 0.26 h	2.95 ± 0.14 i	64.41 ± 2.45 a	9.83 ± 0.48 f	11.50 ± 0.46 d
lysine	16.51 ± 0.41 d	13.27 ± 0.38 f	15.35 ± 0.45 de	23.38 ± 0.61 a	14.28 ± 0.38 e	14.64 ± 0.36 ef	21.06 ± 0.53 bc	2.89 ± 0.13 g	15.06 ± 0.40 e	20.79 ± 0.46 c	22.00 ± 0.52 b	19.38 ± 0.68 c
tryptophan	32.92 ± 1.15 d	25.29 ± 1.02 fg	40.53 ± 2.28 b	27.35 ± 0.99 f	33.75 ± 1.37 cd	29.7 3 ± 1.40 e	35.37 ± 1.59 c	23.66 ± 1.22 g	44.28 ± 2.34 a	5.34 ± 0.25 h	30.81 ± 2.06 de	30.40 ± 1.58 de
total EAA	69.72 ± 1.88 g	71.09 ± 1.75 f	76.96 ± 2.14 e	91.96 ± 2.65 cd	102.3 ± 2.0 b	82.34 ± 1.50 de	121.0 ± 1.6 a	39.21 ± 1.17 h	65.23 ± 2.14 g	128.9 ± 2.97 a	97.36 ± 2.43 bc	87.05 ± 1.85 d
NEAA												
aspartic acid	6.87 ± 0.22 h	24.62 ± 0.78 de	22.82 ± 1.21 ef	30.46 ± 1.37 bc	46.51 ± 1.67 a	49.47 ± 1.45 a	28.58 ± 1.74 cd	19.53 ± 0.82 f	10.61 ± 056 g	33.28 ± 1.27 b	25.97 ± 1.11 d	18.90 ± 0.97 f
serine	26.34 ± 1.59 g	218.9 ± 11.2 a	35.99 ± 1.74 f	33.08 ± 1.41 f	ND	44.87 ± 2.02 e	148.4 ± 6.2 bc	45.85 ± 1.78 e	166.4 ± 8.3 b	115.1 ± 3.5 d	44.03 ± 1.03 e	133.6 ± 7.4 c
glutamic acid	720.9 ± 33.8 e	299.6 ± 12.8 h	1685 ± 53 b	354.7 ± 14.2 g	1686 ± 43 b	369.5 ± 15.5 fg	349.2 ± 15.4 g	867.6 ± 32.7 d	388.6 ± 11.5 f	2998 ± 114 a	1465 ± 75 c	227.1 ± 6.6 i
proline	478.4 ± 16.7 g	201.5 ± 8.4 i	1609 ± 55.7 c	1253 ± 48 d	722.0 ± 32.5 e	391.3 ± 21.8 h	51.32 ± 2.76 j	1397 ± 59 d	536.4 ± 24.4 f	3396 ± 156 b	4116 ± 148 a	1344 ± 67 d
glycine	3.46 ± 0.13 f	5.55 ± 0.18 c	2.67 ± 0.11 g	2.44 ± 0.12 g	4.47 ± 0.15 d	2.35 ± 0.11 h	4.86 ± 0.21 d	2.09 ± 0.15 hi	1.80 ± 0.10 i	3.89 ± 0.15 e	5.66 ± 0.21 b	9.25 ± 0.43 a
alanine	33.38 ± 1.68 e	58.73 ± 2.34 b	47.33 ± 2.19 c	45.44 ± 2.35 c	41.36 ± 1.98 d	45.20 ± 2.50 c	47.26 ± 1.97 c	22.12 ± 1.68 f	16.58 ± 0.82 g	48.21 ± 2.33 c	68.48 ± 3.10 a	58.47 ± 2.74 b
cystine	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
tyrosine	20.45 ± 0.75 g	132.4 ± 5.9 b	34.04 ± 1.75 e	29.83 ± 1.31 f	229.9 ± 9.88 a	18.57 ± 0.84 h	128.6 ± 43.7 b	30.42 ± 1.55 ef	27.44 ± 1.12 f	38.47 ± 1.34 d	17.08 ± 0.69 h	65.02 ± 2.88 c
histidine	71.42 ± 4.05 f	162.4 ± 8.8 b	85.66 ± 3.56 e	114.8 ± 3.9 c	96.28 ± 2.45 d	376.0 ± 1.4 a	89.58 ± 4.55 e	93.82 ± 4.73 de	60.43 ± 3.66 g	68.90 ± 4.15 f	35.22 ± 2.26 h	116.6 ± 5.3 c
arginine	34.73 ± 1.48 g	125.5 ± 4.8 a	70.18 ± 2.77 d	35.87 ± 1.53 g	46.11 ± 1.68 e	17.82 ± 0.98 h	38.25 ± 1.77 f	33.09 ± 1.81 g	37.61 ± 1.45 f	93.89 ± 3.63 b	65.29 ± 2.67 d	79.17 ± 2.80 c
total FAA[c]	1466 ± 48 h	1300 ± 35 h	3670 ± 105 c	1992 ± 43 g	2975 ± 81 d	1397 ± 39 hi	1007 ± 41 j	2551 ± 78 e	1311 ± 36 i	6925 ± 235 a	5931 ± 197 b	2139 ± 115 f
proline/total FAA (%)	32.63	15.50	43.84	62.90	24.27	28.01	34.68	54.76	40.92	49.04	69.40	62.83
total FAA/total AA (%)	1.61	1.86	3.51	3.25	6.91	2.12	2.47	2.99	2.05	0.95	6.22	3.75

