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#### SHORT COMMUNICATION

# *Prunus spinosa* L. pollen - Quantity and nutritional quality

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## Abstract

Pollen production and pollen quality in *Prunus spinosa* L. (Rosaceae), a common early-spring flowering plant from the temperate zone was evaluated. The species is an efficient pollen producer, as it can produce 0.23 mg of pollen per flower and 1.99 g of pollen per 1 m<sup>2</sup> of shrub surface, however, the values may differ considerably between seasons. Its pollen contains a high amount of proteins (22.1–34.2%). The total lipid content in the pollen ranged between 2.7 and 3.6%. The presence of omega-3, omega-6, and omega-9 fatty acids was found. Among omega-3 fatty acids, inflammation inhibitors, i.e.,  $\alpha$ -linolenic acid and arachidonic acid, were detected. The predominant mineral was potassium, followed by calcium and magnesium. *P. spinosa* pollen is an important ingredient of bee pollen loads containing 89.1–98.2% of *Prunus* pollen. *P. spinosa* should be recommended for planting in the agricultural landscape in order to support the early spring diet for pollinators.

## **Keywords**

pollen production; pollen proteins; lipids; fatty acids; chemical elements

## 1. Introduction

Poor nutrition, i.e., lack of food resources or unbalanced diet derivative to landscape changes (e.g., fragmentation of habitats, crop structure, lack of weeds), are indicated as key factors responsible for pollinator decline (e.g., Bożek et al., 2023). In addition to energy, whose main source is nectar, pollinators need diverse nutrients, mainly provided by plant pollen, to conduct numerous metabolic processes (Bożek, 2021; Filipiak et al., 2017). In particular, the role of pollen proteins, lipids, macro- and microminerals, vitamins, and hormones in the pollinator diet is highlighted (Lau et al., 2022). Pollen is also used by the industry (e.g., food and beverage, pharmaceutical and nutraceutical, cosmetic and personal care); therefore, the demand for this product is increasing due to the increase in social awareness of the health-related properties of bee pollen and the tendency to use healthy diets (Oliveira & Ribeiro, 2020).

In temperate climate zone, an adequate quantity and quality of early spring pollen is required to enhance honey bee colony health (Brodschneider & Crailsheim, 2010). In the agricultural landscape, *Prunus spinosa* L. linear or grouped shrubs are considered as non-forest woody plants with diverse ecological functions, e.g., wind barriers or food for animals (Bożek et al., 2023).

In this study, analyses of pollen production and the pollen chemical composition (total protein content, lipid content and composition, mineral element content) in *Prunus spinosa* L. were performed. These data can give preliminary information on the value of *P. spinosa* pollen for insects and for possible human use.

## 2. Material and methods

The research was conducted in 2021–2022 on *Prunus spinosa* L. grown in Dąbrowica (SE Poland). The mass of produced pollen was established with the ether-ethanol

method, and the total protein content was determined based on the Kjeldahl method (Denisow, 2011).

The elemental analyses were performed with the Flame Atomic Absorption Spectrometry Methodology (FAAS). Samples were mineralized in a Mars Xpress CEM (USA), and the elemental analyses were performed using a Varian SpectrAA 20FS spectrophotometer.

The determination of the composition of fatty acids was carried out using a Varian 450-GC gas chromatograph (Varian Inc., Temecula, CA, USA) equipped with an 1177 Split/Splitless injector and a Select<sup>™</sup> Biodiesel CP9080 for FAME capillary column (30 m; 0.32 mm; 0.25 µm) (Agilent Technologies Inc., Santa Clara, CA, USA).

The botanical composition of pollen grains in pollen loads obtained from honey bees was determined in microscopic slides (Nikon Eclipse E 600 light microscope;  $40 \times 15$ ).

All statistical analyses were performed using Statistica ver. 13 (Statsoft, Poland). Analysis of variance (ANOVA) was employed to test the year effect on pollen production data. Differences were tested with Tukey's test (p = 0.05).

### 3. Results and discussion

In *P. spinosa*, the average mass of pollen produced per flower was 0.23 mg (Table 1). The value is within the range (0.11–0.37 mg per flower) established for *P. spinosa* by Denisow (2011). The pollen mass in the flowers differed between the years of the study. It is agreed that pollen productivity is very sensitive to environmental conditions. For example, in shrub and tree species, a drought during bud setting or spring frost can restrict flower and anther formation and pollen development (Dmitruk et al., 2022; Lotocka et al., 2023). The amount of pollen available per 1 m<sup>2</sup> of the shrub surface differed considerably between the seasons. High differences in total pollen resource availability derivative to the alternate flowering rhythm are a known tendency in trees and shrubs (e.g. Denisow, 2011).

