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## Morphopalynological Attributes of Selected Species of Asteraceae and Apocynaceae from District Bannu

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### ABSTRACT

The study was conducted with an aim to investigate the morphology selected species of Asteraceae and Apocynaceae from District Bannu, Pakistan. Palynological research using the standard method of acetolysis have been conducted on the taxa. Pollen slides were mounted and kept as a references material. The pollen grains were measured using light microscopy and then photographed. The qualitative and quantitative pollen features were studied the observation of the current study revealed the highest Polar diameter in *Carthamus tinctorius* (25.5), and lowest *Conyza canadensis* (5). The observation of the current study revealed the highest Equatorial diameter in *Verbesina encelioides* (20), and lowest *Gaillardia pulchella* (5). The observation of the current study revealed the highest P/E ratio in *Gaillardia pulchella* (4), and lowest *Glebionis coronaria* (0.8). The pollen shape of *Carthamus tinctorius*, *Glebionis coronaria*, *Centaurea iberica*, *Ageratum conyzoides* were spheroidal. *Helianthus annuus*, *Verbisina encelioides* were prolate-spheroidal. *Echinops echinatus* were triangular, *Carthamus oxyacantha* were circular while *Tagates erecta*, *Conyza canadensis* and *Parthenium hysterophorus* were spherical. The pollen size of *Carthamus tinctorius*, *Helianthus annuus*, *Gaillardia pulchella*, *Parthenium hysterophorus*, *Verbisina encelioides* were medium and *Glebionis coronaria*, *Centaurea iberica*, *Ageratum conyzoides* were small While *Echinops echinatus*, *Tagates erecta*, *Conyza canadensis*, *Carthamus oxyacantha* were

Online ISSN

3007-3197

Print ISSN

3007-3189

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large. The exine sculpture of *Gaillardia pulchella*, *Carthamus tinctorius*, *Helianthus annuus*, *Glebionis coronaria*, *Echinops echinatus*, *Centaurea iberica*, *Gaillardia oxyacantha*, *Verbisina encelioides*, *Ageratum conyzoides* were Echininate and *Tagates erecta*, *Conyza canadensis* were Psilate while *Parthenium hysterophorus* were Reticulate. The highest Polar diameter was observed in *Vinca rosea* (22.5), and lowest *Cascabela thevetia* (7.5). The highest Equatorial diameter was observed in *Nerium indicum* (25) and *Cascabela thevetia* (10). The highest P/E ratio was observed in *Vinca rosea* (1.28), and *Nerium indicum* (0.6). The pollen shape of *Nerium oleander* were oblate- spheroidal. *Nerium indicum* were spheroidal while *Cascabela thevetia* and *Vinca rosea* were prolate- spheroidal. The pollen size of *Nerium oleander* were medium-large, *Nerium indicum* were medium and *Cascabela thevetia* were large- very large while *Vinca rosea* were large. The exine sculpture of *Nerium oleander* were regulate- psilate, *Nerium indicum* were psilate, *Cascabela thevetia* were reticulate while *Vinca rosea* were perforate. This information are helpful in differentiation among Asteraceous and Apocynaceous taxa.

## Introduction

Palynology, the study of pollen grains and spores, is a vital discipline in plant sciences. The ultrastructural and morphological features of pollen grains provide a wealth of characteristics to identify specific plant taxa, making them invaluable for solving taxonomic problems, determining allergenic pollen (pollinosis), and conducting palynotaxonomical studies. A key attribute of spores and pollen is their microscopic, highly durable nature due to the biopolymer sporopollenin in the outer exine layer, which grants them excellent preservation potential in the geological record. This durability, combined with significant morphological diversity in size, shape, and sculpturing, makes pollen traits highly useful in plant systematics, as they are influenced by strong selective forces involved in reproduction, including pollination, dispersal, and germination. Consequently, palynomorphological investigation using diverse microscopic techniques serves as a significant tool for modern taxonomists in species delimitation and understanding evolutionary relationships (Ahmad et al., 2018; Traverse, 2007; Begum & Mandal, 2016; Ragho, 2020).

The Asteraceae family, one of the largest plant families with an estimated 22,500–

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30,000 species, is a prime subject for palynological study. The family is defined by its unique pseudanthium inflorescence, the capitulum, and is divided into multiple subfamilies and tribes. Palynological data are particularly fruitful for resolving taxonomic issues at specific and generic levels within this vast family. Asteraceae pollen is eurypalynous, exhibiting a great diversity of forms, but is generally isopolar, radially symmetrical, and tricolporate. Exine ornamentation varies from psilate (smooth) to echinate (spiny) and lophate (ridged). The use of advanced techniques like Scanning Electron Microscopy (SEM) is crucial for observing these micro-morphological variations, which are often invisible under light microscopy, allowing for detailed analysis of exine sculpturing and the development of new descriptive terminology. These pollen characteristics are not only essential for delimiting species and understanding phylogenetic relationships but also push the paleobotanical record back, with fossil evidence suggesting the family's first divergences occurred at least 47 million years ago (Funk et al., 2009; Blackmore et al., 2009; Barreda et al., 2010b; Khan et al., 2021).

Similarly, the Apocynaceae family, another large group with over 4600 species, demonstrates the critical importance of pollen morphology, particularly for its complex reproductive biology. The family is subdivided into five subfamilies, with palynological data providing useful information for differentiating them and indicating phylogenetic relationships. A defining palynological feature for the subfamily Asclepiadoideae is the formation of pollinia, where all the pollen grains of a single anther locule are fused into a hard, sac-like structure attached to a translator mechanism for vector transport. This contrasts with the monad grains found in most other angiosperms. The intricate flowers and pollen units (monads, polyads, massulae, or pollinia) are directly correlated with specialized pollination mechanisms. Studying this pollen morphology is therefore fundamental for taxonomy, understanding pollination limitations, and aiding the conservation of endangered medicinal species within the family, as pollen wall architecture and aperture types influence vector trapping, germination success, and overall reproductive fitness (Endress et al., 2014; Rapini, 2012; Walker & Doyle, 1975).

In both families, palynology extends beyond pure taxonomy. The micromorphometric analysis of pollen under Light Microscopy (LM) and SEM is crucial for the accurate

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identification of medicinal plants, the standardization of crude drugs, and the detection of fossil counterparts in sediments for paleoenvironmental reconstruction. The morphological variability of pollen is related to phylogeny and tends to accumulate early in a clade's history. Thus, the study of pollen wall architecture and ornamentation remains an indispensable source of phylogenetic information and has made tremendous contributions to the systematics and phylogeny of angiosperms, providing insight into evolutionary trends and long-term survival strategies of species (Fazal et al., 2013; Wodehouse, 1936; Erdtman, 1952; Mander, 2018).