[a]Means with different letters within the same row are significantly different ($p < 0.05$). [b]ND, not detected and. [c]FAA, free amino acids.

Table 6. Fatty Acid Composition of Pollen Samples[a]

fatty acid	pollen sample											
	BNL	CL	CJ	DI	FE	HA	NNG	PR	RR	SC	VF	ZM
C8:0	ND[f]	ND	ND	1.03 ± 0.11 b	ND	3.90 ± 0.35 a	ND	ND	0.39 ± 0.03 c	ND	ND	ND
C10:0	ND	ND	ND	14.98 ± 0.76 a	ND	0.38 ± 0.01 c	0.72 ± 0.07 b	ND	0.68 ± 0.05 b	ND	ND	ND
C12:0	1.01 ± 0.06 b	ND	0.34 ± 0.02 d	0.29 ± 0.04 d	ND	2.94 ± 0.32 a	ND	ND	0.70 ± 0.02 c	ND	0.13 ± 0.01 e	ND
C14:0	20.70 ± 1.11 a	0.61 ± 0.01 g	0.80 ± 0.02 e	ND	ND	1.17 ± 0.09 c	0.96 ± 0.04 d	ND	0.49 ± 0.02 h	1.74 ± 0.13 b	0.71 ± 0.03 f	ND
C16:0	17.55 ± 1.26 e	35.54 ± 1.65 b	19.91 ± 0.89 e	5.14 ± 0.25 h	14.38 ± 0.11 f	28.89 ± 1.77 c	64.38 ± 3.21 a	5.26 ± 0.15 h	12.59 ± 0.32 g	19.43 ± 0.88 e	35.89 ± 1.26 b	25.14 ± 2.31 d
C16:1	0.90 ± 0.02 b	0.73 ± 0.03 c	ND	0.88 ± 0.01 b	ND	ND	0.72 ± 0.02 c	ND	4.01 ± 0.15 a	4.39 ± 0.22 a	0.24 ± 0.01 d	ND
C18:0	6.41 ± 0.24 b	3.66 ± 0.21 e	4.23 ± 0.10 d	8.52 ± 0.55 a	4.38 ± 0.28 d	3.06 ± 0.17 f	3.85 ± 0.18 de	ND	3.36 ± 0.21 f	2.00 ± 0.09 g	5.25 ± 0.14 c	1.24 ± 0.09 h
C18:1	14.85 ± 0.97 e	11.11 ± 0.95 f	10.07 ± 0.57 fg	34.80 ± 1.21 b	41.24 ± 2.15 a	5.87 ± 0.38 i	23.48 ± 1.10 c	24.74 ± 1.86 c	23.41 ± 1.27 c	18.23 ± 1.31 d	9.30 ± 0.08 g	7.20 ± 0.52 h
C18:2	5.85 ± 0.21 h	7.44 ± 0.29 f	6.29 ± 0.20 g	24.38 ± 0.27 a	4.38 ± 0.02 i	10.21 ± 0.76 d	4.33 ± 0.17 i	8.39 ± 0.85 e	12.36 ± 0.78 c	18.79 ± 1.22 b	2.66 ± 0.34 j	10.26 ± 0.79 d
C18:3	30.82 ± 1.05 f	39.44 ± 1.32 e	46.91 ± 2.11 c	4.11 ± 0.26 i	17.18 ± 1.03 h	42.08 ± 2.11 d	ND	58.52 ± 4.89 a	20.64 ± 1.76 g	33.38 ± 3.00 f	41.45 ± 3.26 d	52.06 ± 5.66 b
C20:0	1.91 ± 0.56 c	1.47 ± 0.22 d	1.72 ± 0.11 b	0.59 ± 0.03 g	6.88 ± 0.26 a	1.09 ± 0.04 f	0.60 ± 0.14 g	0.67 ± 0.02 g	3.50 ± 0.36 b	1.27 ± 0.09 e	1.88 ± 0.03 c	1.59 ± 0.16 d
C22:0	ND	ND	9.73 ± 0.75 c	5.29 ± 0.67 d	11.56 ± 0.66 b	0.41 ± 0.02 h	0.96 ± 0.07 f	2.42 ± 0.11 e	17.89 ± 1.05 a	0.77 ± 0.05 g	2.49 ± 0.09 e	2.51 ± 0.14 e
TS[b]	47.58 ± 1.28 b	41.28 ± 2.94 cd	36.73 ± 1.33 ef	35.84 ± 2.11 e	37.20 ± 2.05 c	41.84 ± 2.35 cd	71.47 ± 2.65 a	8.35 ± 0.37 g	36.81 ± 1.03 e	25.21 ± 1.69 g	46.35 ± 3.01 bc	30.48 ± 1.66 f
TM[c]	15.75 ± 0.77 d	11.84 ± 0.68 e	10.07 ± 0.57 e	28.49 ± 0.98 b	41.24 ± 2.15 a	5.87 ± 0.38 h	24.20 ± 1.14 c	24.74 ± 1.86 b	27.42 ± 1.33 a	22.62 ± 0.78 c	9.54 ± 0.06 f	7.20 ± 0.52 c
TP[d]	36.67 ± 1.11 d	46.88 ± 1.14 c	53.20 ± 2.34 b	35.67 ± 0.38 d	21.56 ± 0.88 e	52.29 ± 2.65 b	4.33 ± 0.17 f	66.91 ± 3.14 a	35.77 ± 1.77 d	52.17 ± 2.15 b	44.11 ± 1.76 c	62.32 ± 3.42 a
TUS[e]/TS	1.10	1.42	1.72	1.79	1.69	1.39	0.40	10.98	1.72	2.97	1.16	2.28

[a]Values are expressed as percentage of total fatty acids. Means with different letters within the same row are significantly different ($p < 0.05$). [b]TS, total saturated fatty acids. [c]TM, total monounsaturated fatty acids. [d]TP, total polyunsaturated fatty acids. [e]TUS, total unsaturated fatty acids. [f]ND, not detected.

