POLLEN FLORA OF PAKISTAN -LXI. VIOLACEAE

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Abstract

Pollen morphology of 5 species of the family Violaceae from Pakistan has been examined by light and scanning electron microscope. Pollen grains are usually radially symmetrical, isopolar, colporate, sub-prolate to prolate-spheroidal. Sexine slightly thicker or thinner than nexine. Tectum mostly densely punctate rarely psilate. On the basis of exine pattern two distinct pollen types viz., *Viola pilosa*—type and *Viola stocksii*-type are recognized.

Introduction

Violaceae is a family with 20 genera and about 800 species (Mabberley, 1987). In Pakistan it is represented by one genus and 17 species (Qaiser & Omer, 1985). Plant perennial herbs, or shrubs leaves simple, alternate rarely opposite, flowers bisexual, zygomorphic or actinomorphic, calyx 5, corolla of 5 petals, anterior petal large and spurred. Androecium of 5 stamens. Gynoecium a compound pistil of 3 united carpels, ovules superior, fruit capsule. The family is of little economic importance except for the garden favorite, Violets, Violas and Pansies.

Pollen morphology of the family has been examined by Erdtman (1952), Lobreau-Callen (1977), Moore & Webb (1978) and Dojas *et al.*, (1993). Moore *et al.*, (1991) examined pollen morphology of the genus *Viola*. Kubitzki (2004) examined the pollen morphology of the family Violaceae. There are no reports on pollen morphology of the family Violaceae from Pakistan. Present investigations are based on the pollen morphology of 5 species representing a single genus of the family Violaceae by light and scanning electron microscope.

Materials and Methods

Polleniferous material was obtained from Karachi University Herbarium (KUH) or collected from the field. The list of voucher specimens is deposited in KUH. The pollen grains were prepared for light (LM) by the standard methods described by Erdtman (1952) and scanning microscopy (SEM). For light microscopy, the pollen grains were mounted in unstained glycerin jelly and observations were made with a Nikon Type-2 microscope, under (E40, 0.65) and oil immersion (E100, 1.25), using 10x eye piece. For SEM studies, pollen grains suspended in a drop of water were directly transferred with a fine pipette to a metallic stub using double sided cello tape and coated with gold in a sputtering chamber (Ion-sputter JFC-1100). Coating was restricted to 150 A. The S.E.M examination was carried out on a Jeol microscope JSM-2. The measurements are based on 15-20 readings from each specimen. Polar axis (P) and equatorial diameter (E), aperture size, apocolpium, mesocolpium and exine thickness were measured (Table 1).

The terminology used is in accordance with Erdtman (1952), Kremp (1965), Faegri & Iversen (1964) and Walker & Doyle (1975).

Table 1. General characters of pollen grains found in pollen type Viola pilosa.

Name of species	Shape	Length in µm	Breath in µm	Colpus length in Exine thickness µm	Exine thickness µm	
Viola canescens Wall. ex Roxb.	Sub-Pr	Sub-Pr 30.01 (32.08±0.84) 22.50 (25.0±0.71) 26.25 (28.03±0.80) 1.50 (1.67±0.06) Densely punctate 37.50 27.50 27.50	22.50 (25.0±0.71) 27.50	26.25 (28.03±0.80) 30.10	1.50 (1.67±0.06) 2.11	Densely punctate
Viola fedischenkoana W. Becker	Sub-Pr		23.75 (28.39±1.20) 32.50	32.50 (36.25±1.25) 23.75 (28.39±1.20) 27.50 (31.14±1.17) 1.50 (1.64±0.04) Densely scabrate 40.11 32.50	1.50 (1.64±0.04) 1.75	Densely scabrate
Viola makranica Omer & Qaiser	Pr-Sp		21.25 (23.12±0.80) 25.11	23.75 (24.30±0.36) 21.25 (23.12±0.80) 20.10 (20.31±0.31) 1.50 (1.87±0.12) Densely punctate 25.11 25.11	1.50 (1.87±0.12) 2.10	Densely punctate
Viola pilosa Blume	Pr-Sp		22.50 (25.0±0.60) 27.50	25.0 (27.50±0.70) 22.50 (25.0±0.60) 22.50 (23.27±0.42) 1.25 (1.53±0.08) 30.0 27.50 1.75	1.25 (1.53±0.08) 1.75	Punct-Scab

Pr = Prolate, Pr-Sp = Prolate sheroidal

General pollen characters of the family Violaceae

Pollen grains are usually radially symmetrical, isopolar, tricolporate, sub-prolate to prolate-spheroidal. Sexine slightly thicker or thinner than nexine. Tectum mostly densely punctate rarely subpsilate. On the basis of exine pattern two distinct pollen types recognized viz., *Viola pilosa*—type and *Viola stocksii*-type.