The protein content in the *P. spinosa* pollen ranged between 22.1 and 34.2% (Table 2). As reported by Brodschneider & Crailsheim (2010), pollen that contains >20% of protein can be regarded as high quality pollen. Such pollen is known to be attractive for many insect pollinators (Di Pasquale et al., 2013) and is of great importance for the resistance of honey bees to diseases (Amdam & Omholt, 2002). De Sá-Otero et al. (2009) determined lower values (ca. 11–20% of proteins) in *P. spinosa* pollen and pointed out that the protein content in plant pollen depends on the season.

Year	Pollen production per					
	Flower (mg)			<b>m</b> <sup>2</sup> ( <b>g</b> )		
	min-max	Mean	±SD	min-max	Mean	±SD
2021	0.13-0.38	0.26 <sub>b</sub>	0.09	1.87-3.29	2.68 <sub>b</sub>	0.87
2022	0.15-0.24	0.19 <sub>a</sub>	0.04	0.78-3.04	1.29 <sub>a</sub>	0.42
mean		0.23 <sub>A</sub>			1.99 <sub>A</sub>	

Means with different letters differ significantly (Tukey's test; p = 0.05).

Table 2 Protein content in *P. spinosa* pollen and honey bee pollen loads collected in 2021–2022 in SE Poland.

Year		Protein (%)	
		Plant pollen	
	min-max	Mean	±SD
2021	22.1-26.8	24.6 <sub>a</sub>	4.1
2022	27.6-34.2	31.2 <sub>b</sub>	3.7
mean		27.9 <sub>A</sub>	

Means with different letters differ significantly (Tukey's test; p = 0.05).

This is in line with our observations, which showed that the protein content differed significantly between the years of the study. This may be related e.g., to changeable environmental conditions (weather factors, abiotic stresses) that have an impact on biochemical processes and can stimulate protein degradation (Borghi et al., 2019).

The total lipid content in the *P. spinosa* pollen ranged between 2.7 and 3.6% (Table 3) and was similar to that determined by Spulber et al. (2018) in monofloral pollen of *Prunus* L. sp. (3.26%) collected in diverse regions of Romania. The lipids in the *P. spinosa* pollen were dominated by saturated fatty acids (SFAs). Our values are different from the proportion of fatty acids detected in Portuguese bee pollen in which *Prunus* pollen grains were detected (Feás et al., 2012). In their study, the levels of SFAs, MUFAs, and PUFAs were in the range of 13.8–30.5%, 4.6–20.6%, and 50–70%, respectively. Such disparity between crude *P. spinosa* pollen and bee

**Table 3** Content and composition of saturated fatty acids, monounsaturated fatty acids, polyunsaturated fatty acids in *P. spinosa* pollen.

Fatty acids		
	%	g/100 g
Saturated fatty acids (SFA)		
hexanoic acid (caproic acid) C6:0	0.22	0.016
octanoic acid (caprylic acid) C8:0	0.32	0.023
decanoic acid (capric acid) C10:0	0.15	0.011
undecanoic acid C11:0	<lod 0.016<="" =="" td=""><td></td></lod>	
n-dodecanoic acid (lauric acid) C12:0	1.15	0.084
tridecanoic acid C13:0	<lod 0.016<="" =="" td=""><td></td></lod>	
tetradecanoic acid (myristic acid) C14:0	6.65	0.483
oleomyristic acid C14:1n5	<lod 0.016<="" =="" td=""><td></td></lod>	
pentadecanoic acid (pentadecylic acid) C15:0	0.23	0.017
hexadecanoic acid (palmitic acid) C16:0	19.86	1.444
heptadecanoic acid (margarine acid) C17:0	0.18	0.013
octadecanoic acid (stearic acid) C18:0	9.41	0.684
eicosanoic a cid (arachidic acid) C20:0	2.71	0.197
cis-11-eicosenoic acid C20:1n9	<lod 0.016<="" =="" td=""><td></td></lod>	
heneicosanoic acid C21:0	<lod 0.016<="" =="" td=""><td></td></lod>	
docosanoic acid (behenic acid) C22:0	5.14	0.374
tricosanoic acid C23:0	0.36	0.026
tetracosanoic acid (lignoceric acid) C24:0	0.63	0.046
Monounsaturated fatty acid (MUFA)		
cis-10-pentadecenoic acid C15:1n5	<lod 0.016<="" =="" td=""><td></td></lod>	
cis-9-hexadecenoic acid (palmitoleic acid) C16:1n7	1.17	0.085
cis-10-heptadecanoic acid C17:1n7	0.19	0.014
oleic acid+elaidic acid C18:1n9c+C18:1n9t	25.99	1.889
gamma-linolenic acid C18:3n6 (gamma)	<lod 0.016<="" =="" td=""><td></td></lod>	
cis-5-acideicosene C20:1n15	1.14	0.083
cis-11.14-eicosadienoic acid C20:2n6	<lod 0.016<="" =="" td=""><td></td></lod>	
cis-11.14.17-eicosatrienoic acid (ETE) C20:3n3	<lod 0.016<="" =="" td=""><td></td></lod>	
erucic acid C22:1n9	<lod 0.016<="" =="" td=""><td></td></lod>	
nervonic acid C24:1n9	<lod 0.016<="" =="" td=""><td></td></lod>	
Polyunsaturated fatty acid (PUFA)		
linoleic acid+9,12-trans-octadecadienoic acid C18:2n6c+C18:2n6t	3.04	0.221
cis-9,12,15-octadecatrienoic acid (α-linolenic acid) C18:3n3 (alpha)	2.55	0.185
dihomo-⊠-linolenic acid C20:3n6	0.10	0.007
arachidonic acid (ARA) C20:4n6	<lod 0.016<="" =="" td=""><td></td></lod>	