## Objectives

- 1: To study the pollen morphology of selected species of Asteraceae and Apocynaceae.
2. To compare pollen morphological attributes of each family.

## Literature Review

Palynology, the study of pollen grains and spores, is a fundamental discipline in plant sciences due to the wealth of taxonomic information provided by the ultrastructural and morphological features of pollen. These characteristics are crucial for identifying specific plant taxa, solving taxonomic problems, determining allergenic pollen (pollinosis), and conducting palynotaxonomical studies. A key attribute of spores and pollen is their microscopic and highly durable nature, attributed to the biopolymer sporopollenin in the outer exine layer, which ensures excellent preservation in the geological record. This durability, combined with significant morphological diversity in size, shape, and sculpturing, makes pollen traits highly useful in plant systematics, as they are influenced by strong selective forces involved in reproduction, including pollination, dispersal, and germination. Consequently, palynomorphological investigation using diverse microscopic techniques serves as a significant tool for modern taxonomists in species delimitation and understanding evolutionary relationships (Ahmad et al., 2018; Traverse, 2007; Begum & Mandal, 2016; Ragho, 2020).

Within the large and cosmopolitan Asteraceae family, palynological studies have proven exceptionally valuable for taxonomic delimitation and phylogenetic inference. Research on various genera,

Online ISSN

3007-3197

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including *Chrysolaena*, *Bellis*, *Verbesina*, *Artemisia*, *Inula*, *Tragopogon*, and *Scorzonera*, consistently demonstrates that pollen morphology is a reliable taxonomic marker. Asteraceae pollen is generally characterized as isopolar, radially symmetrical, and tricolporate, with exine ornamentation ranging from echinate to lophate. For instance, studies reveal that *Chrysolaena* is stenopalynous with a characteristic echinolophate pollen type, while variations in size and sculpturing help distinguish species within *Bellis* and *Verbesina*. Advanced techniques like Scanning Electron Microscopy (SEM) are crucial for resolving fine exine details, such as spine length and perforation patterns, which are often invisible under light microscopy. These micro-morphological characters are not only essential for species identification but also for understanding evolutionary trends, as evidenced by fossil records pushing the family's divergence back to at least 47 million years ago (Blackmore et al., 2009; Barreda et al., 2010b; Khan et al., 2021).

Similarly, palynological research in the Apocynaceae family highlights the critical role of pollen morphology in understanding its complex reproductive biology and systematics. The family exhibits a remarkable diversity in pollen units, from monads to complex pollinia, particularly in the subfamily Asclepiadoideae. Studies on taxa from various regions, including Brazil and Thailand, show that pollen grains are porate to colporate, with exine ornamentation varying from psilate to microreticulate. Specific features, such as granules at the pore base in *Odontadenia* or a distinct margo in *Geissospermum*, provide diagnostic value. The evolution from monads to pollinia is a key phylogenetic marker, with *Cryptostegia grandiflora* representing an intermediate form. Research on *Rauvolfia* species further demonstrates that pollen traits like shape, size, and sexine ornamentation are significant for distinguishing closely related species and have implications for conservation and paleoenvironmental studies (Endress et al., 2014; Rapini, 2012; Walker & Doyle, 1975).

The application of palynology extends beyond pure taxonomy into fields like ecology, allergology, and biotechnology. In Asteraceae, airborne pollen studies identify highly allergenic species like *Helianthus annuus* and *Parthenium hysterophorus*, with immunochemical methods pinpointing cross-reactive allergens. For Apocynaceae, pollen viability studies are crucial for crop improvement, with species like *Thevetia peruviana* showing high viability for

Online ISSN

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hybridization. Furthermore, pollination ecology research on *Calotropis procera* reveals how plant density and pollinator pressure interact to regulate reproduction, demonstrating an Allee effect. These applied studies underscore the practical importance of pollen characteristics in managing allergenic species, optimizing agricultural yields, and developing conservation strategies for endangered medicinal plants.

Finally, the palynological studies in Asteraceae and Apocynaceae provide indispensable insights into taxonomic classification, phylogenetic relationships, and evolutionary history. The integration of light and electron microscopy techniques allows for the detailed characterization of pollen features, which are critical for delimiting species and genera. Moreover, the applied aspects of palynology in allergology, pollination ecology, and conservation highlight its broad relevance. Future research should continue to leverage these micro-morphological tools, complemented by molecular data, to refine taxonomic boundaries and enhance our understanding of the ecological and evolutionary dynamics within these diverse plant families.

Online ISSN

3007-3197

Print ISSN

3007-3189

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Methodology

Field Collection and Plant Processing

The study was initiated with extensive field trips across District Bannu, Pakistan, during June to August to collect pollen-producing flora. Key areas, including Bannu Township, Surrani Zakar khel, and Kakki, were targeted for their rich floral diversity. Essential materials, including cutters, zipper bags, a plant press, and a digital camera, were used to collect and document plant specimens. Four duplicates of each species were collected, voucher numbers were assigned in the field, and plants were pressed and dried with frequent newspaper changes to prevent fungal growth. Flowers were separately collected in labeled zipper bags corresponding to their voucher numbers to prevent cross-contamination. Plant identification was validated using authoritative sources such as The Plant List, Flora of Pakistan, and the Herbarium of Pakistan (ISL).

Table 1: List of Selected Taxa of Asteraceae and Apocynaceae from District Bannu-Pakistan

| S.No | Taxon                                   | Family     | Voucher No | Locality           | Date of collection | Vernacular Name | English name     | Status     |
|------|-----------------------------------------|------------|------------|--------------------|--------------------|-----------------|------------------|------------|
| 1.   | <i>Carthamus tinctorius</i> L.          | Asteraceae | TA-01      | Bannu Township     | 03-8-2024          | Kareza          | Safflower        | Cultivated |
| 2.   | <i>Helianthus annuus</i> L.             | Asteraceae | TA-02      | Bannu Township     | 04-8-2024          | Mirgul          | Common Sunflower | Cultivated |
| 3.   | <i>Glebionis coronaria</i> (L.) Tzvelev | Asteraceae | TA-03      | Bannu Township     | 04-8-2024          | Guldaudi        | Crown daisy      | Cultivated |
| 4.   | <i>Echinops echinatus</i> Roxb.         | Asteraceae | MA-04      | Surrani zakar khel | 05-8-2024          | Untkatara       | Usnakantaka      | wild       |

