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Pollen Morphological Studies of Selected Plants of Asteraceae Family

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Abstract: The present study comprises pollen morphology and viability estimation that may be utilized as an aid for the taxonomic description of 7 species of family Asteraceae. The species chosen are *Chromolaena odorata* (L.) R.M.king & H.Rob., *Cosmos sulphureus* Cav., *Cyanthillium cinereum* (L.) H.Rob., *Pulicaria vulgaris* Gaetrn., *Tagetes erecta* L., *Tithonia diversifolia* (Hemsl.) A.Gray., *Tridax procumbens* L. Pollen grains were properly collected and used for analysis. Normally, the pollens are tricolporate, echinate and spheroidal. The amount of aperture of these species analyzed ranged from 3 to 4 while the pollen wall normally varied from smooth to spinate. Amongst the seven species colporeous aperture kind was found in *Cosmos*, *Cyanthillium*, *Tagetes*, *Tithonia*, and *Tridax*; whereas porate type in *chromolaena*. The shape of pollen is diverse from oblate-spheroidal to prolate spheroidal. The pollen viability rate ranges from 65 - 86%, which proves that the pollen flora of chosen species is well established. The research concludes that the palynological data have diagnostic value and yet affirm their classification as members of a family.

Keywords: Asteraceae, Aperture, Pollen morphology, Pollen viability, Taxonomy.

I. INTRODUCTION

The pollen is the structure employed in the transportation of the male gamete into the female part of the flower; it is composed of a fine to grainy powder that consists of micro gametophytes (pollen grains), which produce the male gametes of seed plants. Pollen grains come in a huge array of shapes (mostly spherical), dimensions, and surface markings specific to the species. Furrows in the pollen grain known as colpi and pores are important criteria for the identification of pollen types (Dutta., 1964). Asteraceae is one of the first families whose pollen grains were studied (Fisher., 1890). The comprehensive study for most of the genera of this family was using an optical microscope. The Asteraceae family has three types of pollen shapes: Psilate (the pollen grain with a smooth surface), Echinata (Echinae are structured with expansive bases and a sharply pointed apex), or Lophate (Pollen contains lacunae surrounded by ridges). The pollen has important characteristics that make it very valuable in taxonomic studies. These characteristics include the shape and size of the pollen grain, its type of ornamentation on its surface, the presence of pores and colpi as well as the number of grooves and pores and their shapes on the surface of the pollen grain (Erdtman.,1971).

The pollen nucleus is abundant in chromatin material and viable pollen stains pink to deep red with aceto-carmin, while sterile pollen does not take any stain and thus continues almost white and transparent (McKellar and Quesenberry., (1992); Marutani et al., (1993)). The primary objective of this study is to provide palynological information about the Asteraceae family which plays a vital role in analyzing how this palynological information becomes useful to determine their phylogenetic features and the quality and fertility of their pollen grain.

II. MATERIALS AND METHOD

A. Plant Materials

Plant materials were collected from Nirmala College Campus. Flowers and buds were collected separately for pollen studies. All the plant materials used for the study come under the family Asteraceae.

B. Study Area (Fig 1.)



C. Acetolysis (Erdtman., 1969)

The pollen grains were first treated with 70% ethyl alcohol and centrifuged for 5 minutes at 1700 rpm. The supernatant is decanted and the pollen grains are treated with an acetolysis mixture. The acetolysis mixture used for processing pollen grains contains glacial acetic acid and sulphuric acid in the ratio 9:1. The tubes were immersed in a boiling water bath for 3 - 5 min with continuous stirring. The samples were allowed to cool and centrifuged at 1700rpm for 3 minutes and the supernatant was decanted. Distilled water is added to the sample and centrifugation is repeated 3 times. After decanting the pollen grains were stained with acetocarmine.

