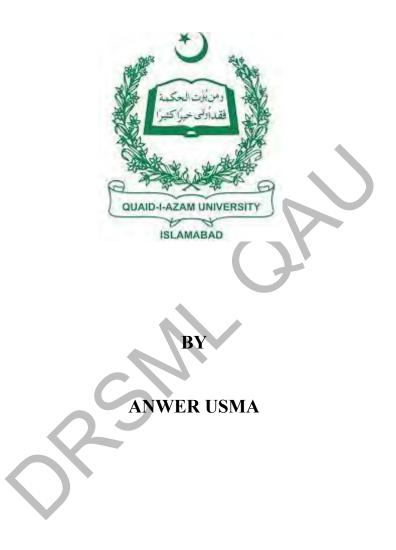
## Phytodiversity using Microscopic Characterization in Potohar Plateau-Pakistan

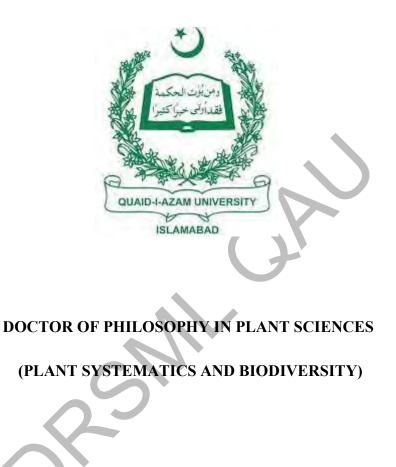


## **Department of Plant Sciences**

Quaid-i-Azam University Islamabad, Pakistan

2023

### Phytodiversity using Microscopic Characterization in Potohar Plateau-Pakistan



BY

ANWER USMA

Department of Plant Sciences, Faculty of Biological Sciences Quaid-i-Azam University Islamabad, Pakistan

2023



IN the name of ALLAH, the Most Beneficient, the Most merciful.

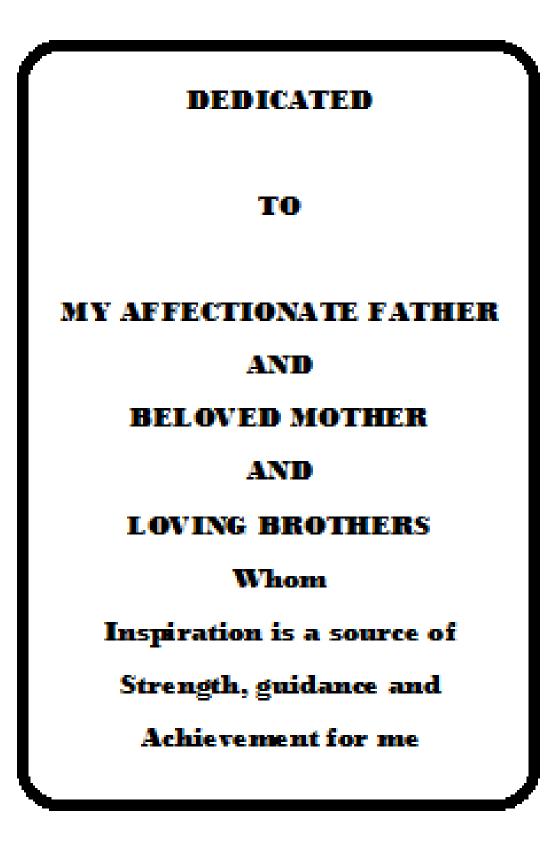
Verify, it is ALLAH who causes the seed grain and the stone fruit to split and sprout. He brings forth the living from the dead and the dead from the living. Such is ALLAH. Then how are you deluded away from the Truth 6:95

"And he to whom wisdom is granted receiveth indeed a benefit overflowing"

(Al-Baqara)

"He who treads the path in search of knowledge, ALLAH would make that path easy, leading to paradise for him"

Sahih Muslim Vol.4; Hadith No.6518



#### CERTIFICATE OF APPROVAL

This is to certify that research work presented in this thesis, entitled "Phytodiversity using Microscopic Characterization in Potohar Plateau-Pakistan" was conducted by Miss Anwer Usma under the supervision of Prof. Dr. Mushtaq Ahmad. No part of this thesis has been submitted anywhere else for any other degree. This thesis is submitted to the Department of Plant Sciences, Quaid-i-Azam University, Islamabad, Pakistan in partial fulfillment of the requirements for the degree of Doctor of Philosophy in the field of Plant Sciences (Plant Systematics and Biodiversity), Department of Plant Sciences, Quaid-i-Azam University, Islamabad, Pakistan.

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#### ANWER USMA

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# Subject: <u>Publication of W – Category Miss. Anwer Usma (PhD Scholar)</u>

This is in reference to circular regarding the publication requirement for  $PhD_{o}$  scholars in Department of Plant Sciences, Faculty of Biological Sciences. It is certified that **Miss.** Anwer Usma has published research papers in W – Categories as given below:

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1	Usma, Anwer, Mushtaq Ahmad, Muhammad Zafar, Shazia	2022	3.9
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	morphological diversity of pollen among Asteraceous taxa from		
	Potohar Plateau-Pakistan." Microscopy Research and		
	<i>Technique</i> 85, no. 7 (2022): 2467-2485.		

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# Contents

S. No.	Title	Page No.
	Chapter 1: Introduction	(1-21)
1.1	Floral Diversity in Potohar-Plateau Pakistan	1
1.2	Introduction to Potohar-Plateau Parts of Pakistan	3
1.3	Dominant Vegetation of Potohar Region	4
1.4	Pollen Morphology of Important Families of Potohar Region (Asteraceae, Amaranthaceae, Fabaceae, Poaceae, Weeds)	6
1.5	Seeds Morphology	14
1.6	Significance of Pollen and Seeds Using LM and SEM	16
1.7	Background Justification of The Study	17
1.8	Objectives of The Present Project	18
	Chapter 2: Material and Methods	(22-45)
2.1	Research Work Outline	22
2.2	Study Area	23
2.3	Climate of the Study Area	23
2.4	Schematic Sketch of Present Study	24
2.5	Field survey and Plants Collection	25
2.6	Herbarium Preparation and Plant Identification	30
2.7	Systematics Studies of Floral Diversity of Potohar-Plateau	30
2.8	Palynological Studies (LM & SEM)	30
2.9	Seed Morphological Studies	33
	Chapter 3: Results and Discussion	(46-282)
	Results	
1	Summary	46
3.1	Section I: Pollen Morphology	

	References	(265-310)
	Recommendation	263
	Conclusion	263
	Fabaceaous Species	
3.2.5	Taxonomic key of seed morphological characters of	257
3.2.4	Seeds Micromorphology of Fabaceous Species	241
3.2.3	Key to seed Morphological features of family Asteraceae	234
3.2.2	Micro-morphological characters of cypsela	213
5.2.1	Seed Morphology of Asteraceous and Fabaceous Plants from Potohar- Plateau Pakistan	212
3.2.1		212
3.2	Section II: Seeds Morphology	
3.1.6	Pollen Micro-Morphology of Weed Species	181
3.1.5	Pollen Micromorphology of Poaceacious Species	154
3.1.4	Pollen Micromorphology of Fabaceous Species	123
3.1.3	Pollen Micromorphology of Amarantheceous Species	98
3.1.2	Pollen Micromorphology of Asteraceaous Species	48
3.1.1	Pollen Micro-morphological Studies of Floral Diversity of Potohar Plateau	47

S. No.	Title	Page No.
1	Panoramic View of collection site at QAU, Islamabad	20
2	Panoramic View of collection site at Chakwal (Thoha Bahadur)	20
3	Collection area of grasses in Kallar kahar (Chakwal)	21
4	Collection area of Grasses in soon Sakaser	21
5	Panoramic view of Kallar Kahar	35
6	View of agricultural fields of Miani village	35
7	Diversity of plants near Uchali Lake	36
8	Pictorial view of Khabeki Lake	36
9	Wild flora near Numl Lake	37
10	Pictorial view of Vegetation on Kala Chitta Hills	37
11	Paranomic view of Soan Sakaser Valley	38
12	Diversity of plants of Khewra Salt Range	38
13	Plant collection during field surveys	39
14	Experimental work in laboratory to study pollen morphology	39
15	Quantitative data analysis of pollen using Microscope	40
16	Qualitative data analysis by studying morphological characters of plants	40
17	Light microscopic study of pollen grains	41
18	Morphological study of plants using stereo microscope	41
19	Preservation of plant specimen	42
20	Mounting of plants on herbarium sheets	42

#### LIST OF PLATES

21	Herbarium specimen photographs of (A) Heliotropium strigosum, (B) Impatiens bicolor, (C) Indigofera linifolia, (D) Symphyotrichum squamatum	43
22	<ul><li>Herbarium specimen photographs of (A) <i>Dactyloctenium scindicum</i>,(B) <i>Croton bonplandianum</i>, (C) <i>Chrozophora plicata</i>,</li><li>(D) <i>Cleome brachycarpa</i></li></ul>	44
23	Ageratum conyzoides (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Echinate Exine Sculpturing	56
24	Artemisia absinthimum (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Surface Sculpturing	57
25	Aster squamatus (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Orientation (D) Echinate Exine Sculpturing	58
26	<i>Bidens bipinnata</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Perforate Ornamentation (D) Echinate Exine Sculpturing	59
27	<i>Bidens pilosa</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Echinate Exine Sculpturing	60
28	Calendula arvensis (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken orientation (D) Exine Sculpturing	61
29	<i>Calendula officinalis</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Surface Sculpturing	62

