

Palynological description of two species of “Shebenik” National Park, Albania

KALLAJXHIU N.

*Department of Biology, FNS
University of Elbasan “Aleksandër Xhuvani”, Elbasan, Albania*

KAPIDANI G.

Department of Biology, FNS, University of Tirana, Tirana, Albania

NAQELLARI P.

TURKU S.

GJETA E.

*Department of Biology, FNS
University of Elbasan “Aleksandër Xhuvani”, Elbasan, Albania*

DAUTI A.

*Department of Chemistry, FNS
University of Elbasan “Aleksandër Xhuvani”, Elbasan, Albania*

Abstract

In this study is presented for the first time in Albanian literature the palynomorphological description of two species found in one of the most important parks in terms of science, economy, history and culture of Albania, “Shebenik” National Park.

*The plants studied are *Daphne mezereum* (Thymelaeaceae) and *Stellaria holostea* (Caryophyllaceae). The main palynomorphological features are defined for each of them. Parameters measured in this study were the types of pollen sizes, pollen shape, aperture characteristic, number of pores and ornamentation type of exine.*

*The results showed that aperture characteristic was pantoporate for both species studied. The ornamentation type of exine was microechinate-microperforate patterns in *Stellaria holostea* and reticulate in *Daphne mezereum*.*

*The study also presents the comparison of the palynomorphological features of *Stellaria holostea* with those of *Stellaria media*, thus determining the differences and similarities between them in size and palynological features.*

Keywords: pollen, pantoporate, *Daphne mezereum*, *Stellaria holostea*, “Shebenik” National Park, Albania

INTRODUCTION

Thymelaeaceae family, according to Flora of Albania includes two genera: *Daphne* L. and *Thymelaea* Scop. There are 6 species in the genus *Daphne* L. -

Daphne mezereum L., *D. gnidium* L., *D. laureola* L., *D. oleoides* Schreber, *D. blagayana* Freyer and *D. cneorum* L. (Qosja *et al.*, 1992).

According to European Flora (Tutin *et al.*, 1968) this family also has the genus *Diarthron* Turcz. and the *Daphne* genus includes about 17 species and 2 subspecies.

The plants of this family are small shrubs, rarely herbs, with simple, entire, usually alternate and exstipulate leaves. Flowers are hermaphrodite or unisexual, regular, 4-merous, usually in small heads or clusters, rarely in racemes or panicles. Sepals are often petaloid, arising from the rim of a tubular, campanulate or urceolate hypanthium, usually similar in colour and texture to the sepals. Petals absent in European genera. Stamens are 8, inserted in 2 whorls on the wall of the hypanthium; filaments short. Ovary is superior, at the base of the hypanthium but free from it, with a single pendent ovule; style is terminal or somewhat lateral. Fruits are nut or drupe.

Species of the *Daphne* genus have leaves often clustered at the ends of the branches. Flowers are hermaphrodite, usually fragrant, in terminal heads or axillary spikes or clusters, rarely in terminal panicles. Hypanthium is tubular or narrowly campanulate; sepals and hypanthium are petaloid; style is terminal. Fruit is a drupe, exposed at maturity; exocarp is succulent, rarely coriaceous (Tutin *et al.*, 1968).

Caryophyllaceae family has a large number of one, two or perennial plants. According to the Flora of Albania (Paparisto *et al.*, 1988), it includes about 27 genus (including *Stellaria* genus, 159 species and 68 subspecies).

The *Stellaria* genus has the following species: *Stellaria nemorum*, *S. media*, *S. neglecta*, *S. pallida* and *S. graminea*. Studies conducted by foreign authors have shown that in Albania, in this genus there are other species that are not included in this flora, such are *Stellaria holostea* and *Stellaria uliginosa* (Barina *et al.*, 2010; Barina *et al.*, 2017).

The flowers of this genus are regular, usually hermaphrodite and have a capsule-type fruit.

According to European Flora, this family has 37 genus and the genus *Stellaria* includes about 17 species and 5 subspecies. The plants are herbs, more rarely small shrubs. Leaves usually are opposite and decussate, more rarely alternate or verticillate, simple, entire, with or without scarious stipules. Flowers are actinomorphic, usually hermaphrodite, often in bracteate dichasia. Sepals are 4-5 and free, or fused and often united by scarious strips of tissue (commissures) alternating with the calyx-teeth. Petals are (0)4-5, free and stamens are usually 8-10, obdiplostemonous. Ovary is superior, unilocular at least above, with 1 to numerous campylotropous ovules on a basal or free-central placenta; stigmas (1)2-5. Fruit is usually a capsule, dehiscing with teeth equalling the styles in number or twice as many; more rarely fruit a berry or achene (Tutin *et al.*, 1964).

Pollen morphology is a very important characteristic that helps in the systematic of different plants. There are many studies in the field of palynology for the plants of these two families. They have shown that the pollen grains of Thymelaeaceae and Caryophyllaceae species are pantoporate, where the number of pores can vary within the family and genus. There may be variations in the size of the pollen grain, diameter of the pore, the thickness and ornamentation of exine layers (Erdtman, 1961; Chanda, 1962; Nowicke *et al.*, 1985; Punt & Hoen, 1995; Bredenkamp & Wyk V., 1996; Beyers & Marais, 1998; Yildiz, 2001; Herber, 2002; Yildiz, 2006; Perveen & Qaiser, 2006; Chengyu & Liu, 2017; Khodayari *et al.*, 2017; Ullah *et al.*, 2018).