Online ISSN

3007-3197

Print ISSN

3007-3189

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| 5.  | <i>Tagetes erecta</i> L.                                         | Asteraceae | MA-05 | Surrani zakar khel       | 05-8-2024 | Ganda          | African Marigold     | Cultivated     |
| 6.  | <i>Conyza Canadensis</i> (L.) Cronquist                          | Asteraceae | MA-06 | Surrani zakar khel       | 09-8-2024 | Daryai boti    | Horseweed            | Cultivated     |
| 7.  | <i>Centaurea iberica</i> Trevir. ex Spreng.                      | Asteraceae | SB-07 | Kotka feroz Surrani      | 11-7-2024 |                | Iberian star-thistle | Non-Cultivated |
| 8.  | <i>Gaillardia pulchella</i> Foug.                                | Asteraceae | SB-08 | Kotka feroz surrani      | 12-7-2024 | Gul e Namda    | Indian blanketflower | Cultivated     |
| 9.  | <i>Carthamus oxyacantha</i> L.                                   | Asteraceae | MM-09 | Kakki                    | 05-7-2024 | Kasanba Qartam | Wild Safflower       | Wild           |
| 10. | <i>Parthenium hysterophorus</i> L.                               | Asteraceae | MM-10 | Kakki                    | 05-7-2024 | Gajar ghas     | Santa Maria feverfew | Cultivated     |
| 11. | <i>Verbesina encelioides</i> (Cav.) Benth. and Hook.f. ex A.Gray | Asteraceae | SD-11 | Dagan shadi khan surrani | 08-7-2024 | Alo Gulay      | Golden crownbeard    | Non-Cultivated |

Online ISSN

3007-3197

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|-----|-------------------------------------------------------|-------------|-------|-----------------------------------|-----------|------------|--------------------------|------------|
| 12. | <i>Ageratum<br/>conyzoides</i><br>Sieber ex<br>Steud. | Asteraceae  | SD-12 | Dagan<br>shadi<br>khan<br>surrani | 08-7-2024 | Visadodi   | Billygoat<br>weed        | Cultivated |
| 13. | <i>Cascabela<br/>thevetia</i><br>(L.) Lippold         | Apocynaceae | TA-13 | Bannu<br>Township                 | 15-6-2024 | Zyerh gul  | Yellow<br>oleander       | Cultivated |
| 14. | <i>Nerium<br/>indicum</i><br>Mill.                    | Apocynaceae | SB-14 | Kotka<br>feroz<br>surrani         | 20-6-2024 | Speen gul  | White flower<br>oleander | Cultivated |
| 15. | <i>Nerium<br/>oleander</i><br>L.                      | Apocynaceae | MM-15 | Kakki                             | 21-6-2024 | Kaneer     | Oleander                 | Cultivated |
| 16. | <i>Catharanthus<br/>roseus</i><br>(L.) G.Don          | Apocynaceae | SD-16 | Dagan<br>shadi<br>khan<br>surrani | 23-6-2024 | Babozi gul | Madagascar<br>periwinkle | Cultivated |

## Laboratory Processing and Slide Preparation

Collected plant specimens were poisoned using a solution of mercuric chloride and ethanol to preserve them for long-term herbarium storage. For palynological analysis, the Wodehouse (1935) method was employed for slide preparation. Anthers were isolated from dried flowers, placed on a slide, and treated with acetic acid to clear debris. A drop of melted glycerin jelly was added as a mounting medium, and a coverslip was applied. Pollen slides were meticulously labeled with their corresponding field voucher numbers to maintain a clear chain of identification throughout the study.

## Microscopic Examination and Analysis



Calculated using the standard formula ( $P/E \text{ Ratio} = \text{Polar diameter} / \text{Equatorial diameter}$ ). Microphotography was conducted at 40x magnification using a compound microscope equipped with an image analyzer to document the morphological details of the pollen grains for systematic analysis.

## ***Plate no 01: Microscopic analysis of Asteraceous and Apocynaceous taxa***

### **Results**

#### **Quantitative Characteristics of Asteraceous Taxa**

Pollen grains were quantitatively measured using LM. Quantitative characteristics includes P/E ratio, polar and equatorial diameter.

#### **Polar Diameter**

Pollen morphology of the species of family Asteraceae e.g. *Carthamus tinctorius*, *Helianthus annuus*, *Glebionis coronaria*, *Echinops echinatus*, *Tagetes erecte*, *Conyza canadensis*, *Centaurea iberica*, *Gaillardia pulchella*, *Carthamus oxyacantha*, *Parthenium hysterophorus*, *Verbesina encelioides* and *Ageratum conyzoides* was also examined under the light microscope. The observations of the

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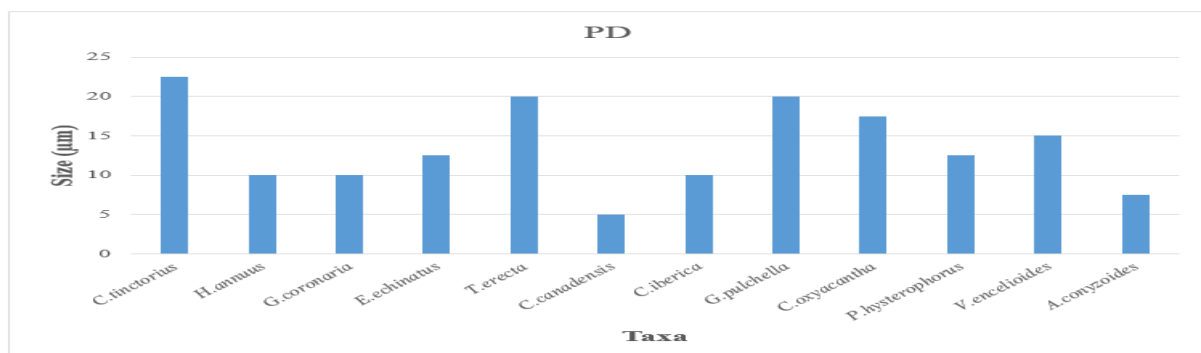
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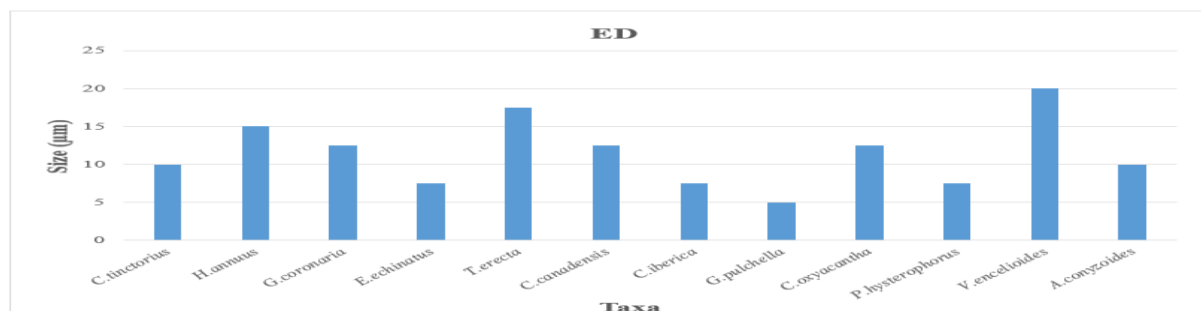
current study revealed the highest PD in *Carthamus tinctorius* (22.5), *Tagatus erecta* (20), *Gaillardia pulchella* (20), *Carthamus oxyacantha* (17.5), *Verbesina encelioides* (15), *Echinops echinatus* (12.5), *Parthenium hysterophorus* (12.5), *Helianthus annuus* (10), *Glebionis coronaria* (10), *Centaurea iberica* (10), *Ageratum conyzoides* (7.5) and *Conyza canadensis* (5).



