



Palynological Features of *Iris sintenisii* (Iridaceae)

N. Kallajxhiu^{1*}, G. Kapidani², P. Naqellari¹, B. Pupuleku¹, S. Turku¹, A. Dauti¹
and A. Jançe³

¹Department of Biology, FNS, "A. Xhuvani" University of Elbasan, Albania.

²Department of Biology, FNS, University of Tirana, Albania.

³Albanian University, Tirana, Albania.

Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IJPSS/2016/26258

Editor(s):

(1) Genlou Sun, Professor, Biology Department, Saint Mary's University, 923 Robie Street, Halifax, Nova Scotia, B3H 3C3, Canada.

(2) Scd Victoria Anatolyivna Tsygankova, Department for Chemistry of Bioactive Nitrogen-Containing Heterocyclic Compounds, Institute of Bio-organic Chemistry and Petrochemistry, Plant Physiology, NAS, Ukraine.

Reviewers:

(1) Alejandra Leal, Universidad Simón Bolívar Edo. Miranda, Venezuela.

(2) Anonymous, Oswego State University of New York, USA.

(3) Ortrud Monika Barth, Instituto Oswaldo Cruz, Fiocruz, Rio de Janeiro, Brazil.

(4) Paola Ubiergo Corvalán, Universidad Autónoma de Chiapas, México.

(5) Anonymous, Universidade Estadual de Feira de Santana, Brazil.

(6) Richa Pandey, GIPL, Gujarat Forest Department, Gujarat, India.

Complete Peer review History: <http://sciencedomain.org/review-history/15346>

Short Research Article

Received 7th April 2016
Accepted 1st July 2016
Published 9th July 2016

ABSTRACT

This study is a palynological description of the pollen grains of *Iris sintenisii*, Iridaceae. The pollen samples were collected in the hills of Krasta next to Elbasan city, Albania. The palynological features of the pollen grains of this specie were studied for the first time and were compared with that of *Iris germanica*. The slides were prepared using two methods: Acetolysis (according to Erdtman, 1960) and basic fuchsin method (according Smoljaninova & Gollubkova, 1953). Important morphological features of the pollen grains as symmetry, form, size, apertures and also the thickness of exine were examined. The results have shown that the pollen grains of *Iris sintenisii* are smaller than the pollen grains of *Iris germanica*. The thickness of the exine of *Iris sintenisii* is thinner than the exine of pollen grains of *Iris germanica*.

*Corresponding author: E-mail: kallajxhiunikoleta@hotmail.com;

Keywords: *Iris sintenisii*; pollen grains; pollen morphology; basic fuchsine; acetolysis method.

1. INTRODUCTION

Iris is a genus of 260–300 species of flowering plants with showy flowers. It takes its name from the Greek word for a rainbow, which is also the name for the Greek goddess of the rainbow, Iris. Some authors stated that the name refers to the wide variety of flower colors found among the many species [1]. The *Iridaceae* Family is represented by six genera in the Flora of Albania. These are: *Hermodactylis* Miller, *Iris* L., *Gynandriris* Parl., *Crocus* L., *Romulea* Maratti and *Gladiolus* L. [2]. Six species of *Iris* occur in our flora. They are: *Iris germanica*, *Iris pseudacorus*, *Iris pumila*, *Iris pallida*, *Iris aphylla* and *Iris sintenisii* [2]. For some of these species, the palynological features were studied [3-10,8,11].

Elbasan city is located in the center of Albania, Krasta region, and lies down in the valley of the Shkumbin river. It is surrounded by low hills planted mainly with olives and pines. The climate of Elbasan area is a typical Mediterranean, with an average annual temperature of 15-16°C. The cold winter is associated to the characteristic wind of Krasta, blowing from the east through the valley of the Shkumbin river [12].