30	<i>Carthamus oxyacantha</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Surface Sculpturing	63
31	<i>Carthamus lanatus</i> (A) Floral Parts Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Echinate Exine Sculpturing	64
32	Causinia prolifera (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Echinate Exine Sculpturing	65
33	Cirsium arvense (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Surface Sculpturing	66
34	<i>Eclipta prostrate</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Echino-Lophate Ornamentation (D) Surface Sculpturing	67
35	<ul><li><i>Erigeron bonariensis</i> (A) Floral Parts .Scanning Electron</li><li>Micrographs of Pollen (B) Polar View (C) Rounded orientation</li><li>(D) Echinate Exine Sculpturing</li></ul>	68
36	<i>Erigeron Canadensis</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Surface Sculpturing	69
37	Ixeris polycephala (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Echinate Pollen Ornamentation (D) Surface Sculpturing	70
38	<i>Lactuca sativa</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B)Polar View (C) Equatorial View (D) Echinate Sculpturing	71

39	Lactuca serriola (A) Floral Parts .Scanning Electron	72
	Micrographs of Pollen (B) Polar View (C) Elliptical sunken	
	orientation (D) Echinate Exine Sculpturing	
40		72
40	Launea naudicaulis (A) Floral Parts .Scanning Electron	73
	Micrographs of Pollen (B) Polar View (C) Rounded sunken	
	orientation (D) Surface Sculpturing	
41	Launea procumbens (A) Floral Parts .Scanning Electron	74
	Micrographs of Pollen (B) Polar View (C) Elliptical sunken	
	orientation (D) Micro-reticulate Echinate Exine Sculpturing	
42	Parthenium hysterophorus (A) Floral Parts .Scanning Electron	75
	Micrographs of Pollen (B) Polar View (C) Rounded sunken	
	orientation (D) Surface Sculpturing	
43	Silybum marianum (A) Floral Parts .Scanning Electron	76
	Micrographs of Pollen (B) Polar View (C) Echinate Pollen	
	orientation (D) Surface Sculpturing	
44	Sonchus arvensis (A) Floral Parts .Scanning Electron	77
	Micrographs of Pollen (B) Polar View (C) Elliptical sunken	
	orientation (D) Echinate perforate-tectate Exine Sculpturing	
45	Sonchus asper (A) Floral Parts .Scanning Electron Micrographs	78
	of Pollen (B) Polar View (C) Elliptical sunken orientation (D)	
	Echinate Exine Sculpturing	
46	Sonchus oleraceous (A) Floral Parts .Scanning Electron	79
	Micrographs of Pollen (B) Polar View (C) Elliptical sunken	
	orientation (D) Surface Sculpturing	

47	Sphagneticola trilobata (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken	80
	orientation (D) Echinate Exine Sculpturing	
48	<i>Taraxacum offinale</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken	81
	orientation (D) Echino-lophate perforate Exine Sculpturing	
49	Taraxacum platycarpum (A)Floral Parts .Scanning Electron	82
	Micrographs of Pollen (B)Polar View (C) Rounded sunken orientation (D) Echinate Surface Sculpturing	
50	Verbisinia eleusoides (A) Floral Parts .Scanning Electron	83
	Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Echino-lophate Exine Sculpturing	
51	Zinnia elegans (A) Floral Parts .Scanning Electron Micrographs	84
	of Pollen (B) Polar View (C) Echinate with pointed spines ornamentation (D) Surface Sculpturing	
52	Achyranthus aspera (A) Floral Parts .Scanning Electron	102
	Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Micro-Echinate Exine Sculpturing	
53	Aerva javanica (A) Floral Parts .Scanning Electron	103
	Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing	
54	Amaranthus gracizens (A) Floral Parts .Scanning Electron	104
	Micrographs of Pollen (B) Polar View (C) Sunken Pore	
	Ornamentation (D) Surface Sculpturing	
55	Amaranthus hybridus (A) Floral Parts .Scanning Electron	105
	Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Micro-Echinate Exine Sculpturing	
	orientation (D) Micro-Echinate Exine Sculpturing	

56	<i>Amaranthus spinosus</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Equatorial View (C) Polar View (D)	106
	Scabrate Exine Sculpturing	
57	Amaranthus viridis (A) Floral Parts .Scanning Electron	107
	Micrographs of Pollen (B) Polar View (C) Sunken Pore	
	orientation (D) Micro-Echinate Exine Sculpturing	
58	Atriplex stoeksii (A) Floral Parts .Scanning Electron	108
	Micrographs of Pollen (B) Polar View (C) Sunken Pore	
	orientation (D) Scabrate Exine Sculpturing	
59	Celosia Argentia (A) Floral Parts .Scanning Electron	109
	Micrographs of Pollen (B) Polar View (C) Sunken Pore	
	orientation (D) Surface Sculpturing	
60	Chenopodium album (A) Floral Parts .Scanning Electron	110
	Micrographs of Pollen (B)Polar View (C) Equatorial View (D)	
	Scabrate Exine Sculpturing	
61	Chenopodium ambrosioides (A) Floral Parts .Scanning Electron	111
	Micrographs of Pollen (B) Polar View (C) Sunken orientation	
	(D) Micro- Echinate Exine Sculpturing	
62	Chenopodium murale (A) Floral Parts .Scanning Electron	112
	Micrographs of Pollen (B) Polar View (C) Equatorial View (D)	
	Surface Sculpturing	
63	Digeria muricata (A) Floral Parts .Scanning Electron	113
	Micrographs of Pollen (B) Polar View (C) Sunken Pore	
	orientation (D) Surface Sculpturing	
64	Kochia indica (A) Floral Parts .Scanning Electron Micrographs	114
	of Pollen (B) Polar View (C) Sunken Pore orientation (D)	
	Scabrate Exine Sculpturing	

65	Pupalia lappaceae (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Micro-Echinate Exine Sculpturing	115
66	Sauda fruitcosa (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Surface Sculpturing	116
67	Acacia nilotica (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Reticulate Exine Sculpturing (D) Surface Sculpturing	126
68	Acacia modesta (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Reticulate orientation (D) Exine Sculpturing	127
69	<ul><li>Arachis hypagae (A) Floral Parts .Scanning Electron</li><li>Micrographs of Pollen (B) Polar View (C) Scabrate Surface</li><li>(D) Exine Sculpturing</li></ul>	128
70	Astragalus psilocentros (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Moderately Thick Exine Texture (D) Surface Sculpturing	129
71	Astragalus scoripiurus (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing	130
72	<i>Cassia fistula</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing	131
73	Cassia italica (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Moderately Thick Exine (D) Surface Sculpturing	132

74	Cassia occidentalis (A) Floral Parts .Scanning Electron of	133
	Pollen Micrographs (B) Polar View (C) Perforate Sculpturing	
	(D) Surface Sculpturing	
75	Crotolaria burhia (A) Floral Parts .Scanning Electron	134
	Micrographs of Pollen (B) Polar View (C) Equatorial View (D)	
	Surface Sculpturing	
76	Dilbergia sisso (A) Floral Parts .Scanning Electron	135
	Micrographs of Pollen (B) Polar View (C) Thin Exine Texture	
	(D) Surface Sculpturing	
77	Lathyrus aphaca (A) Floral Parts .Scanning Electron	136
	Micrographs of Pollen (B) Equatorial View (C) Thin Exine	
	Texture (D) Perforate Sculpturing	
78	Lespedeza juncea (A) Floral Parts .Scanning Electron	137
	Micrographs of Pollen (B) Polar View (C) Equatorial View (D)	
	Moderately Thick Exine Sculpturing	
70		120
79	Leucaena leucocephalla (A) Floral Parts .Scanning Electron	138
	Micrographs of Pollen (B) Polar View (C) Verrucate Exine	
	Sculpturing (D) Surface Sculpturing	
80	Melilotus indicus (A) Floral Parts .Scanning Electron	139
	Micrographs of Pollen (B) Polar View (C) Equatorial View (D)	
	Surface Sculpturing	
81	Pigomia pinnata (A) Floral Parts .Scanning Electron	140
	Micrographs of Pollen (B) Polar View (C) Equatorial View (D)	
	Surface Sculpturing	
82	Prosopis cineraria (A) Floral Parts .Scanning Electron	141
	Micrographs of Pollen (B) Polar View (C) Equatorial View (D)	
	Surface Sculpturing	

Prosopis juliflora (A) Floral Parts .Scanning Electron	142
Surface Sculpturing	
Pueraria tuberosa (A) Floral Parts .Scanning Electron	143
Micrographs of Pollen (B) Polar View (C) Reticulate Exine	
Sculpturing (D) Surface Sculpturing	
Rhynchosia capitata (A) Floral Parts .Scanning Electron	144
Micrographs of Pollen (B) Polar View (C) Reticulate Perforate	
Exine Texture (D) Surface Sculpturing	
Sophora mollis (A)Floral Parts .Scanning Electron Micrographs	145
of Pollen (B)Polar View (C) Perforate Exine Sculpturing (D)	
Surface Sculpturing	
Tephrosia purpurea (A) Floral Parts .Scanning Electron	146
Micrographs of Pollen (B) Polar View (C) Reticulate	
sculpturing(D) Thin Exine Texture	
Visin manufic lis (A) Elevel Derte Securing Electron	147
	,
Reneulate-Periorate Sculpturing	
Brachiaria brizantha (A) Floral Parts .Scanning Electron	157
Micrographs of Pollen (B) Prominent Pore Orientation (C)	
Polar View (D) Surface Sculpturing	
Cenchrus biflorus (A) Floral Parts .Scanning Electron	158
Micrographs of Pollen (B) Polar View (C) Sunken Pore	
	1
-	<ul> <li>Micrographs of Pollen (B) Polar View (C) Reticulate Exine Sculpturing (D) Surface Sculpturing</li> <li><i>Rhynchosia capitata</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Reticulate Perforate Exine Texture (D) Surface Sculpturing</li> <li><i>Sophora mollis</i> (A)Floral Parts .Scanning Electron Micrographs of Pollen (B)Polar View (C) Perforate Exine Sculpturing (D) Surface Sculpturing</li> <li><i>Tephrosia purpurea</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Reticulate sculpturing(D) Thin Exine Texture</li> <li><i>Vicia angustifolia</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Reticulate-Perforate Sculpturing</li> <li><i>Brachiaria brizantha</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Reticulate-Perforate Sculpturing</li> <li><i>Brachiaria brizantha</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Reticulate-Perforate Sculpturing</li> <li><i>Brachiaria brizantha</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Prominent Pore Orientation (C) Polar View (D) Surface Sculpturing</li> <li><i>Cenchrus biflorus</i> (A) Floral Parts .Scanning Electron</li> </ul>