Key to the pollen types

Pollen type: Viola pilosa-type (Fig. 1 A-D).

Pollen class: Tricolporate **P/E ratio:** 105–131

Shape: Prolate-spheroidal and subprolate. **Apertures:** Colpus long sunken with acute ends.

Exine: Sexine thicker than nexine.

Ornamentation: Densely punctate or scabrate–punctate

Measurements: Size: Polar axis P = 31.98 (37.75 \pm 1.2) 40.21, and equatorial diameter E = 21 (38.5 \pm 2.1) 40.5 μ m. Colpi 20.61 (27.93 \pm 1.4) –35.25 μ m long. Exine 1.5-1.75 μ m thick, sexine thicker than nexine. Tectum densely punctate or densely scabrate–punctate.

Species included: *Viola canescens* Wall. ex Roxb., *Viola fedtschenkoana* W. Becker, *V. pilosa* Blume., *Viola makranica* Omer & Qaiser.

Key to the species

1. +	Pollen grains sub-prolate	2
-	Pollen grains prolate-spheroidal	3
2. +	Tectum densely punctate	Viola canescens
-	Tectum scabrate-punctate	Viola fedtschenkoana
	Tectum punctate-scabrate	
-	Tectum densely punctate	Viola makranica

Pollen type: *Viola stocksii*-type Pollen class: Tricolporate

P/E ratio: 1.06

Shape: Prolate-spheroidal.

Apertures: Colpus short sunken with acute ends.

Exine: Sexine thinner than nexine.

Ornamentation: Sub-psilate, densely punctate.

Measurements: Size: Polar axis P(21.7-) 24.45 \pm 0.54 (-29.41) μm and equatorial diameter E (19.61-) 23.01 \pm 0.55 (-28.11) μm, tricolporate, triangular, with apertures on the sides of an angular grain in polar view, colpi (21.21-) 22.61 \pm 0.68 (-28.13) μm long, with vestibuli, colpal membrane sparsely granulated. Mesocolpium (14.23-) 16.78 \pm 0.55 (-19.61) μm. Apocolpium (0.70-) 1.26 \pm 0.14 (-2.41) μm. Exine (1.26-) 1.33 \pm 0.02 (-1.41) thick μm, sexine thinner than nexine. Tectum sub-psilate, densely punctate.

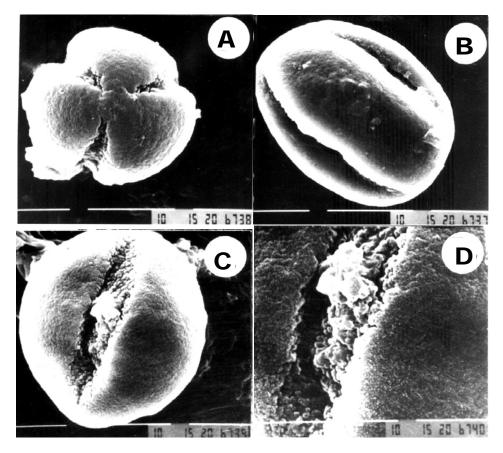


Fig. 1. Scanning Electron micrographs of pollen grains. *Viola canescens* A, Polar view, B, Equatorial view; *Viola fedtschenkoana*: C, Equatorial view; D, Exine pattern. Scale bar = $10 \,\mu m$.

Species included: Viola stocksii Boiss.

Discussion

Violaceae is more or less a stenopalynous family. Pollen grains of this family have similar apertural type i.e., tricolorate pollen. Erdtman (1952) also reported similar pollen in the genus *Viola*. However, it shows little variation in their pollen shape classes and tectum types. Present investigations are based on 5 species representing a single genus i.e., *Viola* L.. On the basis of tectum two distinct pollen types are recognized viz., *Viola pilosa*-type and *Viola stocksii*-type. *Viola pilosa*-type is characterized by densely punctate or punctate–scabrate exine ornamentation. Four species are included in this pollen type. However, these species can easily be seperated on the basis of shape and exine pattern (see key to the species). *Viola* stocksii is easily recognized by having subpsilate to densely punctate tectum. Within the family Violaceae three subfamilies i.e., the Violoideae, the Leonioideae and the Fusispermoideae have been recognized. Cronquist

(1968) kept the family Violaceae in the order Violales. However, Kenneth *et al.*, (2001) and Tokuoka (2008) on the basis of phylogenetic analysis from plastid *rbcL* and *matK* sequences kept the family Violaceae in the order Malpighiales. Pollen data do not support the above phylogenetic treatment because family Violaceae is stenopalynous taxon. Order Malpighiales are quite diverse palynologically, like in the family Polygalaceae has polycolporate pollen (Perveen & Qaiser, 2001), family Linaceae has colporate pollen with baculate tectum (Perveen & Qaiser, 2008).

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(Received for publication 25 February 2008)