D. Preparation of Specimens for Microscopic Observation

For Light Microscopic observation, the acetolysed pollen grains were washed with distilled water and added two drops of 0.01 % diluted acetocarmine for two minutes. At least five slides per specimen were made. Then pollen grains were mounted in glycerine. Pollens were observed and photographed under a compound light microscope with a camera

E. Pollen Viability Studies – Acetocarmine Technique (Stanley and Linskens., 1974).

Pollen viability was done by acetocarmine staining using dissecting forceps, scalpel, and needle. Anthers of various species were opened and transferred the pollen dust onto a microscopic cavity slide containing a drop of sucrose solution (Approximate 10 gram of sucrose is mixed with 100 ml of distilled water to make the solution for germination).

Mature anthers were mashed and pollen grains were mixed thoroughly with the aceto-carmine stain. Coverslips were gently placed on different slides for each species.

The slides were then examined under a light microscope. To determine pollen fertility, darkly stained pollen grains were marked as fertile and viable, and unstained or very lightly stained ones were recognized as sterile or non-viable.

Pollen fertility was calculated by dividing the number of viable pollen grains by the total number of grains counted in the field of view and averaging them for all plants in that species. Pollen viability was expressed as the percentage of pollen fertility in each plant species.

III. RESULTS AND DISCUSSION

Pollen architecture has significance in the taxonomy of angiosperms and in revealing inter-relationship among them. The pollen grains of Asteraceae are known to be spherical, slightly flattened or helianthoid. They are principally tricolporate, echinate, and the colpus number and size differs significantly.

In addition, the Asteraceae is considered to be a eurypalynous family and has zonocolporate pollen grains. In the present study a total of seven species belonging to the Asteraceae family were investigated for pollen morphology and viability estimation. It is presented in Table 1.

- 1) *Chromolaena Odorata* (L.) R.M.King & H.Rob: Pollen is tricolporate, shape of pollen is oblate spheroidal, number of aperture is three and aperture type is pirate. Pollen wall is smooth.
- 2) *Cosmos Sulphureus* Cav: Pollen is colporate, shape of pollen is spheroidal, number of aperture is three and aperture type is colporus. Pollen wall is echinate.
- 3) *Cyanthillium Cinereum* (L.) H.Rob: Pollen is tricolporate, shape of pollen is spheroidal, number of aperture is three and aperture type is colporus. Pollen wall is echinate.
- 4) *Pulicaria Vulgaris* Gaertn: Pollen is tricolporate, shape of pollen is spheroidal, number of aperture is three and aperture type is colporus. Pollen wall is echinate.
- 5) *Tagetes Erecta* L: Pollen is tricolporate, shape of pollen is spheroidal, number of aperture varies from three to four and aperture type is colporus. Pollen wall is echinate.
- 6) *Tithonia Diversifolia* (Hemsl.) A.Gray: Pollen is colporate, shape of pollen is prolate spheroidal, number of aperture is three and aperture type is colporus. Pollen wall is spinate.
- 7) *Tridax Procumbens* L: Pollen is tetracolporate, shape of pollen is prolate spheroidal, number of aperture is four and aperture type is colporus. Pollen wall is spinate.

Table: 1. Pollen morphological studies of the selected plant species of Asteraceae family

Species Name	Pollen shape	Pollen aperture	No. of aperture	Pollen wall	Aperture type
<i>Chromolaena odorata</i> (L.) R.M.king & H.Rob.	Oblate Spheroidal	Porate	3	Smooth	Porate
<i>Cosmos sulphureus</i> Cav.	Spheroidal	Colporate	3	Echinate	Colporus
<i>Cyanthillium cinereum</i> (L.) H.Rob.	Spheroidal	Tricolporate	3	Echinate	Colporus
<i>Pulicaria vulgaris</i> Gaetrn.	Spheroidal	Tricolporate	3	Echinate	Colporus
<i>Tagetes erecta</i> L.	Prolate Spheroidal	Tricolporate	3-4	Echinate	Colporus
<i>Tithonia diversifolia</i> (Hemsl.) A.Gray.	Prolate Spheroidal	Colporate	3	Spinate	Colporus
<i>Tridax procumbens</i> L.	Prolate Spheroidal	Tetra colporate	4	Spinate	Colporus