Table 7. Mineral Concentration of Pollen Samples (Micrograms per Gram Dry Matter Basis)[a]

element	pollen sample											
	BNL	CLL	CJL	DIL	FEL	HAL	NNG	PRL	RR	SC	VFL	ZML
Al	99.54 ± 0.13 h	218.2 ± 0.1 a	119.5 ± 0.1 f	135.2 ± 0.1 d	129.2 ± 0.1 e	111.2 ± 1.5 g	94.51 ± 0.23 i	115.1 ± 1.7 e	125.8 ± 0.1 ef	151.4 ± 0.2 c	143.4 ± 1.5 c	107.9 ± 0.1 g
As	5.24 ± 0.21 d	6.92 ± 0.18 c	11.70 ± 0.42 b	14.71 ± 0.19 a	6.72 ± 0.21 c	12.15 ± 0.42 b	13.97 ± 0.51 a	2.19 ± 0.15 f	2.19 ± 0.11	3.21 ± 0.08 e	14.01 ± 0.02 a	2.42 ± 0.04 f
Ca	2820 ± 6 b	1995 ± 14 e	1543 ± 11 g	2618 ± 3 c	2080 ± 14 e	1719 ± 8 f	1867 ± 7 f	828 ± 5 i	3053 ± 5 a	2278 ± 6 d	3016 ± 30 a	999 ± 5 h
Cd	0.23 ± 0.01 ef	0.23 ± 0.01 ef	0.27 ± 0.01 d	0.49 ± 0.01 a	0.35 ± 0.01 b	0.31 ± 0.01 c	0.37 ± 0.01 b	0.25 ± 0.01 de	0.21 ± 0.01 f	0.16 ± 0.00 g	0.26 ± 0.01 d	0.15 ± 0.00 h
Co	0.37 ± 0.01c	0.41 ± 0.01 b	0.43 ± 0.02 b	0.26 ± 0.01d	0.20 ± 0.01 f	0.22 ± 0.01 e	0.45 ± 0.02 b	0.54 ± 0.02 a	0.53 ± 0.02 a	0.27 ± 0.01d	0.44 ± 0.02 b	0.25 ± 0.01 d
Cr	ND[b]	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Cu	15.6 6 ± 0.13 d	19.98 ± 0.12 c	20.08 ± 0.03 bc	21.43 ± 0.10 b	8.31 ± 0.10 f	15.24 ± 0.02 d	15.92 ± 0.17 d	15.84 ± 0.23 d	19.65 ± 0.09 c	25.11 ± 0.03 a	18.74 ± 0.17 c	12.21 ± 0.03e
Fe	89.4 ± 0.2 e	104.6 ± 0.1 d	97.7 ± 0.1 d	129.6 ± 0.2 c	207.8 ± 0.6 a	88.3 ± 0.5 e	76.4 ± 0.4 f	131.4 ± 0.3 c	136.8 ± 0.1 c	135.9 ± 0.1 c	158.3 ± 0.5 b	75.2 ± 0.4 f
Ge	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Hg	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
