

Palynological Studies in Tribe Aveneae (Poaceae) from Potohar of Pakistan

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Abstract

Pollen morphology of four species belonging to three genera of tribe aveneae (Poaceae) was examined by light microscope (LM) and scanning electron microscope (SEM). The study showed that pollen in all species were circular in polar view, however, there are variations in equatorial view of pollen and other quantitative characters that is, polar and equatorial diameter, pore diameter and exine thickness, that are valuable in the identification and differentiation of species. Average pollen fertility in the tribe is 77.37%. Scabrate type of sculpturing is found in all species except *Polypogon monspeliensis* which showed the verrucate type of sculpturing and can be differentiated from *Polypogon fugax* on the basis of its sculpturing pattern. The study revealed that pollen characters are important in the taxonomy of grasses at the specific and generic level and can be useful in delimiting taxa of different tribes.

Key words: Aveneae; Palynological studies; Potohar; Scabrate.

1. Overview of the study

Pollen morphology is study of structure of pollens. Valuable information can be obtained from full and careful study of pollen, leading first to the understanding both of morphology and use of valuable characters of pollen in taxonomy. The present work reports the first detailed palynological studies of 4 different species of tribe Aveneae using LM and SEM. The main aim of the present work is to find the importance of pollen morphology in identification of plants at generic and specific level.

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2. Literature review

The tribe aveneae belongs to sub family Pooideae, having about 65 genera mainly in temperate regions of both hemispheres, extending to mountainous regions of the tropics; out of these 17 genera and 55 species in Pakistan [1]. There are about 700 genera and 11000 species of grasses [2]. This number is 1000 more than the number suggested by [3] i.e. 10000 species. Potohar is 80 square miles north south by 180 square miles east west, lying North of salt range and between the river Jehlum to its East and South east and Indus in the north and North West. Potohar is a large region located between 32°-30° to 34° North Latitude and 71°- 45 ° to 73 ° -45 ° East Longitude. Its total area is 23,160 square kilometer [4]. In this study, from Potohar, 4 species belonging to 3 genera of this tribe are collected. Genus *Polygogon* has two species, while *Avena* and *Phalaris* have one species each. Pollen morphology has proved to be a valuable tool in plant taxonomy. Pollen morphology was not considered in the earlier taxonomic studies. Palynology can be helpful in solving problems related to grass systematics and can provide basis for additional features for identification of plant species [5]. Pollen morphology of grasses has been studied by [6-8]. In this study, both qualitative and quantitative characters of pollen in species of tribe aveneae were studied, to identify and differentiate species at the specific and generic level, as some characters such as pollen grain size and sculpturing pattern are of significance in taxonomy of grasses [9].

3. Method

Present study was conducted in the experimental Taxonomy lab and Herbarium of Quaid-i-Azam University, Islamabad. The research work is confined to palynological studies of 4 species belonging to 3 different genera of tribe aveneae collected from Potohar region of Pakistan.

3.1 Preparation for Light Microscopy

Light microscopy (LM) was used to study pollen morphology and the terminology used is that of [10-11]. Florets were dissected and anthers were placed on the slide with the help of forceps, added a drop of 45% acetic acid and crushed with iron rod. Pollens were acetolysed according to modified method of [10, 12]. Stirred with needle for equal distribution of pollens, placed the cover slip and sealed the slide edges by transparent nail polish. Slides were labeled with their name, locality and voucher number. The slides were kept in wooden slide cases in vertical position. The following pollen parameters were studied under light microscope for pollen morphology; shape in polar and equatorial view, polar diameter, equatorial diameter, P/E ratio, number of pores, Pore diameter and exine thickness.

3.2 Preparation for Scanning Electron Microscopy (SEM)

The anthers were collected from freshly collected specimens. The anthers were crushed to release pollens. The pollen grains were suspended in distilled water on slide. A drop of water containing pollen was transferred to metallic stub. A hair brush was used to prevent clumping of pollen during evaporation and coated with gold in vacuum coater and examined with, a Joel microscope (JSM 1200). The method for SEM pollen preparation was followed after [13].

3.3 Preparation for Pollen fertility

To determine pollen fertility, acetocarmin and glycerin jelly was used by the modified techniques used by [14]. Anthers were squashed in a drop of acetocarmin. Debris was removed gently and cover slip was placed on it. The slides were observed at low magnification (X10). The number of stained and unstained pollen was counted. Fully stained pollen was considered fertile, while unstained and deformed pollen were considered infertile.