Pollens of the taxa studied are mostly echinate. The pollen aperture was tricolporate in *Cyanthillium cinereum*, *Pulicaria vulgaris*, *Tagetes erecta*, colporate in *Cosmos sulphureus* and *Tithonia diversifolia*. The pollen aperture in *Chromolaena odorata* and *Tridax procumbens* were porate and tertacolporate respectively. The pollen of all members showed colporous type of aperture except *Chromolaena odorata*. The pollen shape was spheroidal in the case of *Cosmos sulphureus*, *Cyanthillium cinereum*, *Pulicaria vulgaris* and oblate spheroidal in *Chromolaena odorata*. The members, like *Tagetes erecta*, *Tithonia diversifolia*, *Tridax procumbens* showed prolate spheroidal pollen. The number of aperture was three in all the members except *Tridax procumbens*.

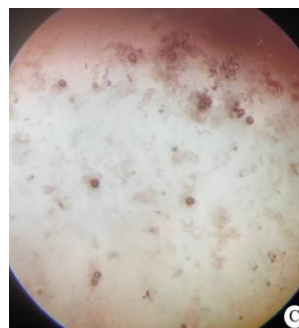
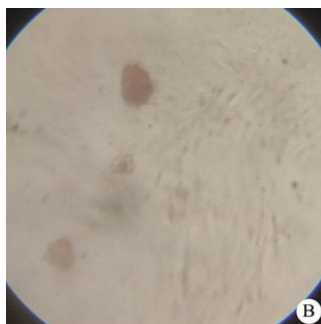
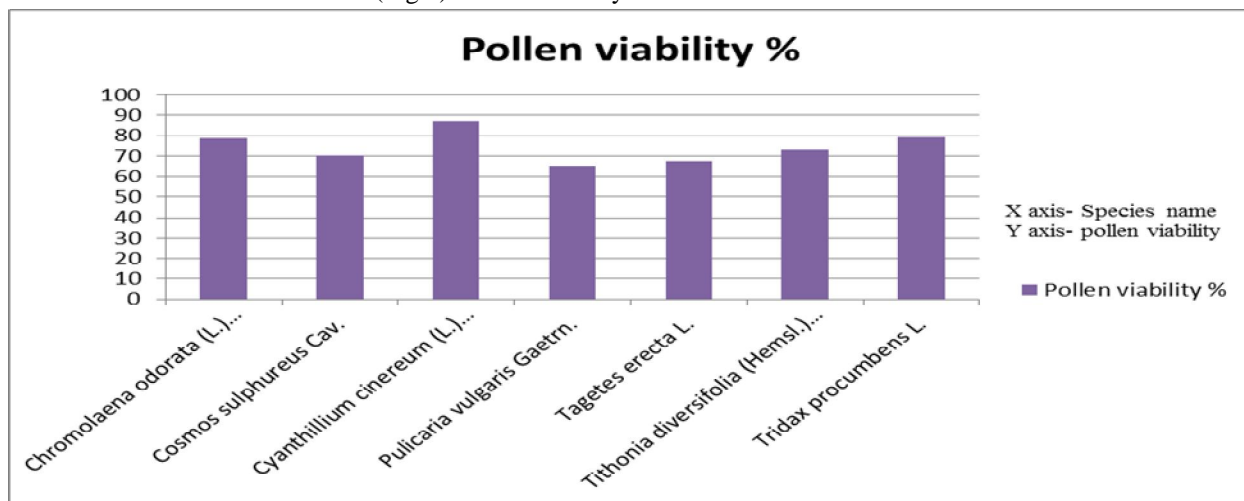
A. Pollen Viability Studies

Results of pollen viability studies are given in Table 2, (Figure 2) of the seven taxa studied, the maximum viability showed in *Cyanthillium cinereum* (87%) followed by *Tridax procumbens* (79.31%), and *Chromolaena odorata* (78.43%). The minimum viability resulted in *Pulicaria vulgaris* (65.2%). The viable pollen stains pink to deep red with aceto-carmin, while sterile pollen does not take any stain and thus continues as almost white and transparent.