The palynomorphological features of pollen grains of *Iris sintenisii* were studied for the first time in the present paper. The aim of this study is to investigate palynomorphological similarities and differences in front of *Iris germanica*, as the only species studied in Albania [3]. The pollen grain morphology of *Iris germanica* was presented by Kapidani, G. [3]. The author has used acetolysis method (according to Erdtman,

1960) and basic fuchsine method (according Smoljaninova & Gollubkova, 1953) [3]. Summary of details of *Iris germanica* pollen grain morphology are presented in the Table 1 and Table 2.

2. MATERIALS AND METHODS

Iris sintenisii is an herbaceous perennial plant with an erect stem welling up from a rhizome. It grows in grassy places, scrub forests and blooms in the period of May to June [2].

Samples of *Iris sintenisii* flowers and pollen grains were collected during an educational tour realized with students in the habitat of Krasta (Elbasan) in Albania and were taken from different individuals in the same habitat (Fig. 1). The “Flora of Albania” of Vangjeli et al. [2] was used for identification of this species. Care has been taken during the preparation of slides in order to remove all the waste of the anthers.

Two analytical methods were applied to pollen grain preparation: The acetolysis method according Erdtman [13], and the Basic fuchsine method according Smoljaninova & Gollubkova [14]. Three to five microscope slides were prepared for pollen grain study and using the two methodologies. Micrographs of pollen grains in polar and equatorial views were presented with magnification of 400 x, taken by using the Digital Microscope/Camera Software, Motic Images Plus 2.0 ML, B₁ Series. Morphological characteristics of 31 pollen grains were evaluated. For each characteristic the minimum, maximum and average values were defined.

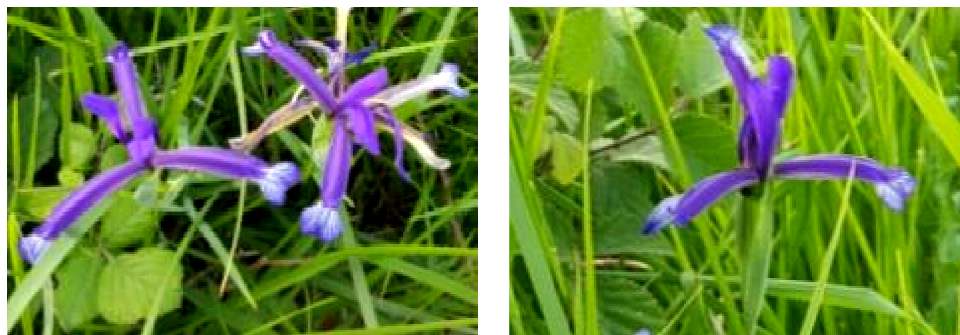


Fig. 1. *Iris sintenisii*

2.1 The Acetolysis Methodology According to Erdtman [13]

The flower buds were kept in ethanol. Anthers were soaked in glacial acetic acid, centrifuged and in sequence submitted to the acetolysis mixture (concentrated acetic anhydride and sulfuric acid in a 9:1 ratio). The mixture was placed in water bath at a temperature of 70-80°C for 5 to 20 seconds. The sediment obtained was washed three to four times with distilled water. Pollen grains were observed under a microscope after adding a drop of a 1:1 glycerin-water mixture. Very dark pollen grains were submitted again in another test-tube by adding 1-2 drops of sodium chloride and 1-2 drops of concentrate sulfuric acid till the material became lighter. After centrifugation the sediment was left for a half hour in 1:1 glycerin-water mixture and centrifuged again. Glycerin-gelatin, prepared according to Kissler [12,14,15], was used to prepare the pollen slides, and sealed with paraffin.

2.2 The Basic Fuchsin Methodology According to Smoljaninova & Golobkova [14]

Solutions of basic fuchsin were prepared according to two variants:

- Basic Fuchsin, alcohol 75% and phenol in the ratio of 1:700:100,
- Basic Fuchsin, ethyl alcohol 96% and xylol in the ratio 1:600:800.

A few drops of 100% ethanol were added to the pollen grains placed on a microscope slide. In case of quick ethanol evaporation, extra drops can be added. Oil spread from the pollen grains was removed with blotting paper from the slide. Then drops of the staining solution were added. Phenol and xylol were used to obtain a better transparency. Glycerin-gelatin was used to prepare the pollen slides also.