The aim of this study was to identify the palynomorphological features of the pollen grains of the species *Daphne mezereum* and *Stellaria holostea* as well as to determine the differences and similarities with the features of other species of their genera studied by other local and foreign authors.

MATERIAL AND METHODS

The study material was obtained fresh during field expeditions by various plant individuals, in "Shebenik" National Park, in Ruen of Rrajca, in Albania on 13. 05. 2022 and the determination of plants was made by the botanist Dr. GJETA Ermelinda, based on (Barina *et al.* (2017).

"Shebenik" National Park is declared in 2008 and it is one of the newest national parks in Albania. It is located in Librazhd District in the Elbasan Region, on the border with Northern Macedonia and has a total area of 34 507.9 ha, where with DCM no. 59 dated 26.01.2022 to the previous surface of 33 927.66 ha was added 589 ha of new surface. The Park ranges in altitude from 300 to 2,200 m above sea level and includes a diversity of climatic conditions, geological types, landscapes, habitats and plant and animal species. "Shebenik" National Park represents an important scientific, economic, historical and cultural role etc.

During the expedition, flowers were carefully taken from individuals of different populations. To carry out the palynological study, 4-5 microscope slides have been prepared for each of them. Two analytical methods have been used for the preparation of pollen slides: acetolysis according to Erdtman (1960) and basic fuchsine method according to Smoljaninova & Gollubkova (1953).

For each species, measurements of 31 pollen grains were made. Are defined pollen diameter, pores diameter, distance between two pori and exine thickness. Also, ornamentation types of exine are defined.

Terminology followed here is that of Erdtman (1969), Faegri & Iversen (1975), Moore *et al.*, (1997) and Kapidani (2004). For conducting the study, a

Biological Microscope (Motic BA310 Series LED-Digital) was used, with 400 x magnification. The study was accompanied with photos of species in their habitat and microscopic photographs of pollen grains in the different views.

This study is a novelty in the Albanian literature as these species have been studied for the first time from the palynomorphological point of view in Albania.

RESULTS AND DISCUSSION

Morphopalinological description

Family: **Thymelaeaceae**

Genus: **Daphne L.**

Species: ***Daphne mezereum* L.- Spurge laurel**

Daphne mezereum L. is a nanophanerophyte plant and it is a deciduous shrub 25-200 cm, of bushy habit, with erect or ascending, greyishbrown branches; young shoots hirsute. Leaves reached up 30-80 x 8-25 mm and are oblong-lanceolate, glabrous or ciliate, thin, narrowed to a short petiole. Flowers are pinkish-purple, very fragrant, appearing before or with the leaves, borne in clusters of 2-4 in the axils of the fallen leaves of the previous year, forming intercalary spikes. Hypanthium is 5-8 mm, villous; sepals villous beneath, glabrous above, slightly shorter than the hypanthium. Drupes are exposed before maturity, somewhat calcicole. Most of Europe except the extreme west, south and north (Tutin *et al.*, 1968). *Daphne mezereum* is known for attracting bees. It has nectar/pollen rich flowers.



Fig. 1 *Daphne mezereum* (in its habitat)

Pollen grains of *Daphne mezereum* are monade with spheroidal shape and pollen class is pantoporate. Outline in polar view is circular. The diameter of the pollen grains varies from (27.5-30.25) 26.40 μm . The pollen has a crotonoid tectum. The number of pores is greater than 6. The pore diameter is approximately 3.5 μm . The thickness of exine is about 2.25 μm and exine ornamentation was reticulate.

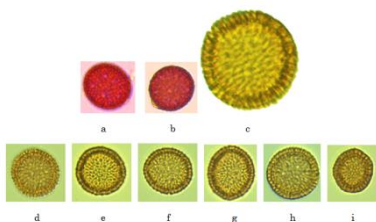


Fig. 2 *Daphne mezereum* pollen grains

a, b pollen grains of *Daphne mezereum* (basic fuchsin method, 400 x magnification); c. pollen in polar view (acetolysis method, 1000 x magnification); d, e, f, g, h, i pollen in different views (acetolysis method, 400 x magnification) (photo: Kallajxhiu N.)

The pollen grains of *D. mezereum* are very similar palynologically to those of other species of the genus *Daphne* but may vary in size. Based on their size, it turns out that pollen grains are included in the group of medium-sized grains (25-50) μm (Kapidani, 2004).

Family: **Caryophyllaceae Juss.**

Genus: **Stellaria L.**

Species: ***Stellaria holostea* L. - Greater Stitchwort**

Stellaria holostea is a perennial herbaceous flowering plant where stems up to 60 cm, ascending, weak, diffuse, sharply quadrangular, usually rough. Its leaves reach up to 30-80 mm, lanceolate-acuminate, rough on margins and on midrib beneath.