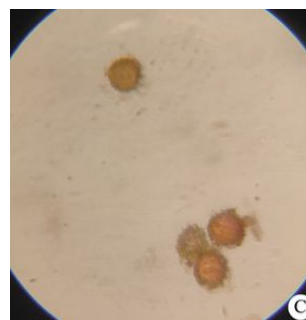
Table: 2. Pollen viability studies of the selected plant species of Asteraceae family

Name of the species	Total no. of pollen grains	Non-viable pollen grain	Viable pollen grains	Pollen viability %
<i>Chromolaena odorata</i> (L.) R.M.king & H.Rob.	102	22	80	78.43
<i>Cosmos sulphureus</i> Cav.	17	5	12	70.05
<i>Cyanthillium cinereum</i> (L.) H.Rob.	30	4	26	87.00
<i>Pulicaria vulgaris</i> Gaetrn.	23	8	15	65.20
<i>Tagetes erecta</i> L.	46	15	31	67.39
<i>Tithonia diversifolia</i> (Hemsl.) A.Gray.	67	18	49	73.13
<i>Tridax procumbens</i> L.	29	6	23	79.31

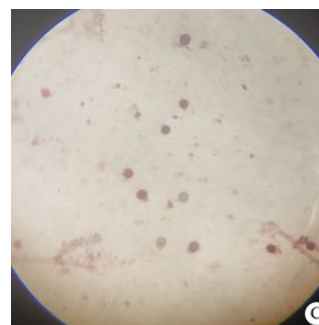
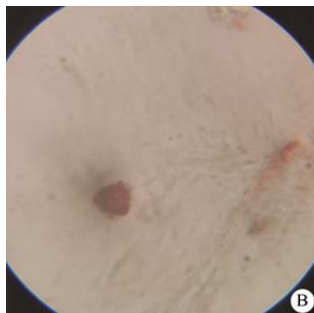
(Fig 2). Pollen viability in Asteraceae members.



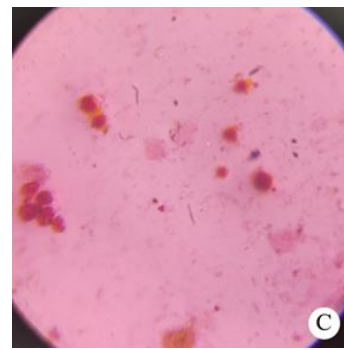
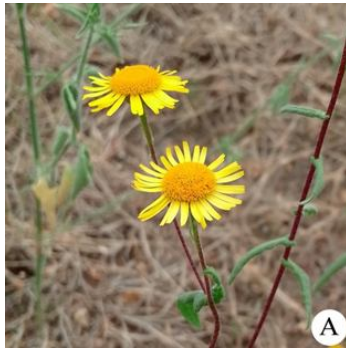
(a) *Chromolaena odorata* (L.) R.M.King & H.Rob. (b) microscopic view of pollen grain (c) pollen grain viability



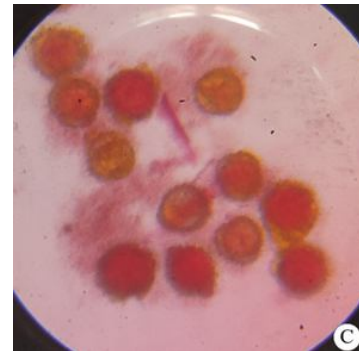
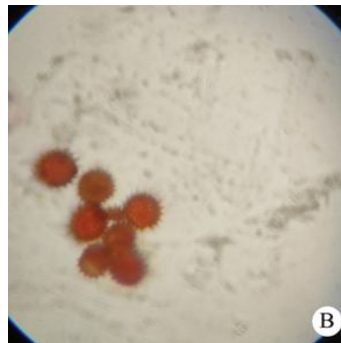
(a) *Cosmos sulphureus* Cav. (b) microscopic view of pollen grain (c) pollen grain viability