The best result was obtained using the acetolysis methodology to study the pollen grain sporoderm. Pollen grains shape, size of apertures and, in some cases, the observation of sculpture elements of the exine could be best analyzed when fuchsin stain was used.

3. RESULTS AND DISCUSSION

3.1 Morphological and Palynological Description

Family: Iridaceae.

Genus: *Iris* L.

Species: *Iris sintenisii* Janka.

The pollen grains of *Iris sintenisii* are monolete, with an elliptical contour when in equatorial view and circular when in polar view (Fig. 2). The aperture may reach the poles. Pollen grain dimensions comprise a polar diameter of 55.26 µm average, and an equatorial diameter of 39.45 µm average (Table 1). The sculpture of the exine is reticulate and the thickness of the exine comprises a little more than two micrometer.

Comparing pollen grain morphology of *Iris sintenisii* and *Iris germanica* [3], similarities of morphological features were observed, but changes in size were identified. Based upon the morphological features, as shown in Table 1, the pollen grains of *Iris sintenisii* are smaller than the pollen grains of *Iris germanica*. Some data are summarized in Tables 1 and 2.

It can clearly seen in Fig. 3 that the pollen grains of *Iris sintenisii* have a thicker exine than the pollen grains of *Iris germanica*. Also, the sculptures of the exine of *Iris sintenisii* are reticulate, whereas the sculpture of the exine of *Iris germanica* is verrucate.



Fig. 2. *Iris sintenisii* pollen grains. a. Polar view, fuchsin stained; b. Polar view, acetolysed; c. Equatorial view, acetolysed. (Obtained at 400x magnification; photo: N. Kallajxhiu)

Table 1. Pollen grain dimensions of *Iris sintenisii* and *Iris germanica*

Pollen grain features	Minimum <i>Iris sintenisii</i>	Minimum <i>Iris germanica</i>	Maximum <i>Iris sintenisii</i>	Maximum <i>Iris germanica</i>	Average <i>Iris sintenisii</i>	Average <i>Iris germanica</i>
Polar diameter (µm)	50.50	56.80	57.96	73.80	55.26	66.10
Equatorial diameter (µm)	37.26	44.00	53.40	61.10	39.45	51.80

Table 2. Comparison of thickness and sculpture of *Iris sintenisii* and *Iris germanica* exines

Palynological features	Average dimension of <i>Iris sintenisii</i>	Average dimension of <i>Iris germanica</i>
Thickness of exine (µm)	2.07	1.50
Sculpture of exine	Reticulate	Verrucate

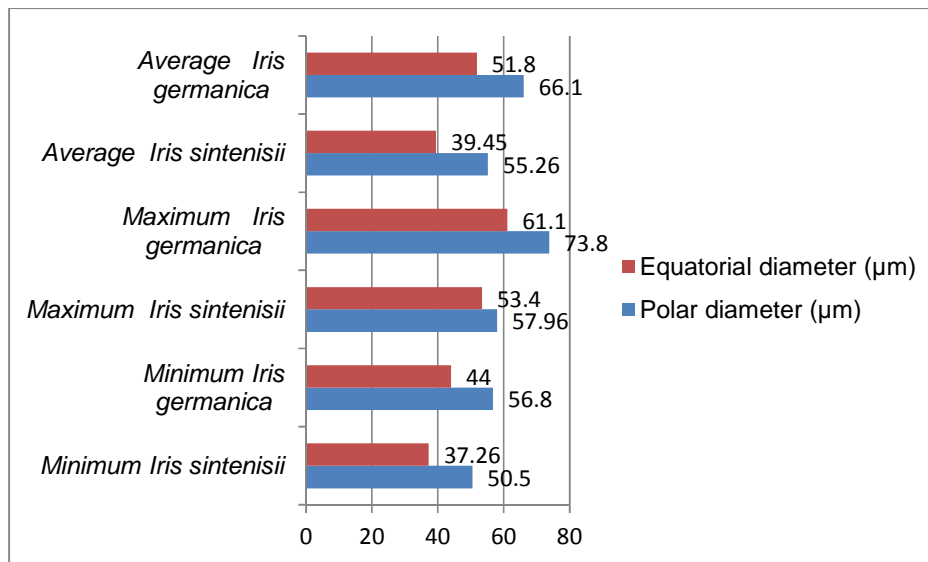


Fig. 3. Graphical presentation of comparative pollen grain size

4. CONCLUSIONS

From the data presented, it was concluded that:

The pollen grains of *Iris sintenisii* have great similarity with the pollen grains of *Iris germanica* regarding the morphological features, in terms of:

- Pollen grains have an elliptical contour in equatorial view and a circular contour in polar view.
- The aperture reaches almost up to the pole.

The differences of the dimensions between the pollen grains are shown below:

- The pollen grains of *Iris sintenisii* are smaller than the pollen grains of *Iris germanica*.

- The exine of *Iris sintenisii* is thicker than the exine of *Iris germanica*.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Manning John, Goldblatt Peter. The *Iris* family: Natural history & classification. Portland, oregon: Timber Press. 2008;200-204. ISBN 0-88192-897-6
2. Vangjeli J, Ruci B, Mullaj A, Paparisto K, Qosja XH. Flora e Shqipërisë. Vëllimi IV.

- Akademia e Shkencave e Republikës së Shqipërisë, Tiranë. 2000;1-502,257-267.
3. Kapidani G. Bazat e palinologjisë. Spore dhe polene të disa bimëve të sotme të Shqipërisë. Seiko, Elbasan. 1996;49.
 4. Kallajxhiu N. Studimi alergopalinologjik i bimëve alergjike të rrethit të Elbasanit dhe i alergjive të shkaktuara prej tyre. Rama Graf. 2011;38-39.
 5. Ducker CS, Knox BR. Pollen and people. Biotechnology and ecology of pollen. Springer, New York. 1985;399-404.
 6. Surova TD, Gumbatov ZJ. Ultrastruktura obovlocka pilcevi zeren Kavkazni prectavitel *Taxus baccata* (Taxaceae). Botanicheskii Zhurnal. 1986;71(7):886-888.
 7. Pupuleku B. Studimi melissopalinologjik i mjaltërave të rajonit Elbasan dhe i poleneve të disa bimëve mjaltore. Ph D, Universiteti i Tiranës; 2002.
 8. Donmez EO, Isik S. Pollen morphology of turkish amaryllidaceae, *Ixioliriaceae* and *Iridaceae*. Grana. 2008;147(1):15-38.
 9. Marcos Vinicius Dantas-Queiroz & Cynthia Fernandes Pinto da Luz. Palynotaxonomy of Iridaceae Juss. From Goias and Tocantins States, Brazil; Braz. J. Bot; 2015.
 10. Božena Mitić, Heidemarie Halbritter, Renata Šoštarić, Toni Nikolić. Pollen morphology of the genus *Iris* L. (Iridaceae) from Croatia and Surrounding area: Taxonomic and phylogenetic implications. Plant Systematics and Evolution, January. 2013;299(1):271-288.
 11. Nur Münevver Pinar, Emel Oybak Dönmez. Pollen morphology of turkish *Iris* L. (Iridaceae) with reference to evolutionary trends at the infrageneric level. Israel Journal of Plant Sciences. 2000;48(2):129-141.
 12. Qirjazi P. Gjeografia fizike e Shqipërisë, Tiranë. 1998;2:115-290.
 13. Erdtman G. The acetolysis method. Svensk Botanicheskii Tidskrift. 1960;54: 561 – 564.
 14. Smoljaninova LA, Gollubkova VF. Metodike issledovani pilci. Doklady Akademii Nauk SSSR T LXXXVIII. 1953; 1:125-126.
 15. Sladkov AN. The introduction to the pollen analysis. Akademia Nauk, Moscow. 1967; 32-75.

© 2016 Kallajxhiu et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:

The peer review history for this paper can be accessed here:
<http://sciencedomain.org/review-history/15346>