Graph 01: Polar diameter (µm) of different pollen of family Asteraceae

### Equatorial Diameter

Pollen morphology of the species of family Asteraceae e.g. *Carthamus tinctorius*, *Helianthus annuus*, *Glebionis coronaria*, *Echinops echinatus*, *Tagetes erecte*, *Conyza canadensis*, *Centaurea iberica*, *Gaillardia pulchella*, *Carthamus oxyacantha*, *Parthenium hysterophorus*, *Verbesina encelioides* and *Ageratum conyzoides* was also examined under the light microscope. The observations of the current study revealed the highest ED in *Verbesina encelioides* (20), *Tagetes erecte* (17.5), *Helianthus annuus* (15), *Glebionis coronaria* (12.5), *Conyza canadensis* (12.5), *Carthamus oxyacantha* (12.5), *Carthamus tinctorius* (10), *Ageratum conyzoides* (10), *Echinops echinatus* (7.5), *Centaurea iberica* (7.5), *Parthenium hysterophorus* (7.5) and *Gaillardia pulchella* (5).



Graph 02: Equitorial diameter (µm) of different pollen of family Asteraceae

### P/E Ratio

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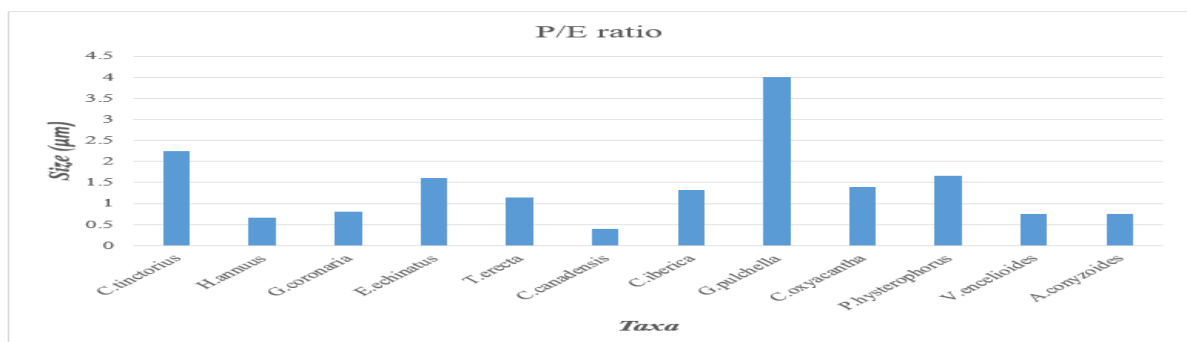
3007-3197

Print ISSN

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Pollen morphology of the species of family Asteraceae e.g. *Carthamus tinctorius*, *Helianthus annuus*, *Glebionis coronaria*, *Echinops echinatus*, *Tagetes erecta*, *Conyza canadensis*, *Centaurea iberica*, *Gaillardia pulchella*, *Carthamus oxyacantha*, *Parthenium hysterophorus*, *Verbesina encelioides* and *Ageratum conyzoides* was also examined under the light microscope. The observations of the current study revealed the highest P/E ratio in *Gaillardia pulchella* (4), *Carthamus tinctorius* (2.25), *Echinops echinatus* (1.6), *Parthenium hysterophorus* (1.66), *Carthamus oxyacantha* (1.4), *Centaurea iberica* (1.33), *Tagetes erecta* (1.14), *Glebionis coronaria* (0.8), *Verbesina encelioides* (0.75), *Ageratum conyzoides* (0.75), *Helianthus annuus* (0.66) and *Glebionis coronaria* (0.8).



Graph 03: P/E ratio (µm) of different pollen of family Asteraceae

### Qualitative Characteristics of Asteraceous Taxa

Pollen grains were qualitatively measured using LM. Qualitative characteristics includes shape, size and exine sculpturing.

#### Shape

The Pollen shape of *Carthamus tinctorius*, *Glebionis coronaria*, *Centaurea*, *Ageratum conyzoides* were spheroidal. *Helianthus annuus*, *Verbisina encelioides* were prolate-spheriodal. *Echinops echinatus* were triangular, *Carthamus oxyacantha* were circular while *Tagates erecta*, *Conyza canadensis* and *Parthenium hysterophorus* were spherical.

#### Size

The pollen size of *Carthamus tinctorius*, *Helianthus annuus*, *Gaillardia pulchella*, *Parthenium hysterophorus*, *Verbisina encelioides* were medium and *Glebionis coronaria*, *Centaurea iberica*, *Ageratum conyzoides* were small while *Echinops echinatus*, *Tagates erecta*, *Conyza canadensis*, *Carthamus oxyacantha* were large.

Online ISSN

3007-3197

Print ISSN

3007-3189

<http://amresearchreview.com/index.php/Journal/about>

## Exine Sculpturing

The exine sculpture of *Carthamus tinctorius*, *Helianthus annuus*, *Glebionis coronaria*, *Echinops echinatus*, *Centaurea iberica*, *Gaillardia pulchella*, *Carthamus oxyacantha*, *Verbisina encelioides*, *Ageratum conyzoides* were Echinate and *Tagates erecta*, *Conyza canadensis* were Psilate while *Parthenium hysterophorus* were Reticulate.

