

Pollen morphology investigations of three *Buddleja* L. (Scrophulariaceae) taxa used in landscaping of parks and gardens from Eskişehir province

İsmühan Potoğlu Erkara ^{1,a,*} ^(D), Okan Sezer ^{1,b} ^(D)

¹Eskişehir Osmangazi University, Faculty of Science, Department of Biology, Eskişehir, Turkey

*Corresponding author: ismuhan@ogu.edu.tr

(Received: May 24, 2023 / Accepted: July 10, 2023)

Abstract

In this study, pollen morphology of three *Buddleja davidii* cultivars (*B. davidii* Franch cv. *monum*, *B. davidii* Franch cv. *royal red* and *B. davidii* Franch cv. *white profusion*) from Scrophulariaceae, which are used for landscaping in Eskişehir province, were investigated by light and scanning electron microscopy. Pollen grains of investigated cultivars are tricolporatae/tetracolporatae type and sphaeroidal shaped. Exine was seen to be tectatae-psilate-perforatae ornamentation. As a result of microscopic examinations, the distinctions between taxa were revealed statistically. Comparisons between studied taxa will contribute to taxonomy for classification of *Buddleja davidii* taxa.

Keywords: Buddleja, pollen morphology, Eskişehir, Turkey

Introduction

Plant taxonomy, the science of classifying and identifying plant species, plays a crucial role in understanding the diversity and evolutionary relationships within the plant kingdom. One discipline that has greatly contributed to plant taxonomy is palynology, the study of pollen grains and spores. Palynology provides invaluable insights into plant evolution, species identification, and ecological interactions.

Palynology offers a unique perspective on plant taxonomy by focusing on microscopic features that can be preserved for thousands or even millions of years. By examining pollen grains and spores, palynologists can identify plant taxa, determine their evolutionary relationships, and reconstruct past environments. These microscopic structures act as a botanical fingerprint, providing essential clues for plant classification and systematic studies.

Palynology finds extensive applications in both modern and fossil plant studies. In modern plant taxonomy, palynological data complement morphological and molecular approaches, especially in cases where traditional characteristics may be limited or ambiguous. Pollen morphology, including size, shape, and surface ornamentation, can serve as reliable diagnostic features, aiding in identifying and differentiating closely related species [1, 2].

The genus *Buddleja*, commonly known as butterfly bush, comprises a diverse group of flowering plants within the family Scrophulariaceae. *Buddleja* species are predominantly shrubs or small trees characterized by their showy inflorescences and attractive flowers that attract butterflies and pollinators. Taxonomically, the genus *Buddleja* is classified within the order Lamiales and the family Scrophulariaceae. It is further divided into several sections and species, with approximately 100 recognized species distributed across different regions of the world. The taxonomy of the genus *Buddleja* has been studied extensively using various approaches, including morphological, anatomical, and molecular techniques. These studies have contributed to the identification and description of new species, as well as the clarification of relationships within the genus. The taxonomic revision and classification of *Buddleja* have relied on comprehensive investigations that incorporate multiple lines of evidence, such as floral characteristics, leaf morphology, reproductive traits, and genetic analyses. Further taxonomic studies are essential to refine the understanding of the *Buddleja*, its species boundaries, and its evolutionary history [3, 4].

Numerous taxa within the Scrophulariaceae family have been extensively investigated in terms of their palynological characteristics; however, studies focusing specifically on the *Buddleja* taxa are notably limited. This study aims to determine the palynological features of *B. davidii* cultivars and assess their potential usefulness in distinguishing taxa at the infraspecific level.

Materials and Methods

The materials of the study are three *Buddleja davidii* cultivars (*B. davidii* Franch cv. *monum*, *B. davidii* Franch cv. *royal red* and *B. davidii* Franch cv. *white profusion*), used for herbal design in the parks and gardens in Eskişehir city center. Pollen samples of the plants were obtained from the flowers in the trees located on the parks and gardens. Pollen samples of the investigated taxa were taken from dried plants found in the Herbarium (OUFE) of Osmangazi University Faculty of Science. The examination of current pollen grains under light microscope was done by Wodehouse (1935) method, and examination of fossil pollen by Erdtman (1969) method. Morphological examinations of the pollen grains were done under Nikon binocular microscope, oil immersion objective (x100). 50 times measurements were made for all parameters to determine the average values. Standard deviation and variations have been calculated. Each range in the ocular micrometer is 0.98 µm. Microphotographs were taken with a Nikon 80i type microscope and a KAMERAM Digital camera in the Department of Biology, Faculty of Science, Eskişehir Osmangazi University. The magnification of the photos is x1000. For Scanning electron microscopy (SEM) examinations, unacetholyzed pollen grains were placed on the fixing plate and covered with gold and examined under Jeol 5600 LV Scanning electron microscope (SEM) [5-10]. Various basic palynological books and various studies have been used for the diagnosis of pollen [1, 2; 5-11].