91	Cenchrus ciliaris (A) Floral Parts .Scanning Electron	159
	Micrographs of Pollen (B) Polar View (C) Slightly sunken	
	Pore orientation (D) Scabrate Exine Sculpturing	
92	Cenchrus pennisetiformis (A) Floral Parts .Scanning Electron	160
	Micrographs of Pollen (B) Polar View (C) Oriented Pore	
	orientation (D) Surface Sculpturing	
93	Cymbopogon jwarancusa (A) Floral Parts .Scanning Electron	161
	Micrographs of Pollen (B) Polar View (C) Sunken Pore	
	Orientation (D) Fine gemmate Exine Sculpturing	
94	Cynadon dactylon (A) Floral Parts .Scanning Electron	162
	Micrographs of Pollen (B) Polar View (C) Scabrate-verrucate	
	Exine Sculpturing (D) Surface Sculpturing	
95	Dactyloctenium aegyptium(A) Floral Parts .Scanning Electron	163
	Micrographs of Pollen (B) Polar View (C) Sunken Pore	
	orientation (D) Surface Sculpturing	
96	Dichanthium annulatum (A) Floral Parts .Scanning Electron	164
	Micrographs of Pollen (B) Polar View (C) Prominent Pore	
	orientation (D) Gemmate-verrucate Exine Sculpturing	
97	Eleusine indica (A) Floral Parts .Scanning Electron	165
	Micrographs of Pollen (B) Polar View (C) Psilate-Scabrate	
	Exine Sculpturing (D) Surface Sculpturing	
98	Pennisetum glaucum (A) Floral Parts .Scanning Electron	166
	Micrographs of Pollen (B) Polar View (C) Equatorial View (D)	
	Surface Sculpturing	
99	Pennisetum typhoids (A) Floral Parts .Scanning Electron	167
	Micrographs of Pollen (B) Polar View (C) Equatorial View (D)	
	Granulate to perforate tectate Exine Sculpturing	

100	<i>Poa annua</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing	168
101	Saccharum spontaneum (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Surface Sculpturing	169
102	Steria viridis (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Gemmate–scabrate Exine Sculpturing (D) Equatorial View	170
103	Zea mays (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Prominent Pore Orientation (C) Polar View (D) Scabrate, psilate Surface Sculpturing	171
104	<ul><li>Anagallis arvensis (A) Floral Parts .Scanning Electron</li><li>Micrographs of Pollen (B) Polar View (C) Sunken orientation</li><li>(D) Surface Sculpturing</li></ul>	186
105	Asphodelus tenuifolius (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Angular orientation (D) Reticulate Exine Sculpturing	187
106	<ul> <li>Boerhivia procumbense (A) Floral Parts .Scanning Electron</li> <li>Micrographs of Pollen (B) Polar View (C) Rounded orientation</li> <li>(D) Scabrate Exine Sculpturing</li> </ul>	188
107	<ul><li>Brachychiton acerifolius (A) Floral Parts .Scanning Electron</li><li>Micrographs of Pollen (B) Polar View (C) Perforate orientation</li><li>(D) Surface Sculpturing</li></ul>	189
108	<i>Broussonetia papyrifera</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing	190

109	Bryophyllum pinnatum (A) Floral Parts .Scanning Electron	191
	Micrographs of Pollen (B) Polar View (C) Sunken orientation	
	(D) Surface Sculpturing	
110	Capparis decidua (A) Floral Parts .Scanning Electron	192
	Micrographs of Pollen (B) Polar View (C) Sunken orientation	
	(D) Surface Sculpturing	
111	Chorchorus depressus (A) Floral Parts .Scanning Electron	193
	Micrographs of Pollen (B) Polar View (C) Wavy orientation	
	(D) Surface Sculpturing	
112	Cissampelos pareira (A) Floral Parts .Scanning Electron	194
	Micrographs of Pollen (B) Polar View (C) Sunken orientation	
	(D) Exine Sculpturing	
113	Citharexylum spinosum (A) Floral Parts .Scanning Electron	195
	Micrographs of Pollen (B) Polar View (C) Equatorial View (D)	
	Exine Sculpturing	
114	Cleome brachycarpa (A) Floral Parts .Scanning Electron	196
	Micrographs of Pollen (B) Polar View (C) Sunken orientation	
	(D) Surface Sculpturing	
115	Cleome viscosa (A) Floral Parts .Scanning Electron	197
	Micrographs of Pollen (B) Polar View (C) Reticulate and	
	Scabrate Surface (D) Exine Sculpturing	
116	Commelina benghalensis (A) Floral Parts .Scanning Electron	198
	Micrographs of Pollen (B) Polar View (C) Sunken & Angular	
	orientation (D) Exine Sculpturing	
117	Convolvulus arvensis (A) Floral Parts .Scanning Electron	199
	Micrographs of Pollen (B) Polar View (C) Scabrate	
	Ornamentation (D) Exine Sculpturing	

118	Cucumis melo sub agerestis (A) Floral Parts .Scanning Electron Micrographs of Pollen (B)Polar View (C) Equatorial View (D) Exine Sculpturing	200
119	<i>Datura innoxia</i> (A)Floral Parts .Scanning Electron Micrographs of Pollen (B)Polar View (C) Scabrate & Reticulate Ornamentation (D) Echinate Exine Sculpturing	201
120	<ul><li><i>Euphorbia granulate</i> (A) Floral Parts .Scanning Electron</li><li>Micrographs of Pollen (B) Polar View (C) Sunken orientation</li><li>(D) Exine Sculpturing</li></ul>	202
121	<ul> <li>Oxalis corniculata (A) Floral Parts .Scanning Electron</li> <li>Micrographs of Pollen (B) Polar View (C) Sunken orientation</li> <li>(D) Exine Sculpturing</li> </ul>	203
122	Solanum surattense (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Reticulate Ornamentation (D) Surface Sculpturing	204
123	<i>Trianthema portulastrum</i> (A) Floral Parts .Scanning Electron Micrographs of Pollen (B) Polar View (C) Perforate Exine Ornamentatin (D) Surface Sculpturing	205
124	Cypsela micrographs of <i>Ageratum conyzoides</i> (A) General View (B) Surface Sculpturing , <i>Artemesia absanthimum</i> (C)General View (D) Surface Sculpturing	206
125	Cypsela micrographs of <i>Aster squamatus</i> (A) General View (B) Surface Sculpturing <i>,Biden pilosa</i> (C) General View (D) Surface Sculpturing	234
126	Cypsela micrographs of <i>Bidens bipinnata</i> , (A) General View (B) Surface Sculpturing , <i>Calendula arvensis</i> (C) General View (D) Surface Sculpturing	235

127	Cypsela micrographs Calendula officinalis (A) General View	236
	<ul><li>(B) Surface Sculpturing , <i>Carthamus oxyacantha</i> (C) General View (D) Surface</li></ul>	
128	Cypsela micrographs of <i>Carthamus lanatus</i> (A) General View	237
	<ul><li>(B) Surface Sculpturing , <i>Causinia prolifera</i> (C) General View</li><li>(D) Surface Sculpturing</li></ul>	
129	Cypsela micrographs of <i>Cirsium arvense</i> (A) General View (B)	238
	Surface Sculpturing , <i>Eclipta prostrate</i> (C) General View (D) Surface Sculpturing	
130	Cypsela micrographs of Erigeron bonariensis (A) General	239
	View (B) Surface Sculpturing ,. <i>Erigeron Canadensis</i> (C) General View (D) Surface Sculpturing	
131	Cypsela micrographs of <i>Ixeris polycephala</i> (A) General View	240
	(B) Surface Sculpturing , <i>Lactuca sativa</i> (C) General View (D) Surface Sculpturing	
132	Cypsela micrographs of <i>Lactuca serriola</i> (A) General View (B)	241
	Surface Sculpturing ,. <i>Launea naudicaulis</i> (C) General View (D) Surface Sculpturing	
133	Cypsela micrographs <i>Launea procumbens</i> (A) General View(B)	242
	Surface Sculpturing , <i>Parthenium hysterophorus</i> (C) General View (D) Surface Sculpturing	
134	Seed micrographs of <i>Acacia nilotica</i> (A) General View (B)	247
	Surface Sculpturing , <i>Accacia modesta (</i> C) General View (D) Surface Sculpturing	
135	Seed micrographs Arachis hypagae (A) General View (B)	248
	Surface Sculpturing, <i>Astragalus psilocentros</i> (C) General View (D) Surface Sculpturing	

136	Seed micrographs of Astragalus scoripiurus (A) General View	249
	(B) Surface Sculpturing, Cassia fistula (C) General View (D)	
	Surface Sculpturing	
137	Seed micrographs, Cassia italic (A) General View (B) Surface	250
	Sculpturing, Cassia occidentalis (C) General View (D) Surface	
	Sculpturing	
138	Seed micrographs of Crotolaria burhia (A) General View (B)	251
	Surface Sculpturing, Dilbergia sisso (C) General View (D)	
	Surface Sculpturing	
139	Seed micrographs of <i>Lathyrus aphaca</i> , (A) General View (B)	252
	Surface Sculpturing , <i>Lespedeza juncea</i> (C) General View (D)	
	Surface Sculpturing	
140	Seed micrographs of Leucaena leucocephalla (A) General View	253
	(B) Surface Sculpturing, Melilotus indicus (C) General View (D)	
	Surface Sculpturing	
141	Seed micrographs <i>Pigomia pinnata</i> (A) General View (B)	254
	Surface Sculpturing, Prosopis cineraria (C) General View (D)	
	Surface Sculpturing	
142	Seed micrographs of <i>Prosopis juliflora</i> (A) General View (B)	255
	Surface Sculpturing, Pueraria tuberose (C) General View (D)	
	Surface Sculpturing	
143	Seed micrographs of <i>Rhynchosia capitata</i> (A) General View	256
	(B) Surface Sculpturing , Sophora mollis (C) General View (D)	
	Surface Sculpturing	