K	4739 ± 23 d	4785 ± 12 d	6358 ± 11 c	2353 ± 24 f	5894 ± 11 c	4351 ± 12d	8204 ± 127 a	4520 ± 30 de	4382 ± 19 e	6113 ± 20 c	7149 ± 122 b	5035 ± 5 d
Mg	1405 ± 11 c	1457 ± 9 c	1262 ± 18 d	321 ± 1 i	2835 ± 12 a	57 1 ± 3 h	2777 ± 49 a	712 ± 8 g	1089 ± 6 e	1939 ± 1 b	2202 ± 19 b	815 ± 1 f
Mn	24.21 ± 0.54 f	33.85 ± 0.12 d	357.4 ± 2.0 a	8.69 ± 0.02 h	31.75 ± 0.18 d	16.70 ± 0.03 g	99.40 ± 0.17 c	32.95 ± 0.17 d	156.1 ± 2.3 b	30.33 ± 0.04 de	27.29 ± 0.47 e	24.06 ± 0.02 f
Mo	0.21 ± 0.01	ND	ND	ND	ND	0.12 ± 0.01	ND	ND	ND	0.37 ± 0.01	0.22 ± 0.01	0.44 ± 0.02
Na	459.1 ± 4.1 e	569.0 ± 3.1 d	616.3 ± 2.1 c	590.7 ± 5.1 d	274.1 ± 0.82 i	292.5 ± 0.88 h	846.4 ± 16.8 a	462.0 ± 1.3 e	379.1 ± 6.1 f	327.0 ± 2.0 g	689.9 ± 13.4 b	294.3 ± 0.5 h
Ni	0.55 ± 0.01 g	1.87 ± 0.02 c	3.97 ± 0.01 a	1.36 ± 0.05 d	0.88 ± 0.02 e	0.53 ± 0.02 g	0.70 ± 0.02 f	1.06 ± 0.03	2.77 ± 0.08 b	0.70 ± 0.03 f	0.88 ± 0.03 e	0.76 ± 0.02 f
P	6758 ± 13 cd	5806 ± 9 d	6937 ± 19 c	2136 ± 2 h	6618 ± 15 cd	2587 ± 6 g	9587 ± 23 a	5835 ± 17 d	4804 ± 11 e	7043 ± 17 c	8587 ± 111 b	4651 ± 9 e
Pb	ND	0.52 ± 0.01 d	0.12 ± 0.00 g	1.06 ± 0.03 b	0.23 ± 0.01 e	0.56 ± 0.02 d	0.05 ± 0.00 i	0.10 ± 0.00 h	0.64 ± 0.02 c	0.96 ± 0.02 b	1.37 ± 0.04 a	0.13 ± 0.00 f
Se	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
Zn	34.08 ± 1.10 e	44.22 ± 0.21 d	65.30 ± 2.91 a	28.25 ± 0.10 f	30.43 ± 1.14 f	43.95 ± 0.72 d	46.25 ± 0.57 d	50.16 ± 0.17 c	43.01 ± 0.50 d	45.84 ± 0.58 d	55.04 ± 0.02 b	54.62 ± 0.89 b