4. Findings and discussion

The qualitative and quantitative characters of pollen of 4 species belonging to tribe aveneae mention in Table. 1, are as follows:

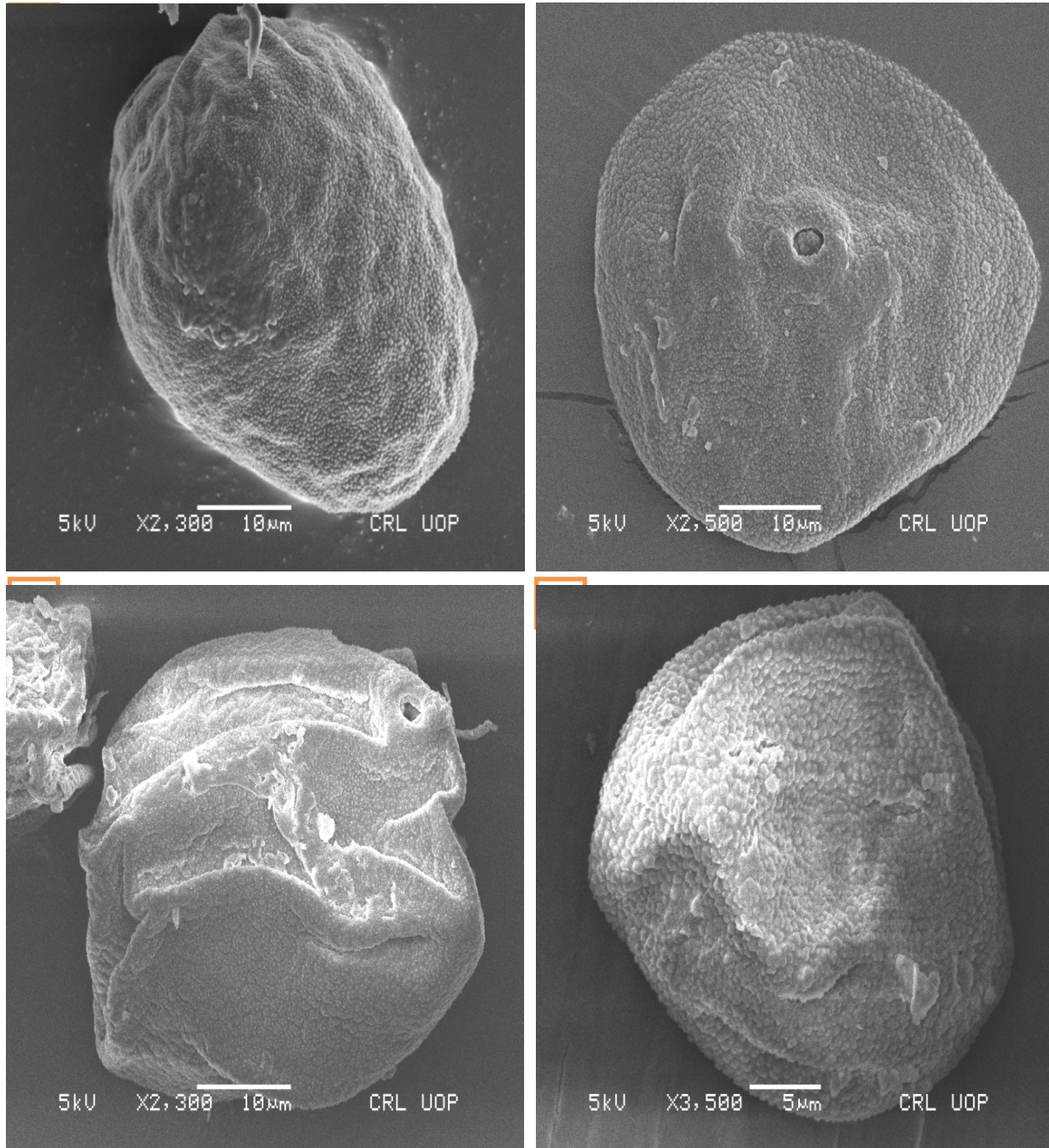


Figure 1
A: Pollen of *Avena sativa* showing scabrate sculpturing, B: *Phalaris minor* pollen with scabrate sculpturing, C: *Polypogon fugax* pollen with scabrate sculpturing, D: Pollen of *P. monspeliensis* showing verrucate sculpturing.

4.1 *Avena sativa* L.

The pollen is circular in polar view and spherical to sub-prolate to prolate in equatorial view; polar axis diameter is 35.12 (33.5- 41.75) μm and equatorial axis diameter 34.37 (27.6-48.75) μm . The P/E ratio is 1.02 while pollen is monoporate and ectoporate. Pollen fertility is 81.36%. The exine thickness is 1.02 μm . The Pore diameter is 1.45 μm (1.23 to 1.65 μm). Sculpturing scabrate and scabrae are widely spaced (Fig. 1A).

4.2 *Phalaris minor* Retz.

Pollen is monad type. Pollen is circular in shape in polar view and spheroidal to sub prolate in equatorial view; polar axis diameter is 34.2 (29.5- 37.5) μm and equatorial axis diameter is 33.52 (20- 35) μm . The P/E ratio is 1.02 while pollen is ectoporate or endoporate and monoporate. The pore diameter is 2.7 (2.4 - 3.2) μm and exine thickness is 1.03 (1.0 - 1.5) μm . The pollen fertility is 72.39%. Sculpturing is scabrate and scabrae are narrowly spaced (Fig. 1B).