#### LIST OF FIGURES

S.No.	Title	Page No.
1	Flow sheet showing research outline	22
2	Schematic Sketch of present project	24
3	Schematic Representation of Pollen Parameters Characterization	32
4	Parameters studied for seed morphological characters.	34
5	Map of study area showing collection sites.	45
6	Mean values of Pollen diameter among Asterecious taxa	85
7	Variation in P/E ratio index of Asterecious taxa	85
8	Mean data variations of colpi length and width in Asteracious taxa	86
9	Mean exine thickness in Asteraceous Species.	86
10	Data variations of pore length and width in Asteraceous species	87
11	Variations in mean values of Spines Length & Width among Asteraceous taxa	87
12	Mean values of Pollen diameter among Amaranthaceous taxa	117
13	Data variations of pore length and width in Amaranthaceous species	117
14	Variation in P/E ratio index of Amaranthaceous taxa	118
15	Mean exine thickness in Amaranthaceous Species.	118

	TT	
16	Mean values of Pollen diameter among Fabaceous taxa	148
17	Variation in P/E ratio index of Fabaceous taxa	148
18	Mean data variations of colpi length and width in Fabaceous taxa	149
19	Mean exine thickness in Fabaceous Species.	149
20	Mean values of Pollen diameter among Poaceous taxa	172
21	Mean exine thickness in Poaceous Species.	172
22	Data variations of pore length and width in Poaceous species	173
23	Mean values of Pollen diameter among Weeds	206
24	Variation in P/E ratio index of Weeds	206
25	Mean data variations of colpi length and width in Weeds	207
26	Mean exine thickness in Weeds.	207

### LIST OF TABLES

S.No.	Title	Page No.
1	Check List of Floral Diversity of Potohar-Plateau with Collection Site	42
2	Qualitative data of pollen of family Asteraceae members from Potohar region of Pakistan	48
3	Quantitative data of pollen of family Asteraceae members from Potohar region of Pakistan	52
4	Fertility and Sterility percentages of examined taxa of Asteraceae	55
5	Qualitative data of pollen of family Amaranthaceae members from Potohar region of Pakistan	98
6	Quantitative Data of Amaranthaceae members from Potohar Region of Pakistan	99
7	Fertility and Sterility percentages of examined taxa Of Amaranthaceae	101
8	Qualitative data of pollen of family Fabaceae members from Potohar region of Pakistan	124
9	Quantitative data of pollen of family Fabaceae members from Potohar region of Pakistan	125
10	Qualitative characters of some species of family Poaceae.	154
11	Quantitative pollen morphological characters of some species of family Poaceae	155
12	Fertility and Sterility percentage of selected species of Poaceae family	156
13	Qualitative data of pollen of weeds members from Potohar region of Pakistan	181

14	Quantitative data of pollen of weeds members from Potohar region of Pakistan	183
15	Pollen Fertility percentages of examined taxa of Weeds	185
16	Quantitative Seed micro-morphological attributes of Asteraceous species.	213
17	Qualitative Seed micro-morphological attributes of Asteraceous species.	214
18	SEM Ultrastructural Seed coat micro-morphological features of Cypsela	215
19	Micro- morphological attributes of seeds of family Fabaceae	241

#### Abstract

This Project is the systematics exploration of floral diversity of Potohar Plateau-Pakistan using quantitative and qualitative micro-morphological characterization. Floral diversity of the study area is first time documented systematically with comprehensive qualitative and quantitative micromorpholgical analysis. Field trips were conducted in different areas of Potohar Plateau including Attock, Chakwal, Mianwali, Soon sakesar, Miani Village, Uchali Lake, Kabbaki Lake, Kalla Chitta Hills, Rawalpindi and Islamabad. The plants were collected, identified, pressed, dried, preserved and deposited in the Herbarium of Pakistan (ISL), Quaid-i-Azam University Islamabad. The highest number of species belongs to family Asteraceae (29 species), Fabaceae (22 species), Amaranthaceae (17 species), Poaceae (15 species) and remaining species belongs to different families. The first section is confined to Palynomorphological characters studied by Light Microscope (LM) and Scanning Electron Microscope (SEM). The Pollen micro-morphological variations observed during this study include pollen shape and size, Polar and equatorial diameter, pore size, P/E ration, apertures, Length and width of spines and exine thickness. Similarly remarkable variations regarding pollen exine sculpturing was reported among various families from reticulate, microechinate, scabrate, verrucate, microechinate-aerolate types. The Second section is confined to Seeds morphological characters studied by Light Microscope (LM) and Scanning Electron Microscope (SEM). The Seeds micro-morphological variations observed during this study include Length and width of seed, L/W ratio, color, shape, anticlinal wall pattern, periclinal wall thickness, anticlinal wall protuberance, periclinal wall elevation, surface sculpturing.

Total 103 plant species were studied for palynological description via light microscopy (LM) and scanning electron microscopy (SEM). In family Asteraceae largest pollen size was found in *Bidens pilosa* (97.5um), while the lowest pollen size was observed in *Carthamus lanatus* (12.5um). In family Amaranthaceae largest pollen was found in *Amaranthus gracizens* (81um), while the lowest pollen size was observed *Digeria muricata* (11.50um). In Fabaceae largest pollen is observed in *Lespedeza juncea* (38um), while lowest size in *Prosopis cineraria* (14.3um) Based on systematics exploration of various macro and micro characters (LM and SEM), the diagnostic palynological characters includes exine sculpturing, shape of pollen, number of colpi and spines.

Seed micromorphology was studied in 40 species belong to two families Asteraceae and Fabaceae. Seed shape in asteraceous and fabaceous species observed was Linear, Linear fusiform, curved-ring shaped, oblanceolate, obovoid, cylindrical, Pointed and Linear lanceolate. The highest seed length observed in Ageratum conyzoides that is 7.56 While lowest in Erigeron Canadensis 1.6. The Highest seed width observed in Parthenium hysterophorus 3.9, While lowest in Erigeron bonariensis 0.5. Taxonomic keys based on palynological and seed morphological characters were developed for identification of different species belongs to different families.

The taxonomic keys developed on the basis of Palynology and seed morphology were useful for classification and delimitation of selected taxa within families. This study further recommends to work on these plants with respect to their conservation status and write monograph on these plants of Potohar-Plateau based on Palynomorphological and seeds morphological characters in addition to advanced systematics tools and features. The study further recommends correlating this data with phylogenetic information in order to use at global perspective for systematic of different families.



# 1.1 Floral Diversity in-Potohar-Plateau Pakistan

The flora of Pakistan exhibits its diversified climatic zones distribution ranging from temperate to tropical and arid to semi-arid. Heterogeneous climatic conditions with wide ranges of ecological zones. Atmospheric temperature, humidity and precipitation differ intensely in various altitudinal regions. In the south-western locales of this land, the elevation is quite low resulting into high temperature accompanied by low precipitation, such varied climatic condition results with the arid and semi-arid ecology. In ecological terms, there exist five kind of rangelands including the alpine pastures, the semi-alpine pastures, the temperate rangelands, and the arid rangelands (Hua et al., 2015).

The Potohar Plateau part of largest province Punjab selected for present project includes (9) cities Attock, Chakwal, Mianwali, Soon sakasar, Miani Village, Uchali Lake, Kabbaki Lake, Kalla Chitta Hills, Rawalpindi and Islamabad. North Punjab, usually known and termed as the Potohar -Plateau, stretches towards the south of northern hills, bordered in the west by Indus River and by Jhelum River in the east (Majeed et al., 2010). In the north Punjab region, Scrub vegetation ranges are located between Indus and Jhelum rivers in Attock, Rawalpindi, Jhelum and Chakwal Districts counting Islamabad. Acacia modesta is the leading species; in addition to that Amaranthus gracizens and dodonea viscosa are also found profoundly (Hussain, 2009). There is a variety of Plants and trees blooming in the area is reliant on rainfallas well as ground water, and the region is undergoing an undulated orography (Hashmi et al., 2012). The prevalent species of Plant families in this region belongs to Asteraceae, Amaranthaceae, Fabaceae, Poaceae, Brassicaceae, Asphodelaceae, Nyctaginaceae, Capparaceae, Chenopodiaceae, Tiliaceae, Menispermaceae, Verbenaceae, Cucurbitaceae, Euphorbiaceae at the same time, contingent upon their medicinal value (Bibi et al., 2014). Alongside the roads, field and canals, Dalbergia sisso is widely planted (Bajwa et al., 2003). Grass species which are native to Potohar region and the most abundantly found are Aristida mutabilis, Cenchrus ciliaris, Cynodon dactylon, Dichanthium annulatum, Cenchrus biflorus, Dactylolactenium scindicum, Eleusine indica (Hameed et al., 2008). Dominant species found in the area are Zizyphus nummularia, Acacia modesta, Melia azedarach, Albizia lebbeck, Dodonea viscosa, Cassia fistula and Dalbergia sisso (Qureshi et al., 2014; Qureshi et al 2018). Lanta camara, Xanthium strumarium and Prosopis juliflora were recorded as the chief nonnative species in this region (Malik and Husain, 2006). Other species of plants include Ajuga bracteosa, Rumex dentatus, Solanum nigrum, Chenopodium murale, Cleome brachycarpa, Capparis decidua, Atriplex stocksii, Corchorus depressus, Celosia argentea.