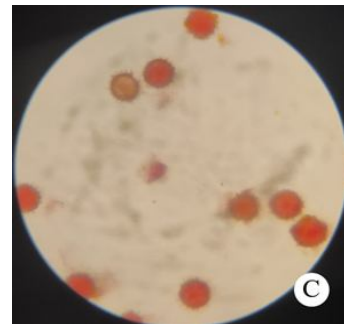
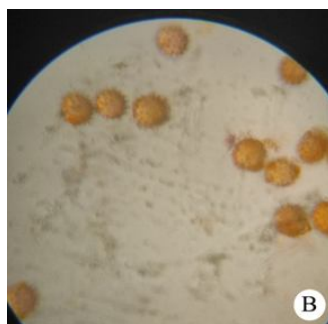
(a) *Cyanthillium cinereum* (L.) H.Rob (b) microscopic view of pollen grain (c) pollen grain viability



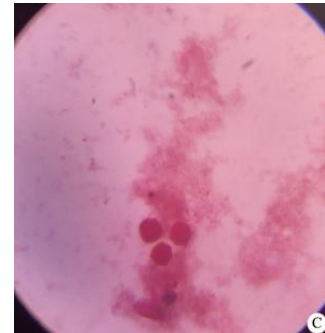
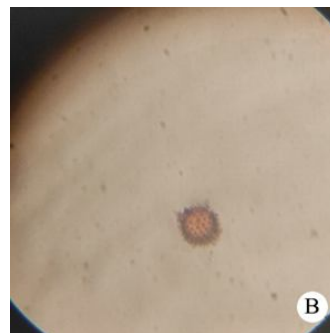
a) *Pulicaria vulgaris* Gaertn. (b) microscopic view of pollen grain (c) pollen grain viability



(a) *Tagetes erecta* L. (b) microscopic view of pollen grain (c) pollen grain viability



a) *Tithonia diversifolia* (Hemsl.) A. Gray (b) microscopic view of pollen grain (c) pollen grain viability



(a) *Tridax procumbens* L. (b) microscopic view of pollen grain (c) pollen grain viability

Pollen grains in the Asteraceae family are comparatively eurypalynous with less variance exhibited in the pollen morphology. Aperture is generally tricolporate to tricolpate occasionally tetracolpate or tetracolporate as discovered in *T. procumbens*. Pollen grains within this family are echinate in ornamentation.

This agrees with all the morphological descriptions of species of Asteraceae from the current study. They may be distinguished from comparable echinate pollen from different families by the comparatively small size of the pollen and also the spines, in addition to the irregular structure of the spines. Although Erdtman (1969) reported in genus *Artemisia* and also some other anemophilous genera of the Asteraceae, the pollen grains are with no apparent spines. Rowley et al., (1981) verified the existence of spines that are brief and difficult to see light microscopy. Within this analysis tricolporate, apertures were detected in *Cyanthillium cinereum*, *Pulicaria vulgaris*, *Tagetes erecta*, and colporate in *Cosmos sulphureus* and *Tithonia diversifolia*. Whereas porate and tertacolporate kind of pollen aperture revealed in *Chromolaena odorata* and *Tridax procumbens* respectively. Pollen viability study revealed the maximum viability detected in *Cyanthillium cinereum* followed by *Tridax procumbens*. The minimal viability is found in *Pulicaria vulgaris*.

IV. CONCLUSION

Implementing the variants in pollen morphology into the species researched showed that species from the family with similar pollen characters tend to be more closely associated and consequently exhibit interspecies relationships indicating reasons for them to maintain precisely the same family while those with distinct pollen characters aren't quite closely related and indicate grounds for them to become distinct species. Pollen fertility rate ranges from 65-86%, which demonstrates that the pollen flora of chosen species is well established..

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