Results and Discussion

Species: *Buddleja davidii* cv. *monum* Pollen Type: Tricolporatae Pollen Shape: Sphaeroidea P/E= 0.90 μm (W); 1.20 μm (E) Exine: Average thickness 1.08 μm (W); 0.96 μm (E) Aperture: Colpus long and slender; the borders are not clear, the ends are stubby. The pores are prominent and located at the midpoint of the colpus. Structure: Tectatae Sculpture: Psilatae-perforatae

Wodehouse Method			Erdtman Method		
	Μ	S	Μ	S	
Р	15.28	± 1.74	15.40	± 2.38	μm
Ε	14.16	± 2.14	13.06	± 1.14	μm
clg	13.86	± 2.06	12.36	± 1.80	μm
clt	4.25	± 1.22	4.32	± 1.18	μm
plg	5.78	± 1.66	5.12	± 1.12	μm
plt	3.48	± 1.56	3.35	± 1.54	μm
L	14.06	± 1.24	14.68	± 1.42	μm
t	4.04	± 0.78	4.40	± 0.80	μm
i	0.82	± 0.08	-	-	μm
Ex	1.08	± 0.12	0.96	± 0.10	μm

Table 1. Morfometrical Data of Buddleja davidii cv. monum

Species: Buddleja davidii cv. royal red

Pollen Type: Tricolporatae/Tetracolporatae

Pollen Shape: Sphaeroidea P/E= $1.10 \mu m$ (W); $1.08 \mu m$ (E)

Exine: Average thickness 1 µm (W); 0.86 µm (E)

Aperture: Colpus slender and long, well-defined, pointed ends. Pores are prominent.

Structure: Tectatae

Sculpture: Psilatae-perforatae

Wodehouse Method			Erdtman Method		
Р	M 17.28	S ± 2.48	M 15.13	S	
				± 1.44	μm
Ε	16.16	± 1.24	15.06	± 1.12	μm
clg	15.86	± 1.28	14.36	± 1.18	μm
clt	7.30	± 1.16	5.22	± 1.18	μm
plg	5.62	±1.64	5.42	± 1.06	μm
plt	3.64	± 1.20	3.25	± 0.74	μm
L	15.32	± 2.12	14.98	± 2.24	μm
t	8.18	± 1.66	7.04	± 0.80	μm
i	0.88	± 0.14	-	-	μm
Ex	1.00	± 0.14	0.86	± 0.14	μm

Table 2. Morfometrical Data of Buddleja davidii cv. royal red

Species: Buddleja davidii cv. white profusion

Pollen Type: Tricolporatae

Pollen Shape: Sphaeroidea P/E= $1.10 \ \mu m$ (W); $1.12 \ \mu m$ (E)

Exine: Average thickness 1.22 µm (W); 1 µm (E)

Aperture: Colpus long and slender; the borders are not clear, the ends are stubby. The pores are prominent and located at the midpoint of the colpus.

Structure: Tectatae

Sculpture: Psilatae-perforatae

Table 3. Morfometrical Data of Buddleja davidii cv. white profusion

Wodehouse Method			Erdtman Method		
	M 18.28	S ± 1.68	M 26.22	S	
Р				± 2.44	μm
Е	13.16	± 3.24	24.06	± 2.12	μm
clg	13.86	± 1.28	22.36	± 1.18	μm
clt	5.30	± 1.16	6.22	± 1.18	μm
plg	5.62	±1.64	5.42	± 1.06	μm
plt	6.74	± 1.20	5.25	± 0.74	μm
L	12.32	± 2.12	21.98	± 2.24	μm
t	5.18	± 1.66	5.04	± 0.80	μm
i	0.88	± 0.14	-	-	μm
Ex	1.22	± 0.14	1.00	± 0.00	μm

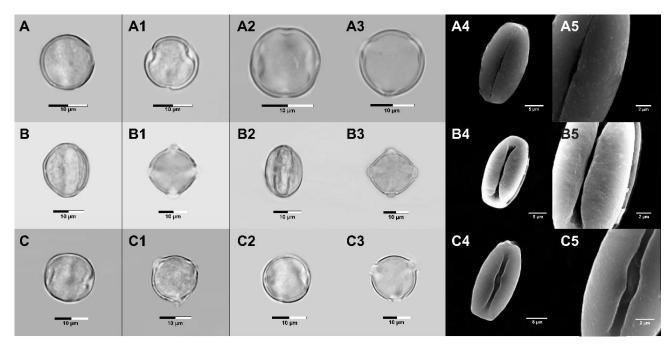


Figure 1. LM (A-C: Equatorial (W), A1-C1: Polar (W), A2-C2: Equatorial (E), A3-C3: Polar (E)) and SEM (A4-C4): Equatorial view, A5-C5: Exine ornamentation microphotographs of investigated taxa (A-A5: cv. monum; B-B5: cv. royal red; C-C5: cv. white profusion)

In this study, palynological characteristics of three different cultivars of *Buddleja davidii*, namely cv. *monum*, cv. *royal red*, and cv. *white profusion*, were examined and compared.