Salt range of Pakistan is blessed with rich floral diversity (Ahmad and Waseem, 2004). There are some natural resources existing in Salt Range. The climate of this Range is dry subtropical while the vegetation of is partially every every even while the vegetation of this locality is classified into three main types which are (i) Thorny-ever green tropical, (ii) semi ever green sub-tropical and (iii) degraded scrubs. Dodonea viscosa, Olea cuspidate and Acacia modesta that are typically present at hill swatches at an leviation of 750 m or higher are made up of the semi ever green sub-tropical (Ahmad et al., 2012).Plant species of drytropical thorny ever green forests includes Capparis decidua, Maytenus royleanus, Salvadora oleoides and Acacia nilotica while the plants that are under intense compression with inhibited growth constitute degraded scrublands (Ahmad et al., 2010). Salt accepting species are also found in some areas together with saline soil like Fagonia indica and Withania coagulans. The plants which are found in saline soil are Salsola foetida, Amaranthus gracigenzs and the grasses in the same saline soil include Cynodon dactylon (Ahmad et al., 2009). Nerium oleander is also reported to exist in this region (Ahmad et al., 2012). There is a great deal of diversity of grasses in the salt range with sixty two species (62) species of eleven (11) tribes involving the substantial volume of greenswards in northern Punjab, Pakistan (Ahmad et al., 2009). Pond borders and banks of streams have Saccharum spontaneum which is an exceptional soil binder. Acacia nilotica, Eucalyptus *camaldulensis* and *Zizyphus mauritiana* are other common trees of this locale while bushes that are also reported in this area include Cannabis sativa, Calotropis procera, Lantana camara, Prosopis cineraria. Furthermore, Aerva javanica, Anagalis arvensis, Amaranthus hybridus, Abutilon indicum, Cenchrus ciliaris, Cynodon dactylon, Eragrostis poides in consort with *Saccharum bengalense* have been reported to sprout here (Sarwar et al., 2017). Nearly sixty edible species of grass have already been documented within the study zone

and the most frequently arising species among them are *Cynodon dactylon. Aristida mutabilis, Heteropogan contortus* (Sarwar et al., 2017).

## 1.2 Introduction to Potohar-Plateau Parts of Pakistan

Potohar -Plateau is characterized by low annual precipitation (0-800mm) and severe lack of available water. The Potohar Plateau is divided into three different zones i.e Arid, semiarid and hyper-arid. In total one third land area of the world comprises of arid land. There are three major types of climate exists in Potohar-Plateau i.e Tropical, Mediterranean and continental. Distribution patterns of rainfall in both summer and winter season is different. Rainfall effects the Potohar-plateau in many ways and depending on edaphic characteristics the potential for soil erosion is quite different. Diurnal fluctuations of temperature also effect the growth of plant in Potohar-Plateau. Plant growth also affect the atmospheric humidity which is generally low. As a result of diurnal temperature changes, rocks disintegrate mechanically, root systems of the plant break up root particles subsequently soil in the Potohar-Plateau formed. In Potohar-plateau water holding capacity depends on the soil texture, structure and depth of the soil. In the area weathering of minerals and leaching of nutrients are slowed down due to decrease rainfall (Dregne, 2011). Vegetative growth in the Potohar regions are (i) Succulent perennials, (ii) non-succulent perennials, (iii) ephemeral annuals. Xerophytic plants of Potohar-plateau have a layer of cutin, extensive root system and different anatomical characteristics that help the plant to survive in this environment. Vegetation of Potohar-Plateau is quite different and classified as desert, semi desert, low rainfall wood land savanna, and in ever green scrub. In desert annual rain fall is less than 100mm, in semi desert 100 to 300mm in land savanna (300-600mm) and more than 500mm in ever green scrub (Jiapaer et al., 2011).

Potohar-Plateau of North Punjab spreads from 32 °10 °34 °9 °N latitude and 71° 10°73° 55° E longitude encompassing the district Attock (Jang, Hazro, Fateh jang, Hasan Abdal, Pindi Gheb), the district Rawalpindi ( Kotli Sattian, Gujar Khan, Kahuta, Taxila, Kallar Sayyedan), Jhelum district ( Dina, Sohawa, Pind Dadan Khan) and the district of Chakwal (Choa Saidan Shah, Kallar Kahar, Talagang, Lawa).

# 1.3. Dominant Vegetation of Potohar Region

The Potohar region is very rich in Grass flora, which is economically very important for livestock and wildlife feed, shelters for many birds and mammals, construction work, hay production, and civilian medicinal purposes (Ahmad et al., 2010). Chaudhry et al. (2001) Reported 41 species from the Chumbi Surla Wildlife Sanctuary, Ahmad et al. (2010) enlisted 60 grasses from Soone Valley and Nawaz et al. (2012) reported 33 species from the Salt Range.

The predominant grasses are *Heteropogon contortus* and *Cynodon dactylon* along the hills, and the salt patches are dominated by *Ochthochloa compressa* and *Aeluropus lagopoides*. *Chrysopogon serrulates*, *Cymbopogon jwarancusa* and *Pennisetum orientalis* are moderately high and predominant. *Aristida adscensionis*, Bromus spp. are high altitude predominant. The genus Lorium. (Nawaz et al., 2012; Hameed et al., 2012) also exist in the dominant vegetation of this region.

Other dicotyledonous plants that dominate the region are Phulai (*Acacia modesta*), Zaitoon (*Olea ferruginea*), and baikhar (*Justicia adhatoda*). Many alien invaders such as Chatak Chandni (*Parthenium hysterophorus*), Sanatha (*Dodonaea viscosa*), Panch phalli (*Lantana camara*) and Masqat (*Prosopis glandulos*) are endemic on the low hills (Hameed et al., 2012).

Studies of vegetation response to climate change focus on mountainous areas of recent times. Slight elevations can change the overall vegetation structure, species distribution pattern, and species composition along with physiological and other climatic factors (Parmesan, 2006).

Altitude changes are directly related to atmospheric pressure and temperature decline, but other factors such as soil nutrient and water availability, growth phase, radioactivity, and wind speed may also affect (Charrier et al., 2015). In general, grasses are more likely to adapt to extreme environments, so a wide range of species are expected to be resistant to different habitat types (Vitasse et al., 2009).

Species distributional pattern, species composition and community structure significantly altered with altitudinal gradient as reported by a number of researchers all over the world (Li et al., 2011; Abbasvand et al., 2014). Similar finding has been recorded

in the Potohar region, particularly at higher altitudes. Variability in climatic and physiographic conditions are exceptionally high in the region, such as high mountain peaks of Tret, Daleh and Sakesar, sandy desert in Khushab district, heavily salt affected foothills near Lillah and Pind Dadan Khan, vast valleys like Vanhar, Soone and Soan, protected areas like Chinji, ChunbiSurla, Kala Chitta hills, Lehri Jindi and Domeli Diljabba, drier hills of Kala bagh, cooler Murree foothills, etc. (Mahmood et al., 2012). These variation is strong enough to impose a drastic change in distributional pattern and ecology of inhabitant grass species. (Bijlsma & Loeschcke et al., 2005). Species colonizing lower altitudes were different from those of higher altitudes in their structure, distributional range and contribution towards community structure (Lenoir et al., 2008).

High altitude species like *Lolium persicum*, *L. perenne*, *Themeda anathera*, *Arthraxon lancifolius*, *Apluda mutica*, *Rostraria cristata*, *Koeleria cristata* and K. macrantha restrict to limited areas. Vegetation of the Potohar region is constantly under threat due to anthropogenic activities like firewood collection, mining activities, construction work, overexploitation of medicinal/ economic plant resources, deforestation for agricultural purposes, habitat fragmentation, grazing and browsing of livestock (Wilson et al., 2005).

As Consequence, quite a few species are endangered, and facing extinction at local and international level (Nawaz et al., 2012). Moreover, species like *H. contortus*, *A. adscens*, *D. scindicum* in addition to *C. serrulatus* and *C. jwarancusa* dominated the vegetation community, all these species are well adapted to xeric conditions (Nawaz et al., 2012).

Environmental conditions abruptly changed at the highest altitude regarding temperature, atmospheric pressure, amount of annual rainfall and nutrient availability (Adler & Levine, 2007). Lower altitudes of the Potohar region generally shared similar climatic conditions with few exceptions. This might be a strong reason for close association of two or three dominant species at almost all altitudinal ranges, where a sudden change in climates and vegetation observed. *Chrysopogon serrulatus*, *C. jwarancusa* and *C. dactylon* dominated the vegetation component all over the Potohar region, all showed significant

association among themselves (Patterson, 1980). *Saccharums pontanium,* a species of aquatic habitat, is Chang et al (2012) as reported by, it has been shown to be associated with *Imperata cylindrica* or *Cynodon dactylon,* which share a similar habitat.

# 1.4 Pollen Morphology of Important Families of Potohar Region (Asteraceae, Amaranthaceae, Fabaceae, Poaceae, Weeds)

Palynology play a significant role in the formation of natural groups and help in the evaluation of taxonomic relationship among various taxa. The study of Pollen morphology, an effective support to phylogeny and plant taxonomy has been recognized by numerous researchers (Bahadur et al., 2018; Bano et al., 2019; Nazish et al., 2019; Ullah et al., 2019). Likewise, Mbagwu and Edeoga (2006) use pollen characters to establish possible evidence of connection among particular groups of Angiosperms. The morphology of pollen grains is important for documentation and classification of plants, validation and confirmation (Arora and Modi, 2008).

### (A) Asteraceae

Family Asteraceae consist of 1530 genus and approximately 23000 species which are distributed within 3 subfamilies and 17 tribes (Hayat and Khan,2009). It is the largest family of angiosperms and cosmopolitan in distribution. In Pakistan this family comprises of 650 species distributed in 15 tribes (Ahmad et al 2013). Members of this family are mostly herbs rarely trees and shrubs. (Skvarla et al, 1977) worked on the unique architecture of exine of this family and also studied the advance phylogeny and taxonomy of family Asteraceae.