**Table 2: Pollen Characteristics (qualitative) of Asteraceae Taxa**

| S.no | Taxa                            | Pollen shape       | Pollen size | Exine sculpturing |
|------|---------------------------------|--------------------|-------------|-------------------|
| 1    | <i>Carthamus tinctorius</i>     | Spheroidal         | Medium      | Echinate          |
| 2    | <i>Helianthus annuus</i>        | Prolate-spheroidal | Medium      | Echinate          |
| 3    | <i>Glebionis coronaria</i>      | Spheroidal         | Small       | Echinate          |
| 4    | <i>Echinops echinatus</i>       | Triangular         | Large       | Echinate          |
| 5    | <i>Tagates erecta</i>           | Spherical          | Large       | Psilate           |
| 6    | <i>Conyza canadensis</i>        | Spherical          | Large       | Psilate           |
| 7    | <i>Centaurea iberica</i>        | Spheroidal         | Small       | Echinate          |
| 8    | <i>Gaillardia pulchella</i>     | Spheroidal         | Medium      | Echinate          |
| 9    | <i>Carthamus oxyacantha</i>     | Circular           | Large       | Echinate          |
| 10   | <i>Parthenium hysterophorus</i> | Spherical          | Medium      | Reticulate        |
| 11   | <i>Verbisina encelioides</i>    | Prolate-spheroidal | Medium      | Echinate          |
| 12   | <i>Ageratum conyzoides</i>      | Spheroidal         | Small       | Echinate          |

**Table 3: Pollen Characteristics (quantitative) of Asteraceae Taxa**

| S.no | Taxa | Polar diameter | Equatorial | P/E ratio |
|------|------|----------------|------------|-----------|
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Online ISSN

3007-3197

Print ISSN

3007-3189

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|    |                                     |      | diameter |      |
|----|-------------------------------------|------|----------|------|
| 1  | <i>Carthamus tinctorius</i>         | 22.5 | 10       | 2.25 |
| 2  | <i>Helianthus annuus</i>            | 10   | 15       | 0.66 |
| 3  | <i>Glebionis coronaria</i>          | 10   | 12.5     | 0.8  |
| 4  | <i>Echinops echinatus</i>           | 12.5 | 7.5      | 1.6  |
| 5  | <i>Tagetes erecta</i>               | 20   | 17.5     | 1.14 |
| 6  | <i>Conyza canadensis</i>            | 5    | 12.5     | 0.4  |
| 7  | <i>Centaurea iberica</i>            | 10   | 7.5      | 1.33 |
| 8  | <i>Gaillardia pulchella</i>         | 20   | 5        | 4    |
| 9  | <i>Carthamus oxyacantha</i>         | 17.5 | 12.5     | 1.4  |
| 10 | <i>Parthenium<br/>hysterophorus</i> | 12.5 | 7.5      | 1.66 |
| 11 | <i>Verbesina encelioides</i>        | 15   | 20       | 0.75 |
| 12 | <i>Ageratum conyzoides</i>          | 7.5  | 10       | 0.75 |

## Quantitative Characteristics of Apocynaceous Taxa

### Polar Diameter

The pollen morphology of Apocynaceae taxa e.g *Nerium oleander*, *Nerium indicum*, *Cascabela thevetia* and *Vinca rosea* was examined. The highest PD was observed in *Vinca rosea* (22.5), *Nerium indicum* (15), *Nerium oleander* (10) and *Cascabela thevetia* (7.5).

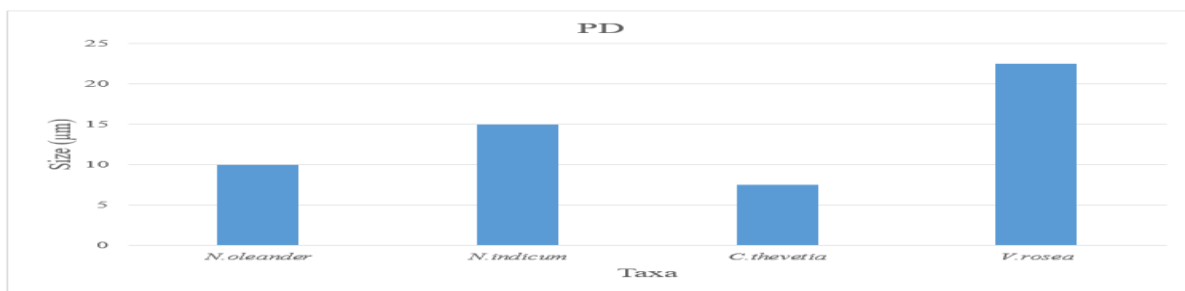
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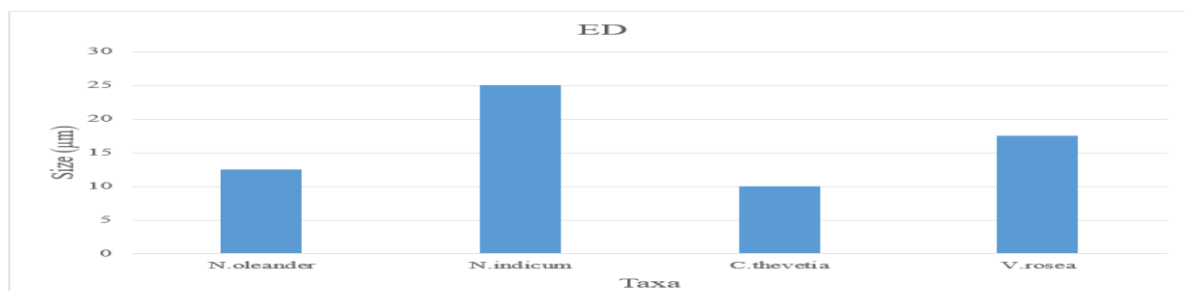
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Graph 01: Polar diameter (μm) of different pollen of family Apocynaceae

## Equatorial Diameter

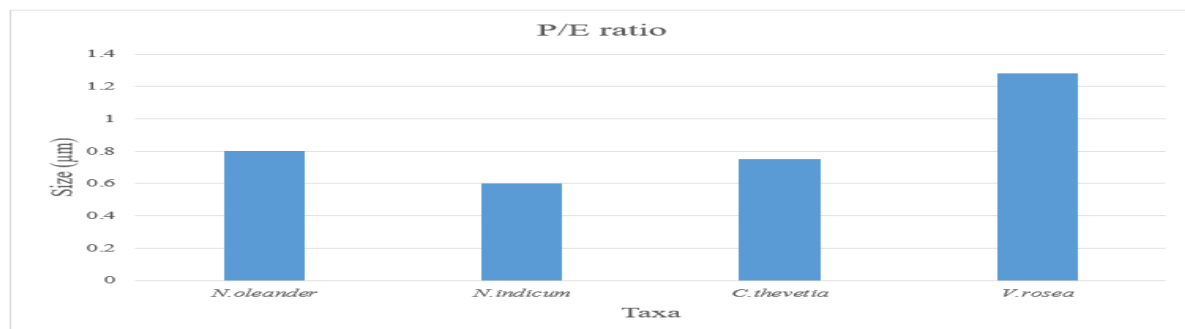
The pollen morphology of Apocynaceae taxa e.g *Nerium oleander*, *Nerium indicum*, *Cascabela thevetia* and *Vinca rosea* was examined. The highest ED was observed in *Nerium indicum* (25), *Vinca rosea* (17.5), *Nerium oleander* (12.5), and *Cascabela thevetia* (10).