[a]Means with different letters within a row are significantly different ($p < 0.05$). [b]ND, not detected.

In conclusion, the above findings might provide a scientific basis for evaluating nutritional values of bee pollen and contribute to a database of food composition. These data might also be used as a reference for health agencies' recommendations, consumers' choices, industry exploration, and production of bee pollen.

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Notes

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ABBREVIATIONS USED

BNL, *Brassica napus* L.; CLL, *Citrullus lanatus* L.; CJL, *Camellia japonica* L.; DIL, *Dendranthema indicum* L.; FEL, *Fagopyrum esculentum* L.; HAL, *Helianthus annuus* L.; NNG, *Nelumbo nucifera* Gaertn.; PRL, *Papaver rhoeas* L.; RR, *Rosa rugosa*; SC, *Schisandra chinensis*; VFL, *Vicia faba* Linn.; ZML, *Zea mays* L.; AOAC, Association of Offical Analytical Chemists; ANOVA, analysis of variance; TDF, total dietary fiber; SDF, soluble dietary fiber; IDF, insoluble dietary fiber; FAMEs, fatty acid methyl esters; GC, gas chromatography; FID, flame ionization detection; DW, dry weight; ICP-AES, inductively coupled plasma atomic emission spectroscopy; SD, standard deviation; EAA, essential amino acids; NEAA, nonessential amino acids; TAA, total amino acids; ND, not detected; FAA, free amino acids; FAO, Food and Agriculture Organization of the United Nations; WHO, World Health Organization; TS, total saturated fatty acids; TUS, total unsaturated fatty acids; TM, total monounsaturated fatty acids; TP, total polyunsaturated fatty acids; PTWI, provisional tolerable weekly intake; JECFA, Joint FAO/WHO Expert Committee on Food Additives

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