Taxonomists use many disciplines to improve identification, classification and systematics position of plants with in taxa. But the most significant tool for identification and classification of closely related plant taxa is the study of their pollen grains. In Pakistan many work has been done on basic and applied palynology, while there is plenty of information published on pollen and spores morphology worldwide. The family Asteraceae is eurypalynous family (Meo et al.,2004) and having zonocolporate pollens found in most

of its genera. The pollens are tricolporate; exine sculpturing is echinate with different variations in number and size of colpi (Qureshi et al 2009). The pollen grains of tribe Astereae, Helianthae and Inuleae has spinulose exine sculpturing while Vernonieae possess spinulose- lophate type of exine sculpturing. Some work has been done on fertility of pollens as it plays vital role in fruit and seed production in flowering plants (Rigamoto et al 2002). In family Asteraceae pollen fertility is high in both diploids and tetraploids (Blackmore et al 2009). Depending upon the climatic conditions fertility of pollens is based. In warm environment some species of genus Tradescantia pollen fertility may be higher than in cold, wet days (Ahmad et al.,2013). Due to the cosmopolitan distribution and predominance this family occupies highest rank among the angiosperm families.

Pollen morphology of family Asteraceae was done for the first time in Pakistan by (Perveen and Qaiser, 1999). (Coutinho et al., 2021) studied different aspects of pollen of different tribes and genus of family Asteraceae. However, at present there is no separate documentation on the pollen morphology of species belonging to family Asteraceae from Potohar region of Pakistan. The aim of this study is to provide detailed information of pollens and their exine structure based on both qualitative and quantitative characters by using Light and Scanning electron microscopy. These characters are helpful in identification of complicated species of the family. This study also aims to determine the pollen fertility of selected species of family Asteraceae which are economically important and this will help to identify the freely reproducing species for conservation purposes.

### (B) Amaranthaceae

Amaranthaceae is a flowering plants family as the word amaranthus is derived from the "anthos" which is a Greek word means everlasting flower (Rastogi & Shukla, 2013). Amaranthaceae is large, diverse and cosmopolitan family. In past, some researchers included chenopodiaceae in the wider circumscription of Amaranthaceae and some authors have represented monophyly in their molecular studies in these families (Cuénoud et al., 2002; Judd, Campbell, Kellogg, Stevens, & Donoghue, 2008). The previous morphological and phylogenetic research revealed that Amaranthaceae and Chenopodiaceae had long been considered a single evolutionary lineage and were closely

related families (Wang, Zhou, Bai, & Zhao, 2018). The AGP IV system in 2016 placed the family Amaranthaceae in order Caryophyllales and includes the plant species previously treated as family Chenopodiaceae. The family Amaranthaceae is estimated to contain a distribution of 180 genera and 2,500 species worldwide (Chase et al., 2016; Iamonico & Mosyakin, 2018; Kanwal & Abid, 2017; Safiallah, Hamdi, Grigore, & Jalili, 2017). The centers of diversity of Amaranthaceae are tropical and warm temperate regions. That is, Central and South America, South Africa, and Australia (Müller & Borsch, 2005). Majority of its species are halophytes and adapted to saline, xerophytic and xerohalophytic communities because they have the ability to grow and survive in high temperature, high irradiance, drought, and salinity (Dehghani & Akhani, 2009; Hedge et al., 1997; Zhigila, Yuguda, Akawu, & Oladele, 2014). In Pakistan, the plant species belong to this family are dominated by herbs and shrubs and dominantly distributed in salt ranges. The halophytes represent about 19% flora of Pakistan. The highest number of halophytic plant species (90) present in Pakistan is included in the family Chenopodiaceae (Khan & Qaiser, 2006). Palynological investigation is a beneficial tool in the taxonomic and phylogenetic identification of many plant families (Bahadur et al., 2018; Clark, Brown, & Mayes, 1980; Khan et al., 2018).

The application of light microscopy (LM) and scanning electron microscopy (SEM) has proven invaluable for observations of the surface morphology of plant materials. The use of SEM for the micro-morphological analysis of pollen grains has become a vital research method (Yang et al., 2017). Ullah et al. (2018) investigated the pollen morphology of subfamily Caryophylloideae and its taxonomic significance using LM and SEM. The micromorphological characters of *Marrubium trachyticum* Boiss. (Lamiaceae) using SEM and LM have been reported (Akçin & Camili, 2018). Butt et al. (2018) also utilized LM and SEM for morpho-palynological study of Cyperaceae. The use of LM and SEM is of special interest for the taxonomist to examine various taxonomic features (Khan, Ahmad, Zafar, & Ullah, 2017). Various authors have studied pollen morphology of family Amaranthaceae and Chenopodiaceae worldwide using LM and SEM (Dehghani & Akhani, 2009; Olvera, Fuentes-Soriano, & Hernández, 2006; Punsalpaamuu, Schluetz, Gegeensuvd, & Saindovdon, 2012; Talebi, Noori, & Nasiri, 2016). Moquin-Tandon was

the first researcher who carried out the palynological study of Chenopodiaceae (Moquin-Tandon, 1849). Borsch (1998) studied the pollen types in Amaranthaceae using scanning electron microscope and revealed new characters such as ektexinous bodies on the membrane of an aperture. Olvera et al. (2006) studied the pollen morphology and systematics of Atripliceae from herbarium specimens and showed that the quantitative morphology of pollens provides insight for conflicts based on macro-morphology. The pollen characters including pollen diameter, pore diameter, pore number, exine thickness, operculum diameter and operculum spinules of subfamily Suaedoideae in Iran have been reported by Dehghani and Akhani (2009). Talebi et al. (2016) also conducted a palynological study on some Amaranthus taxa from Iran using light and scanning electron microscope and their results showed circular and poly pentaporate pollens with scabrate surface sculpturing. The comprehensive study on the pollen characters of some plants of Chenopodiaceae has been carried out by Punsalpaamuu et al. (2012) in Mongolia. Their study reported poly pantaporate pollen grains having exine without perforations. A palynological investigation of 13 Alternanthera species distributed in Mexico has been studied by Sánchez-del Pino, Fuentes-Soriano, Solis-Fernández, Pool, and Alfaro (2016) who indicated the pollen morphological characters (Pollen ornamentation, number of nanospines on pore membrane and metareticula form) as a beneficial tool to discriminate three types of pollens (Spheroidal, de decahedrical, dimorphic) within Alternanthera. The pollen grains morphology and biometry of 17 taxa belonging to Amaranthaceae family have been studied by Angelini et al. (2014) who reported spheroidal and radially symmetrical pollen grains with echinae and perforations on the surface.

The family Amaranthaceae is one of the rich families of the Potohar Plateau of Pakistan as it includes a higher number of halophytes present in salt range areas of Potohar Plateau. The pollen morphology of its members possesses rich diversity (i.e., radially symmetrical, usually apolar, pantoporate, polypantoporate, shape spheroidal, tectum punctate scabrate, rarely spinulose; Perveen & Qasier, 2012; Hussain et al., 2018). Pollen grains of Amaranthaceae are pantopolyporate and some genera of the family such as *Chenopodium* L. have similar exine structure (Pinar & İnceoglu, 1999b). Perveen and Qasier (2012), studied the pollen morphology of 40 Amaranthaceae taxa belonging to 14

genera, which was based on exine and surface ornamentation. Shinwari, Shinwari, and Khan (2004) described two major types of pollens (periporate and fenestrate) in 7 genera and 18 species of Amaranthaceae using light microscopy.

The few researchers in Pakistan have conducted a study on the pollen morphology of family Amaranthaceae and Chenopodiaceae which were treated as separate families but now family Chenopodiaceae is merged into the family Amaranthaceae. The current study has been carried out keeping in view the use of LM and SEM in the significance of pollen studies of Amaranthaceae and its aid in the identification of halophytic flora. The aim of current research work is to evaluate the palyno-morpholoical characters, pollen types, shape and sculpturing and relationship within the different genera of the family using light and scanning electron microscopy

# (C) Fabaceae

Fabaceae Family comprises 730 genera and 19,325 species (Lewis et al. 2005). It is the largest family in terms of the number of endemic species in Brazil (Forzza et al. 2010), with 228 genera and 2896 species (Flora do Brasil 2020). Along with Quillajaceae D. Don, Polygalaceae Hoffmanns. & Link e Surianaceae Arn., the family forms the Fabales (APG IV 2016). Regarding the number of species, Fabaceae represents the third largest family of angiosperms, surpassed only by the families Asteraceae and Orchidaceae (Legume Phylogeny Working Group (LPWG) 2017). Due to the high number of species, the family is considered cosmopolitan, dispersed mainly in tropical regions (Lewis 1987; Judd et al. 2009).

Regardless of the systematic disparity within the family, Fabaceae appears to be a monophyletic group, despite the subfamily Caesalpinioideae being considered a paraphyletic group (LPWG 2013). To make the Caesalpinioideae more homogeneous, Lewis et al. (2005) divided and organized the genera into four tribes (Caesalpinieae, Cassieae, Cercideae, Detarieae). Currently, a new classification for Fabaceae is proposed by the LPWG (2017), which brings a new arrangement of the subfamilies based on morphological and molecular data. In this new classification, the Cercideae, Detarieae,

Duparquetia and Dialiineae tribes, and the large clade formed by Mimosoideae– Caesalpinieae–Cassieae that constituted the subfamily Caesalpinioideae, are now recognized as subfamilies, together with Papilionoideae. Therefore, Fabaceae now consists of six subfamilies: Cercidoideae, Detarioideae, Duparquetioideae, Detarioideae, Caesalpinioideae and Papilionoideae (LPWG 2017).

### (D) Poaceae

Grasses constitute a natural homogenous assemblage of kingdom Plantae belonging to the family Poaceae (Gramineae). Undeniably, Grasses are one of the most captivating families of flowering plants, with their variable life forms and indeed they plays their momentous part for all the living creature on planet Earth (Mitra and Mukherjee, 2005). With 610 genera and about 10,000 species, grasses are one to the largest and most treasured groups of plants (Cope, 1982). After Asteraceae, Leguminosae, Orchidaceae and Rubiaceae, the 5<sup>th</sup> largest family of Kingdom Plantae is the grass family, which is mainly known as Poaceae or Gramineae (Tzevlev and Michaelova, 1989). The family Gramineae can be distinguished by other families in terms of its characteristic morphological features which includes ligulate leaves, sheathing and flowers in spikelets, palea, glumes, lemma and caryopsis. The grasses are not closely related to other monocots. Size of grasses varies from minute unobtrusive herbs which are less than one inch to the giant sized bamboos that length up to 130 feet tall. It is very difficult to count the clear-cut number of grass species; however, according to Tzvelev, Poaceae, the number estimates up to of 11,000 species (Osborne, 2010).