Graph 02: Equatorial diameter (μm) of different pollen of family Apocynaceae

## P/E Ratio

The pollen morphology of Apocynaceae taxa e.g *Nerium oleander*, *Nerium indicum*, *Cascabela thevetia* and *Vinca rosea* was examined. The highest P/E Ratio was observed in *Vinca rosea* (1.28), *Nerium oleander* (0.8), *Cascabela thevetia* (0.75), and *Nerium indicum* (0.6).



Graph 03: P/E ratio (μm) of different pollen of family Apocynaceae

## Qualitative Characteristics of Apocynaceous Taxa

### Shape

The pollen shape of *Nerium oleander* were oblate-spheroidal. *Nerium indicum* were spheroidal while *Cascabela thevetia* and *Vinca rosea* were prolate-spheroidal.

### Size

The pollen size of *Nerium oleander* were medium-large, *Nerium indicum* were medium and *Cascabela thevetia* were large-very large while *Vinca rosea* were large.

### Exine sculpturing

The exine sculpture of *Nerium oleander* were regulate-psilate, *Nerium indicum* were psilate, *Cascabela thevetia* were reticulate while *Vinca rosea* were perforate.

**Table 4: Pollen Characteristics (qualitative) of Apocynaceous Taxa**

| S.no | Taxa                      | Pollen shape       | Pollen size      | Exine sculpturing |
|------|---------------------------|--------------------|------------------|-------------------|
| 1    | <i>Nerium oleander</i>    | Oblate-spheroidal  | Medium-large     | Regulate-psilate  |
| 2    | <i>Nerium indicum</i>     | Spheroidal         | Medium           | Psilate           |
| 3    | <i>Cascabela thevetia</i> | Prolate-spheroidal | Large-very large | Reticulate        |
| 4    | <i>Vinca rosea</i>        | Prolate-spheroidal | Large            | Perforate         |

**Table 5: Pollen Characteristics (quantitative) of Apocynaceous Taxa**

| S.no | Taxa                      | Polar diameter | Equatorial diameter | P/E ratio |
|------|---------------------------|----------------|---------------------|-----------|
| 1    | <i>Nerium oleander</i>    | 10             | 12.5                | 0.8       |
| 2    | <i>Nerium indicum</i>     | 15             | 25                  | 0.6       |
| 3    | <i>Cascabela thevetia</i> | 7.5            | 10                  | 0.75      |
| 4    | <i>Vinca rosea</i>        | 22.5           | 17.5                | 1.28      |

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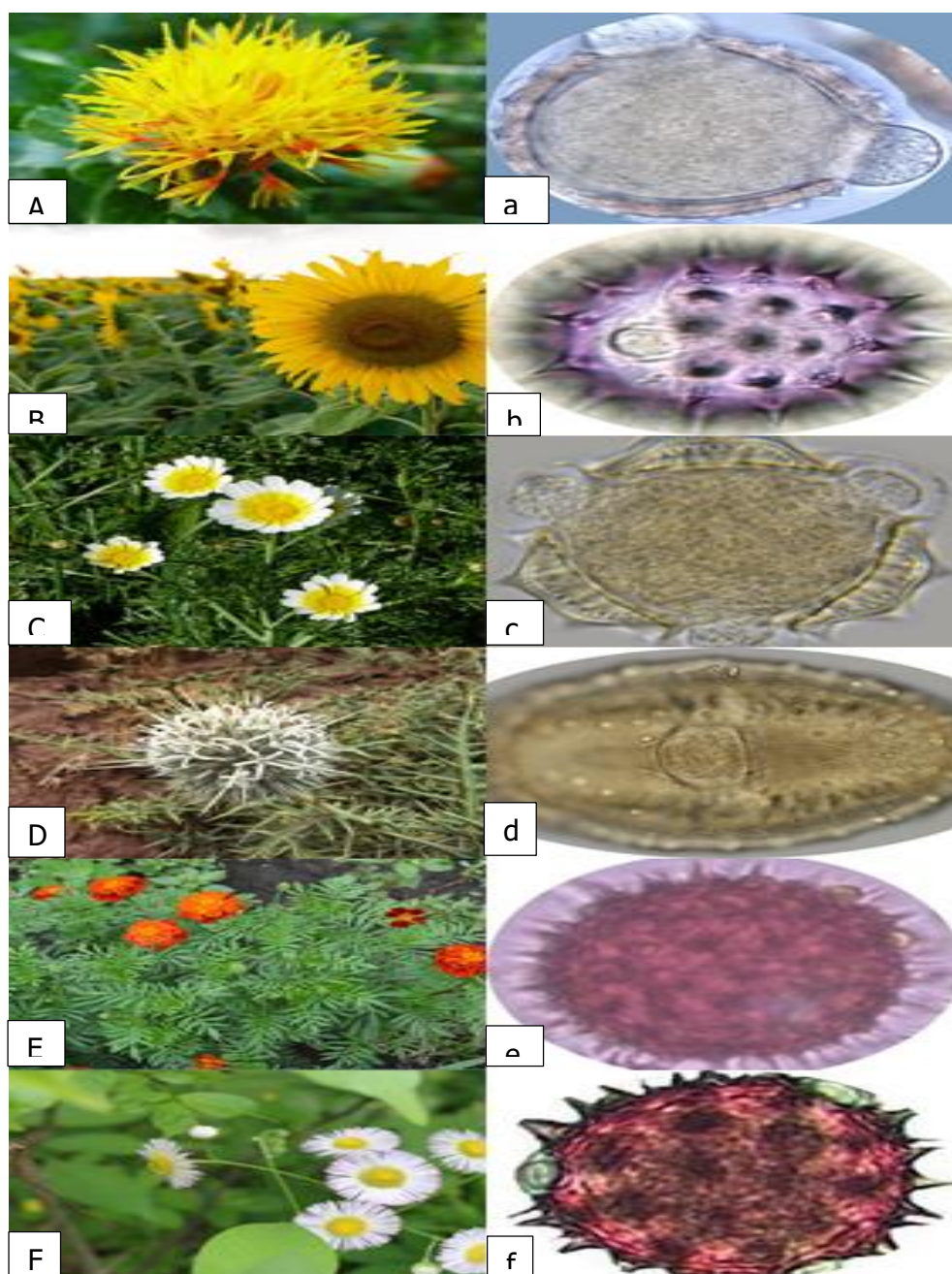