Starting with cv. *monum*, the pollen type was identified as tricolporatae, and the pollen shape was determined to be sphaeroidea with a ratio of $P/E = 0.90 \ \mu m$ (W) and $1.20 \ \mu m$ (E). The average thickness of the exine layer was measured to be $1.08 \ \mu m$ (W) and $0.96 \ \mu m$ (E). The aperture exhibited a colpus structure that was long, slender, and located at the midpoint, with prominent pores. The overall structure was classified as tectatae, and the sculpture was identified as psilatae-perforatae.

In cv. *royal red*, the pollen type was classified as tricolporatae/tetracolporatae, and the pollen shape was also sphaeroidea with a P/E ratio of 1.10 μ m (W) and 1.08 μ m (E). The average thickness of the exine layer was measured to be 1 μ m (W) and 0.86 μ m (E). The aperture exhibited a well-defined, slender, and long colpus structure with pointed ends and prominent pores. Similar to cv. *monum*, the overall structure was categorized as tectatae, and the sculpture was identified as psilatae-perforatae.

Lastly, for cv. *white profusion*, the pollen type was identified as tricolporatae, and the pollen shape was again determined to be sphaeroidea with a P/E ratio of 1.10 μ m (W) and 1.12 μ m (E). The average thickness of the exine layer was measured to be 1.22 μ m (W) and 1 μ m (E). The aperture displayed a colpus structure similar to the previous cultivars, being long, slender, and located at the midpoint, with prominent pores. The overall structure was classified as tectatae, and the sculpture was identified as psilatae-perforatae.

As a result, the examined cultivars of *Buddleja davidii* displayed similar palynological characteristics, including tricolporatae pollen type, sphaeroidea pollen shape, tectatae structure, and psilatae-perforatae sculpture. However, slight variations were observed in the size and thickness of the pollen grains and exine layer among the different cultivars. These variations provide valuable insights into the reproductive biology and evolutionary aspects of these cultivars of *Buddleja davidii*.

Conclusion

Based on the data obtained from the analysis of three different cultivars of *Buddleja davidii*, namely cv. *monum*, cv. *royal red*, and cv. *white profusion*, it can be concluded that these cultivars exhibit similar palynological characteristics. All three cultivars have tricolporate pollen type and sphaeroidea pollen shape, with a tectatae structure and psilatae-perforatae sculpture. However, there are slight differences in the size and thickness of the pollen grains and exine layer among the cultivars. These findings contribute to our

understanding of the reproductive biology and evolutionary aspects of *Buddleja davidii* cultivars. Further research and studies are needed to explore the significance of these palynological features in the differentiation of cultivars and their implications in the taxonomy and breeding of *Buddleja davidii*.

References

- [1] Moore PD, Webb JA, Collinson ME. (1991). Pollen analysis. Oxford Blackwell Scientific Publications, London, pages: 110–112.
- [2] Faegri K, Iversen J. (1975). Textbook of pollen-analysis. 3rd edition. Munksgaard, Copenhagen.
- [3] Coelho GP, Miotto STS. (2018). A taxonomic revision of the genus Buddleja (Buddlejeae, Scrophulariaceae) in Brazil. *Phytotaxa*, 379(2), 187–226.
- [4] APG IV (2016) An update of the Angiosperm Phylogeny Group classification for the orders and families of flowering plants: APG IV. Botanical Journal of the Linnean Society 181: 1–20. https://doi.org/10.1111/boj.12385
- [5] Erdtman G. (1969). Handbook of Palynology Morphology, Taxonomy, Ecology. An Introduction to the Study of Pollen Grains and Spores. Hafner Pub. New York.
- [6] Woodehouse RP. (1935). Pollen grains, their structure, identification and significance in medicine. New York: Hafner, New York, 574.
- [7] Kuprianova A. (1967). Apertures of pollen grains and their evolution in Angiosperms. Paleobot. Playnology, 3: 73–80.
- [8] Aytuğ B, Aykut S, Merev N, Edis G. (1971). İstanbul Çevresi Bitkilerinin Polen Atlası. İ. Ü. Yayın No:1650, O.F. Yayın no:174.
- [9] Charpin J, Surinyach R, Frankland AW. (1974). Atlas of European allergenic pollens. Sandoz Editions, Paris, pp. 20–23.
- [10] Walker JW. (1974). Evolution of exine structure in the pollen of primitive Angiosperms. Am J Bot 61: 891–902.
- [11] Walker JW. (1974). Aperture evolution in the pollen of primitive Angiosperms. Am J Bot 61: 1112– 1137.