The worth of Poaceae family have been acknowledged since time age-old to the modern day cereal crops which are the cultivars of their wild fore-fathers. The grass family mirrors extreme adaptation in response to their alternating atmospheres, capability to share their space with grazing animals and humans. Members of Poaceae have infinite diversity with distinctive forms of living. They propagate, replicate and complete their life in one short cycle. The most important reproductive phenomenon is seed production. The body of plant is just a limited thin leaf; single or double stems but the most significant feature is spike inflorescence which is the main reason of maximum seed production. Manifestly,

grasses are the living creatures which are successful as they survive in hard conditions and grow when conditions become right. As grasses reproduce quickly, due to their short life cycle, therefore they belongs to group of most successful telluric life forms on the earth (Gandhi et al., 2016).

Poaceae is very diverse and one of the most morphological intricate angiosperm families (Lawrence, 1964). This family represents 9,000 species belonging to 651 genera (Khine, 2011). According to Clayton and Renvoize (1986), the total number of grasses in the world is approximately 10,000 sp. 651 genera. Grasses survive and lives in all conceivable habitats and every climatic condition (Mitra and Mukherjee, 2005). Habitats of grasses can be assumed as the highpoint vegetation which can be the semi-arid grasslands of the America as well as the pampas of Asia and the prairies of Africa (Ahmad et al., 2009).

Agrostology is the study of grasses. The grass family is of interest to humans. The cultural significance of cereal grasses times to the period when man was emerging from wild beast stage (Gould, 1968). In Pakistan, in terms of species and genera, Poaceae is one of the leading families of angiosperms (Cope, 1982). As a major portion of diet, most people on earth depends on grasses. Members of the family constitutes approximately 20% of the earth's land, therefore they are considered as ecological dominants of planet Earth (Samreen et al., 2015).

The life form of grasses can be annual or biennial or perennial. Annual means they complete their life in one season, and biennial plants complete their life in two seasons and perennial are those grasses which lives long and they took more than two years to fulfill their life season. C3 and C4 grasses are alienated into two physiological groups on the basis of leaf anatomy. Kranz anatomy is exhibited by C4 type grasses. C4 grasses has radiate mesophyll, but in Poaceae family C4 photosynthesis exhibits numerous independent pedigrees (Gandhi et al., 2016).

# (E) Weeds

Weeds are considered plants that complete with the growth and development of crops. The growth of weeds in agricultural field can limit the production of staple crops and hence causes serious damage to their production (Chaudhri, 1992). According to weeds are those unwanted plants that can grow in irrelevant regions of the agricultural field. They grow in cultivated and wild habitats and cause damage to associated growing plants. Weeds belonging to the family Fabaceae can get essential nutrients from the soil and compete with crops to get this nutrition faster than the other plants(Rahmatullah et al., 2009). These plants obtained nitrogen from their surroundings and converted it into nitrogen-rich compounds to enhance the growth of essential microorganisms, increasing soil fertility(Mehsud et al., 2013). Many researchers work on the weed flora of the world specifically recorded 50 parasitic weeds from Pakistan.

The floral diversity of specific regions has great significance and their natural habitats had often been interpreted by weeds species due to the struggle for survival. Study of pollen grains plays an important role while studying plant taxonomy and biodiversity, as it helps in identifying plant species of particular area (Amarger, 2001).. To identify plant species, many taxonomists use different disciplines of plant systematics like morphology, anatomical studies, and palynological studies to find the exact position of plants within taxa. But the studies of pollen characters are considered the most important tool for classifying and identifying morphologically similar plants(Rashid & Rasul 2011). Previously much research has been done on pollen grains in Pakistan and worldwide.

Weeds play an important role in creating ecological balance in a crop system through supporting different life forms (Zafar et al., 2006. Throughout the world major vegetation of weeds belongs to families like Asteraceae, Poaceae, Amaranthaceae and Fabaceae. Growth of weeds is mostly dependent on the climatic condition of the area (Meo et al., 2005). Meo et al., (2004). Weeds are mostly found growing along the water passages, harvesting costs and fire hazards. In some previously reported studies the most commonly grown weeds of cotton crops are *Cynodon dactylon, Amaranthus viridis, Phyla nodiflora and Phragmites australis*. Common weeds of wheat crops are *Phragmites* sp., *Cynodon* sp., *Ranunculus* spp and *Polypogon* sp., Likewise, *Achyranthes aspera* is a common weed

of sugarcane crop and Plantago lanceolata is important weed of chickpea fields. The correct identification of species is determinant for developing any weed management program according to its different ecological aspects. The taxonomic studies are preliminary research towards the use of identification and classification of weeds of the study area, which could be in the same way useful for weed scientists as well as for botanists. Thus, the morpho-palynological characters of weeds observed through by using microscopic techniques.

### 1.5 Seeds Morphology

Seed micromorphology provides several characters to support the significance of seed morphology in inferring phylogenetic as well as evolutionary relationships and species identification (Attar, Keshvari, Ghahreman, Zarre, & Aghabeigi, 2007; Johnson, Huish, & Porter, 2004). The diversity in seed shape, size, and color can help distinguish taxa (Ninkaew, Pornpongrungrueng, Balslev, & Chantaranothai, 2017). In previous studies seed morphology and anatomical features were considered rather conservative, thus supporting their taxonomic importance (Werker, 1997). Seed attributes have been revealed to be a useful tool for the delimitation of species at family (Speta, 1998) and grouping at the generic level (Jessop, 1975). Seed ultrastructural features particularly the seed surface and cell shape are considered to be a valuable distinguishing character at intraspecific and intrageneric levels (Kubitzki, Rohwer, & Bittrich, 2013). Seed size was reported to be directly related to the number of nutritional reserves that can be allocated to initial seedling development (Veloso et al., 2017).

### (A)Asteraceous Seeds Morphology

Cypsela morphology is one of the most effective taxonomic feature that has an important role in solving different taxonomic problems related to inter- and intra-generic associations among different plant species of Asteraceae (Abid & Qaiser, 2015). Different morphological features (size, beak absence/presence, pappus branching and surface structure) are the main investigating characters of Asteraceous taxa (Karaismailoglu, 2015). Therefore, these micromorphological features of cypsela in

Asteracea could be important diagnostic features for solving different taxonomic relationships within the family. (Ghimire, Suh, Lee, Heo, & Jeong, 2018).

Cypsela morphology has been extensively used as a distinctive taxonomic tool by many researchers for the identification of groups and genera in tribe Anthemideae (Asteraceae) (Cassini, 1823). Likewise, similar study has been conducted on the seed coat and fruit wall of different species of tribes Mutisieae, Cichoreae, and Cynareae (Lavialle, 1912). Extensive work on the Cypsela morphology of several tribes (Compositae) has been conducted from various regions of the world by many researchers (Pak, Park, & Whang, 2001). Variation has been also found in characteristic features of cypselae in Asteraceae for example in the tribe Cynareae, the form of the cells of the testal epidermis and the types of thickenings of their walls provide taxonomically useful information (Kadereit & Jeffrey, 2007).

Apart from this, some work has been conducted on Cypsela morphology representing the family Asteraceae such as Inuleae (Abid & Qaiser, 2002), Anthemideae (Abid & Qaiser, 2009), Senecioneae (Abid & Ali, 2010) and Mutiseae (Abid & Alam, 2011) from Pakistan (Kashmir). In spite of this, limited data exist on the Cypsela morphology of some genera of tribe Lactuceae. For instance, (Abou-El-Naga, 1997) examined the taxonomic significance of 40 carpopodium species of tribe Cichoreae, while (Haque & Godward, 1984) examined the detailed carpopodia of family Asteraceae (2 subfamilies, 10 tribes, 18 genera, and 40 species). The results of their study showed that the subfamily Lactucoideae possess narrower, smoother carpopodia containing limited cells being originated via sideways fusion of unconnected segments. Additionally, LM and SEM has been used to conduct Cypsela seed surface sculpturing and wall anatomical studies of 14 species (genus Lactuca L.) (Zhu et al., 2006) to access their inter- and intrageneric relations among each other. Based on the literature survey, extensive work has been done on the Cypsela morphology of different tribes of Compositeae (including Cichoreae) nationally and internationally but no detailed information is available on the Cypsela morphology of the species discussed here particularly from Pakistan. The aims of the current study are, therefore, to provide a detailed explanation of the Cypsela morphology

and to decide the level to which these micromorphological data can be efficiently used as a taxonomic character to delimit various genera of tribe Cichoreae.

# (B) Fabaceaous Seeds Morphology

Seed characters, mainly outer surface features disclosed by means of scanning electron microscopy (SEM), have been widely employed in solving various problems in systematics and phylogenetic relationship of species (Segarra & Mateu, 2001). The significance of investigating ultrastructural pattern of the seed coat under SEM has been well known for reviewing phenetic linkages and classification of species (Koul, Nagpal, & Raina, 2000; Yoshizaki, 2003). SEM involves high magnification with fine details of ultrastructure that would allow the detection of qualitative variations not possible to observe under optical microscopy (Belhadj et al., 2007). SEM has brought revolutionary trends in study of microscopic organisms with many advantages side by side. The use of SEM was employed to examine the seed borne fungi by Alves and Pozza (2009). Rashid et al. (2018) also reported the use of SEM for systematic studies in tribe Vicieae (Papilionoideae). Souza and Marcos-Filho (2001) gave a comprehensive view of the modulatory role played by mature seed coat in Fabaceae and reported that seed cot structures differ among species and varieties. Erkul, Celep, and Aytaç (2014) examined seeds of 13 Oxytropis DC. species from Turkey using Light and SEM to evaluate the taxonomic relevance of macromorphological and micromorphological seed characters. The study recorded variation in seed size, shape, surface sculpturing pattern, hilum position, and weight. Kirkbride, Gunn, Weitzman, and Dallwitz (2000) developed a CD-ROM that includes an interactive key for the identification of seeds in the 686 legume genera. The characteristics of seeds in the legume family provide an accurate means for their identification. Recently, SEM has been widely used in investigating micromorphological characters of fruit and seed coats (Kaya, Ünal, Özgökçe, Doğan, & Martin, 2011). According to literature, brief information is available on seed morphological characters in tribes of subfamily Papilionoideae (Bortoluzzi, Carvalho-Okano, Garcia, & Tozzi, 2004), so the aim of present study was the use of SEM techniques to search for seed characters constructive in identification of studied taxa.