Plate 0.1: Plant and pollen picture of Asteraceae, [(A,a) *Carthamus tinctorius*], [(B, b) *Helianthus annuus*], [(C, c) *Glebionis coronaria*], [(D, d) *Echinops echinatus*], [(E,e) *Tagetes erecta*], [(F, f) *Conyza canadensis*]

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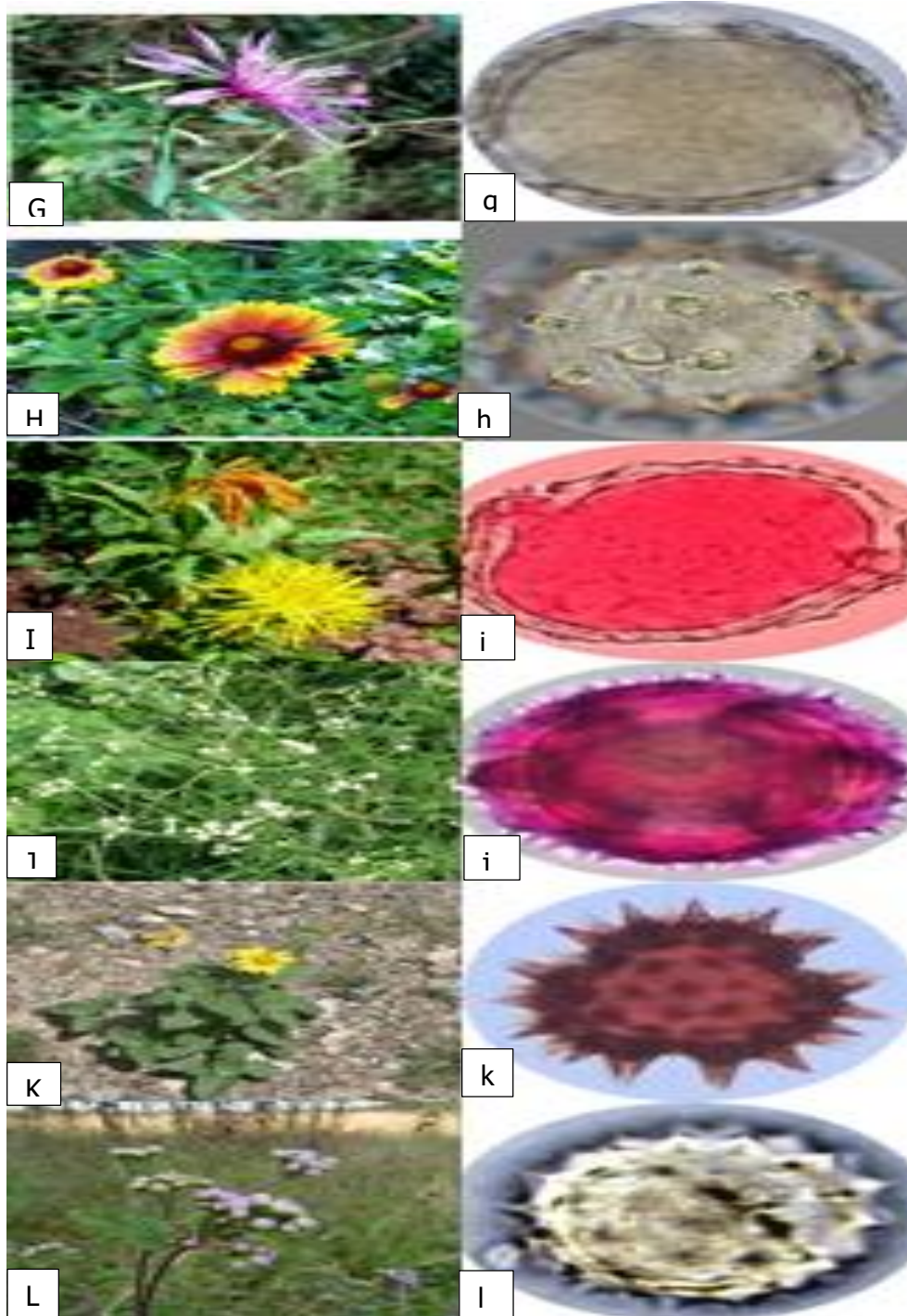


Plate 0.2: Plant and pollen picture of Asteraceae, , [(G, g) *Centaurea iberica*], [(H, h) *Gaillardia pulchella*], , [(I, i) *Carthamus oxyacantha*], , [(J, j) *Parthenium hysterophore*], [(K, k) *Verbena encelioides*], , [(L, l) *Agertum conyzoides*]

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Plate 0.3: Plant and pollen pictures of Apocynaceae. [(A, a) *Nerium oleander* ], [(B, b) *Nerium indicum* ], [(C, c) *Cascabella thevitia* ], [(D, d) *Vinca rosea* ]

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## List of Selected Species of Asteraceae and Apocynaceae Pectorial View of their Pollen Grain

Plate 0.1: Plant and pollen picture of Asteraceae, [(A,a) *Carthamus tinctorius*] , [(B, b) *Helianthus annuus*] , [(C, c) *Glebionis coronaria*], [(D, d) *Echinops echinatus*] , [(E,e) ) *Tagets erecta*] , [(F, f) *Conyza canadensis*]

Plate 0.2: Plant and pollen picture of Asteraceae, , [(G, g) *Centaurea iberica*], [(H, h) *Gaillardia pulchella*], , [(I,i) *Carthamus oxyacantha*], , [(J ,j) *Parthenium hysperophoure*], [( K , k) *Verbesina encelioides*], , [(L , l) *Agertum conyzoides*]

Plate 0.3: Plant and pollen pictures of Apocynaceae. [(A, a) *Nerium oleander* ], [(B, b) *Nerium indicum* ], [(C, c) *Cascabella thevitia* ], [(D, d) *Vinca rosea*

## Discussion

Palynology plays an important role for the delimitation of different families, tribes, genera and species among plants. In this regard, palynological study of Asteraceaeous and Apocynaceaeous taxa was undertaken in order to differentiate among various pollen features of selected species. Different parameters were taken into account for the delimitation of selected species. Details are discussed below.