Continued on next page

Fatty	y acids

, ,	%	g/100 g
Saturated fatty acids (SFA)		0_0
cis-5.8.11.14.17 acid-eicosapentaenoic (EPA) C20:5n3	1.07	0.078
cis-13,16-docosadienoic acid (docosadienoate) C22:2n6	<lod 0.016<="" =="" td=""><td></td></lod>	
cis-4.7.10.13.16.19 acid-docosahexaenoic acid (DHA) C22:6n3	<lod 0.016<="" =="" td=""><td></td></lod>	
SFA	47.01	3.418
MUFA	28.49	2.071
PUFA	6.76	0.491
OMEGA 3	3.62	0.263
OMEGA 6	3.14	0.228
OMEGA 9	25.99	1.889
LOD - Limit of Detection.		





pollen with participation of *Prunus* pollen may be derivative to many factors, i.e., the chemical composition of the pollen of other species present in pollen loads, the weather conditions, and the geographical region (Radev, 2018).

The presence of omega-3, omega-6, and omega-9 fatty acids was found in the analyzed pollen. Omega-3 fatty acids (e.g.,  $\alpha$ -linolenic acid, arachidonic acid) can prevent inflammation by reducing the inflammation mediators and are necessary in the honey bee diet (e.g., Yu et al., 2022).

In total, 3.333 g/100 g of fatty acids were detected in the *P. spinosa* pollen. Among the fatty acids, the highest amounts of oleic and elaidic (1.889 g/100 g), palmitic (1.444 g/100 g), stearic (0.684 g/100 g), and myristic (0.483 g/100 g) acids were recorded. Oleic acid is important during oxidative stress, acting as an antioxidant (Hu et al., 2022). Pollen with high levels of oleic and palmitic acids is regarded to play a significant role in honey bee nutrition (Manning, 2001).

The predominant mineral was potassium, followed by calcium and magnesium. Spulber et al. (2018) reported that samples with a high proportion of *Prunus* pollen originating from Romania exhibited a higher amount of magnesium ( $666.7 \pm 1.05$ ) than potassium ( $4073 \pm 3.21$ ). In our analyses, a high amount of Fe was documented, followed by Zn, Mn, and Cu. Iron-rich *Prunus* sp. pollen was also reported by Spulber

et al. (2018), however, the Fe content in their analyses was almost 3-times higher (150.9  $\pm$  1.11 mg/kg). As shown by Filipiak et al. (2017), the proportion of elements in the insect pollinator nutrition is of great importance for stoichiometrically balanced diets.

We observed honey bees willingly collecting pollen from *P. spinosa* flowers. In both study years, the honey bees predominated and accounted for 68–72% of all insect visitors (Figure 1). Our observations of the high attractiveness of *P. spinosa* pollen were confirmed in analyses of botanical pollen loads. Pollen loads may contain 89.1–98.2% of *Prunus* pollen. *Prunus* pollen constitutes an important ingredient of bee pollen (Bobis et al., 2010; Ceksteryte et al., 2013). It was also found as the primary pollen source for honey bees and wild bumblebees in Michigan (Graham et al., 2023), which indicates its dietary importance irrespective of the geographical region of the temperate zone.

In conclusion, *P. spinosa* should be recommended for planting in the agricultural landscape in order to support the early spring diet for pollinators.

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