Inflorescence lax and its flowers reach up to 15-30 mm in diameter. Calyx has sepals that reach up to 6-9 mm; petals up to twice as long as sepals, bifid to about half-way, rarely absent. Seeds are reddish brown and reached up 2-3 mm diameter (Tutin, 1964).

Greater Stitchwort can be found in northern and central mainland European countries, too. It is one of the most abundant of spring flowers, most common in hedgerows and beside woodland paths and rides. *Stellaria*, the genus name, means star-like. The specific epithet *holostea* comes from the Greek *holosteon* (www.first-nature.com/flowers/stellaria-holostea.php).



Fig. 3 *Stellaria holostea* in its habitat

Pollen grains of *Stellaria holostea* are monade, pantoporate and the shape of pollen is spheroidal. The outline in polar view is circular. Their pores are

evenly distributed throughout the surface of the pollen grain and their number reaches up to 12.

The pores are circular and with an operculum granulate. The diameter of the pore is approximately 5.35 μm and mesopodium, the distance between two pores is about 20 μm .

The pollen grain has an exine where the tectum is microperforate-microechinate and its thickness reaches up to 5.24 μm . Ectexine is thicker than endexine. The diameter of the pollen grain varies from 40-42.5 (41.73) μm .

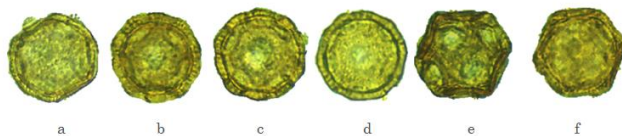


Fig. 4 *Stellaria holostea* pollen grains in different views (acetolysis method, 400 x magnification)
(photo: Kallajxhiu N.)

To determine whether or not there are differences and similarities in the palynomorphological features of *Stellaria holostea* pollen grains, their comparison with the features and dimensions of *Stellaria media* pollen grains was made, studied by Kallajxhiu (2011). The compared data are discarded in Table 2.

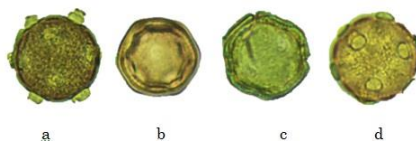


Fig. 5 *Stellaria media* pollen grains, in polar and equatorial view, 400 x magnification
(photo: Kallajxhiu N.)

Table 2. Pollen grains dimensions of *Stellaria holostea* and *Stellaria media*

Pollen grain features	Minimum <i>Stellaria holostea</i>	Minimum <i>Stellaria media</i>	Maximum <i>Stellaria holostea</i>	Maximum <i>Stellaria media</i>	Average <i>Stellaria holostea</i>	Average <i>Stellaria media</i>
Pollen diameter (μm)	40	29.38	42.5	36.31	41.73	32.54

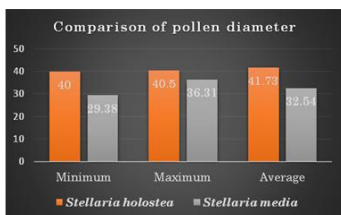


Fig. 6 Comparison data of pollen grains diameter of two species (Stellaria genus)

From Fig. 6 it is clear that the pollen grains of *Stellaria holostea* have a larger diameter than those of *Stellaria media* in the minimum, maximum and average dimensions. It varies from 40-42.5 (41.73) μm thus being classified for the “size” feature of medium pollen grains (Kapidani G., 1996).

Table 3 shows the comparative dimensions of the thickness of the exine, mesopodium, pollen class and exine ornamentation. The data show that in both species of the genus *Stellaria*, the pollen class is pantoporate and the tectum of exine in *Stellaria holostea* is microperforate-microechinate while in *Stellaria media* it is spinulose-punctate.

The exine layer of *S. holostea* is slightly thicker than that of *S. media* (5.24 μm). Also, the diameter of its pores is smaller (5.35 μm) than that of the pores of *S. media* (6.17 μm) and the distance between two pores (mesopodium) of *S. holostea* is approximately 20 μm .

Table 3. Comparison of thickness and ornamentation exine

Palynological of features	<i>Stellaria holostea</i>	<i>Stellaria media</i>	<i>Stellaria holostea</i>	<i>Stellaria media</i>
Pore diameter (μm)	5.35	6.17		
Exine thickness (μm)	5.24	4.39		
Mesopodium (μm)	20	17.5		
Pollen class	pantoporate	pantoporate		
Pollen ornamentation			microperforate-microechinate	spinulose-punctate

CONCLUSIONS

The pollen grains morphology and morphometry in two species studied in Albania (*Daphne mezereum* and *Stellaria holostea*) have various characteristics.

Based on the pollen size, *Stellaria* pollen is medium (25-50 μm) and based on size comparison, *Stellaria holostea* has larger pollen grains in all characteristics than *Stellaria media*, except pore diameter.

The apertures characteristic of the pollen grains was pantoporate in both species studied, where the pores were evenly distributed over the entire

surface of the pollen grain and the exine ornamentation was a microperforate-microechinate in *Stellaria holostea* and reticulate in *Daphne mezereum*.

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