Pollen morphology of the species of family Asteraceaeous e.g. *Carthamus tinctorius*, *Helianthus annuus*, *Glebionis coronaria*, *Echinops echinatus*, *Tagetes erecta*, *Conyza canadensis*, *Centaurea iberica*, *Gaillardia pulchella*, *Carthamus oxyacantha*, *Parthenium hysperophorus*, *Verbesina encelioides*, *Ageratum conyzoides* was also examined under the light microscope. The observation of the current study revealed the highest PD in *Carthamus tinctorius* (25.5), *Tagatus erecta* (20), *Gaillardia pulchella* (20), *Carthamus oxyacantha* (17.5), *Verbesina encelioides* (15), *Echinops echinatus* (12.5), *Parthenium hysperophorus* (12.5), *Helianthus annuus* (10), *Gelbiois coronaria* (10), *Centaurea iberica* (10), *Ageratum conyzoides* (7.5), and *Conyza canadensis* (5).

Pollen morphology of the species of family Asteraceae e.g. *Carthamus tinctorius*,

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3007-3189

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*Helianthus annuus*, *Glebionis coronaria*, *Echinops echinatus*, *Tagetes erecta*, *Conyza canadensis*, *Centaurea iberica*, *Gaillardia pulchella*, *Carthamus oxyacantha*, *Parthenium hysterophorus*, *Verbesina encelioides*, *Ageratum conyzoides* was also examined under the light microscope. The observation of the current study revealed the highest ED in *Verbesina encelioides* (20), *Tagetes erecta* (17.5), *Helianthus annuus* (15), *Glebionis coronaria* (12.5), *Conyza canadensis* (12.5), *Carthamus oxyacantha* (12.5), *Carthamus tinctorius* (10), *Ageratum conyzoides* (10), *Echinops echinatus* (7.5), *Centaurea iberica* (7.5), *Parthenium hysterophorus* (7.5) and *Gaillardia pulchella* (5).

Pollen morphology of the species of family Asteraceae e.g. *Carthamus tinctorius*, *Helianthus annuus*, *Glebionis coronaria*, *Echinops echinatus*, *Tagetes erecta*, *Conyza canadensis*, *Centaurea iberica*, *Gaillardia pulchella*, *Carthamus oxyacantha*, *Parthenium hysterophorus*, *Verbesina encelioides*, *Ageratum conyzoides* was also examined under the light microscope. The observation of the current study revealed the highest P/E ratio in *Gaillardia pulchella* (4), *Carthamus tinctorius* (2.25), *Echinops echinatus* (1.6), *Parthenium hysterophorus* (1.66), *Carthamus oxyacantha* (1.4), *Centaurea iberica* (1.33), *Tagetes erecta* (1.14), *Glebionis coronaria* (0.8), *Verbesina encelioides* (0.75), *Ageratum conyzoides* (0.75), *Helianthus annuus* (0.66), and *Glebionis coronaria* (0.8). The pollen shape of *Carthamus tinctorius*, *Glebionis coronaria*, *Centaurea iberica*, *Ageratum conyzoides* were spheroidal. *Helianthus annuus*, *Verbesina encelioides* were prolate-spheroidal. *Echinops echinatus* were triangular, *Carthamus oxyacantha* were circular while *Tagetes erecta*, *Conyza canadensis* and *Paethenium hysterophorus* were spherical.

The pollen size of *Carthamus tinctorius*, *Helianthus annuus*, *Gaillardia pulchella*, *Parthenium hysterophorus*, *Verbesina encelioides* were medium and *Glebionis coronaria*, *Centaurea iberica*, *Ageratum conyzoides* were small While *Echinops echinatus*, *Tagetes erecta*, *Conyza canadensis*, *Carthamus oxyacantha* were large. The exine sculpture of *Gaillardia pulchella*, *Carthamus tinctorius*, *Helianthus annuus*, *Glebionis coronaria*, *Echinops echinatus*, *Centaurea iberica*, *Gaillardia oxyacantha*, *Verbesina encelioides*, *Ageratum conyzoides* were Echinate and *Tagetes erecta*, *Conyza canadensis* were Psilate while

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*Parthenium hysterophorus* were Reticulate. The current results are similar with the study of zafar *et al.*, 2007; Moreira *et al.*, 2019) with small differences. Qureshi *et al.* (2009) reported the same results in Asteraceae family.

The pollen morphology of Apocynaceae taxa e.g *Nerium oleander*, *Nerium indicum*, *Cascabela thevetia* and *Vinca rosea* was examined. The highest PD was observed in *Vinca rosea* (22.5), *Nerium indicum* (15), *Nerium oleander* (10), and *Cascabela thevetia* (7.5). The pollen morphology of Apocynaceae taxa e.g *Nerium oleander*, *Nerium indicum*, *Cascabela thevetia* and *Vinca rosea* was examined. The highest ED was observed in *Nerium indicum* (25), *Vinca rosea* (17.5), *Nerium oleander* (12.5), and *Cascabela thevetia* (10). Description. The pollen morphology of Apocynaceae taxa e.g *Nerium oleander*, *Nerium indicum*, *Cascabela thevetia* and *Vinca rosea* was examined. The highest P/E ratio was observed in *Vinca rosea* (1.28), *Nerium oleander* (0.8), *Cascabela thevetia* (0.75), and *Nerium indicum* (0.6).

The pollen shape of *Nerium oleander* were oblate- spheroidal. *Nerium indicum* were spheroidal while *Cascabela thevetia* and *Vinca rosea* were prolate- spheroidal. The pollen size of *Nerium oleander* were medium- large, *Nerium indicum* were medium and *Cascabela thevetia* were large- very large while *Vinca rosea* were large. The exine sculpture of *Nerium oleander* were regulate- psilate, *Nerium indicum* were psilate, *Cascabela thevetia* were reticulate while *Vinca rosea* were perforate. Similar results were reported by (Naz *et al.*, 2019; Chatterjee *et al.*, 2014) in species of family Apocynaceae during their study.

## Conclusion

Pollen morphology of 12 species of Asteraceae family and 4 species of Apocynaceae family was analyzed using a light microscope. Both quantitative and qualitative parameters such as polar diameter, equatorial diameter, P/E ratio, size, shape, pollen unit were used for the delimitation of taxa. The study concludes that pollen morphology play an important for the delimitation of taxa. More work is needed in this field in order to complete pollen morphological information of different flora.

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Study need scanning electron microscopic and other cutting edge technologies for finer details of pollen study.

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