4.3 *Polypogon fugax* Nees ex Steud.

Pollen is monad type. Pollen is circular in polar view and spheroid to subprolate or prolate in equatorial view; polar axis diameter is 26.45 (20 - 32.5) μm and equatorial axis diameter is 25.66 (20 - 35) μm . The P/E ratio is 1.02. The exine thickness is 1.13 (0.75 - 1.50) μm , while pollen is exoporate and monoporate. The pollen fertility is 83.30%. Pore diameter is 2.6 (2.2-2.9) μm . Sculpturing scabrate and scabrae are narrowly spaced (Fig. 1C).

4.4 *Polypogon monspeliensis* (Linn.) Desf.

Pollen is monad type. Pollen is circular in polar view and spheroid or prolate in equatorial view; polar axis diameter is 32.25 (27.5 - 34.5) μm and equatorial axis diameter 28.12 (25 - 31) μm . The P/E ratio is 1.14. The exine thickness is 1.1 (0.75 - 1.25) μm , while pollen is monoporate and ectoporate. The pollen fertility is 78.25%. The pore diameter is 2.1 (2.0 - 2.4) μm . Sculpturing is verrucate and verrucae are widely spaced (Fig. 1D).

Table 1. Pollen characters of different species in tribe Aveneae.

Species name	Pv	Ev	Sp	Pad (μm)	Ead (μm)	P/E ratio	Pd (μm)	Et (μm)	Pf (%)
<i>Avena sativa</i>	Circular	sub-prolate to prolate	Scabrate	35.12 (33.5-41.75)	34.37 (27.6-48.75)	1.02	2.6 (2.2-2.9)	1.02	81.36
<i>Phalaris minor</i>	Circular	spheroidal to sub prolate	Scabrate	34.2 (29.5- 37.5)	33.52 (20- 35)	1.02	2.7 (2.4-3.2)	1.03	72.39
<i>Polypogon fugax</i>	Circular	subprolate or prolate	Scabrate	26.45 (20 - 32.5)	25.66 (20 - 35)	1.02	2.6 (2.2-2.9)	1.13	83.30
<i>Polypogon monspeliensis</i>	Circular	spheroid or prolate	Verrucate	32.25 (27.5 - 34.5)	28.12 (25 - 31)	1.14	2.1 (2.0-2.4)	1.10	78.25

Pv: Polar view, Ev: Equatorial view, Sp: Sculpturing pattern, Pad: Polar axis diameter, Ead: Equatorial axis diameter, Pd: Pore diameter, Et: Exine thickness, Pf: Pollen fertility

There is no research work conducted on the pollen morphology of aveneae. Circular nature of pollen is the structural adaptation of grasses for effective pollination by insects [15, 16]. Pollen in *P. monspeliensis* are larger than *P. fugax*, having polar diameter (32.25 μm) and equatorial diameter (28.12 μm) and exine thickness is more in *P. fugax* than *P. monspeliensis*. These variations in size pore diameter and exine thickness serve as a point of differentiation in different species [17]. Pollen is also monoporate and ectoporate in this study, as the number and position of aperture is of prime significance in palynology [18]. Variations are found in equatorial view as spheroidal to oblate spheroidal pollen are found in different species. So these variations observed in qualitative as well as qualitative characters may be helpful in the identification of different species and genera in the tribe. All the species in this tribe showed the scabrate type of sculpturing except *P. monspeliensis* which has verrucate type of sculpturing; hence, sculpturing pattern is an important tool to differentiate *P. monspeliensis* from other species of the genus and tribe. Pollen fertility ranges from 72.39 to 83.30%. The average pollen fertility in the tribe aveneae is 78.82%. Maximum pollen fertility is recorded in *P. fugax*. The degree of fertility of hybrids may give some

indication of the degree of relationship between its parents. In general, hybrids between species of a genus that are not closely related tend to be sterile or of low fertility, whereas hybrids between taxonomically more closely related species or intra-specific taxa tend to be more fertile [19]. Thus, there is a correlation between hybrid fertility and taxonomic relationship. Pollen fertility is a valuable tool in taxonomic studies to distinguish putative hybrids from the parent plants and is also useful to determine the degree of fertility in those plants that are grown under unfavorable conditions [20]. The genetic variations of a flora can be observed by studying their pollen fertility [21]. It is a helpful tool to assess the stability of species in a particular area.

5. Conclusion

This study shows that all the species present in the tribe Aveneae have circular pollen. The variations are valuable in the identification of species. Scabrate type of sculpturing pattern is observed in the tribe except *P. monspeliensis*. Maximum pollen fertility is observed in *P. fugax*, it is concluded from this study that variations exist in qualitative and quantitative characters of pollen, in different species of the tribe and sculpturing pattern in that are helpful in identification, differentiation and delimiting of different taxa.

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