# 1.6 Significance of Pollen and Seeds Using LM and SEM

Although light microscopy continues to be a central element of life science, light microscopy is often regarded as an old technology. Although light microscopy is actually 400 years old, this technology is still evolving and the potential of biological science is not yet fully revealed. The purpose of a light microscope is to provide a magnified image of an illuminated or luminescent sample in the visible spectrum range or adjacent UV or near infrared spectral range. Optical magnification is achieved by passing light through the lens. Since the 1880s, light microscopes have been able to reach the theoretical limit of optical resolution. Unfortunately, many users of biological science are not yet aware of the importance of this technology and continue to receive poor quality images instead. (Dawe et al., 2006).

SEM can provide information Surface topography, crystals Structure, chemical composition And the electrical operation of the top surface A sample of about 1 jar. various Special stages (eg hot, cold, or Designed to allow on-site mechanical testing) Can be installed to enable performance under a variety of conditions Check the status. For example, cathode fluorescence At (light emission) temperature There are many that are close to absolute zero Images stronger than room temperature Light emitted from a cold sample Much quieter Other benefits of SEM. SEM has very profound benefits Most of the samples because of the field Surfaces that are in focus at the same time anything Surface roughness. Optical microscopes operating at high magnification have a very shallow depth of field, so image quality relies heavily on smooth surfaces. The final resolution is 1nm, which can achieve a much higher magnification (up to 1,000,000x). The maximum magnification that can be used with an optical microscope is about 1000 times. An opportunity to learn more than just the surface terrain. Crystal structure, chemical composition and electrical properties. By switching between different imaging methods, you can ensure that the information is cross-correlated (Parry, 2000).

### 1.7 Background Justification of The Study

Potohar region all over the world blessed with unique type of biodiversity including medicinal plants and have a different type of climatic conditions. In potohar regions of Pakistan most of the unique biodiversity exist and distributed across diverse type of landscape and habitats. Due to extreme temperature ranges the vegetation is the area is totally sparse but have a potential of medicinal flora. These medicinal plants resources are needed to be explored and correctly identified for promoting the public health. In potohar region, conservation methodologies are required intensely to preserve the medicinal plant resources. Due to the rich cultural heritage and diversity of medicinal plants in potohar regions of Punjab Pakistan, the proposed project confined to document floral diversity and systematic studies of Plants belonging to different families to solve the taxonomic issues of the Potohar region.

Lesser work on taxonomic studies carried out systematically in this region. This study aims to identify the plants with the help of basic taxonomic tools including morphology, Pollen Morphology and seeds morphology. Without these studies, these valuable plants would have either remained restricted to certain area or may have even extinct without being known to science. The main purpose of the study is to explore the indigenous knowledge of local communities of North Punjab regarding the use of plants which were never surveyed prior to this investigation. The medicinal plants are known to local people. There is a dare need to conserve the wild medicinal plant resources and their associated knowledge. So, this project is an attempt to create awareness among the people about the medicinal values, Palynology and Seeds morphology of these plants of Potohar region, so that this heritage may be wisely used and conserved and perpetuated through judicious management in health sector.

# 1.8 Objectives of The Present Project

The objectives of the research project are as follows

- Preparation of a detailed checklist of floral diversity of Potohar-Plateau regions of Nothern Punjab, Pakistan.
- The number of dominant families including (Asteraceae, Amaranthaceae, Fabaceae, Poaceae) from the Potohar Region characterized using detailed Pollen Morphology and Seeds Morphology.
- Developing a field guide consisting of unique floral pictorial for future studies regarding important flora of Potohar-Plateau

- Application of field photography of Plant species and Morpho-Palynology and Seeds Morphology via LM and SEM for correct identification.
- Construct Taxonomic Keys to Compare different taxonomic characters of the selected plant species to compare the results of LM and SEM.

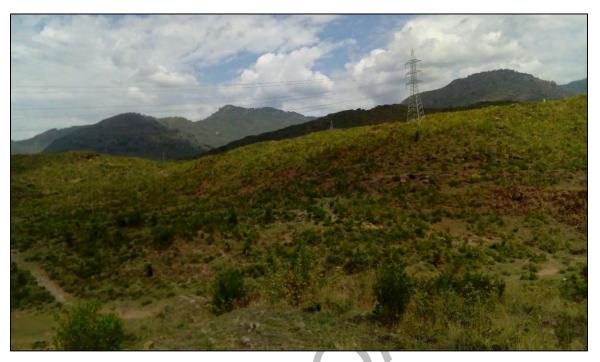


Plate 1: Panoramic View of collection site at QAU, Islamabad

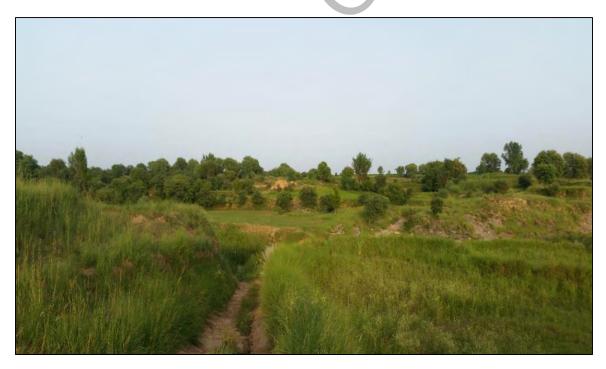


Plate 2: Panoramic View of collection site at Chakwal (Thoha Bahadur)



Plate 3: Collection area of grasses in Kallar kahar (Chakwal)

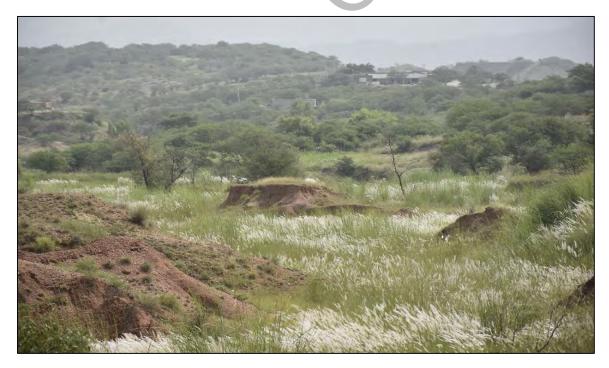
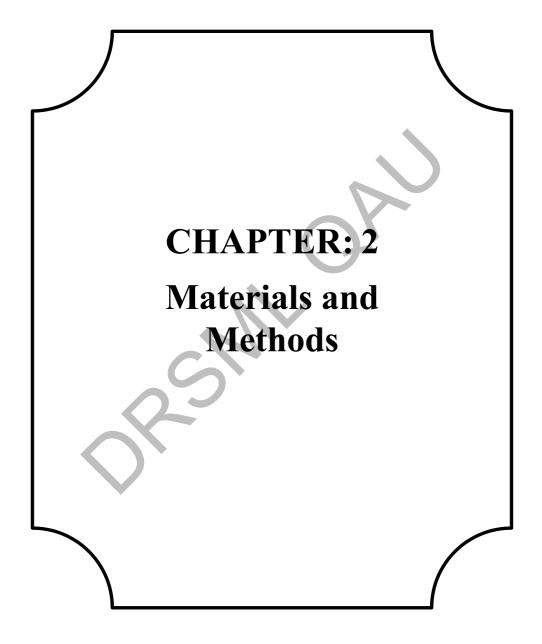


Plate 4: Collection area of Grasses in soon Sakaser



#### 2.1 Research Work Outline

This section summarizes the methodology of the research Project. It briefly describes the selection of the study area, sampling technique and experimentation protocols. It also gives source of sample data, method of data collection and analysis. The main objective of the study is to examine the Taxonomic Characterization of Floral Diversity of Potohar- Plateau. The Laboratory analysis of the selected plants was performed in Plant systematics & Biodiversity Laboratory of Quaid-i-Azam University Islamabad. The study mainly focuses on the Morphology of Plants, Palynological, and Seed morphological perspectives.

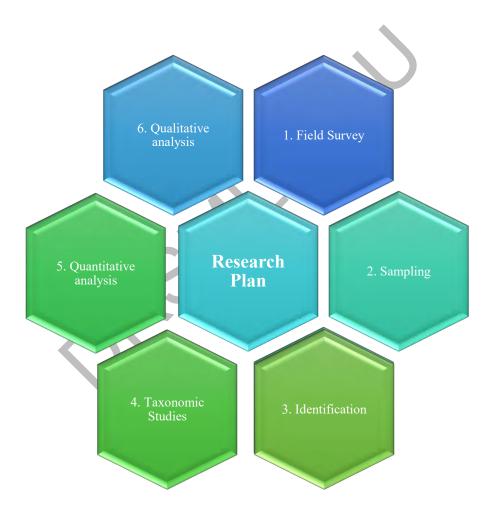


Figure 1: Flow sheet showing research outline

### 2.2 Study Area

Potohar-Plateau in Punjab province,Pakistan that drives name due to extensive rock salt deposits( Ahmad et.,al, 2007). Potohar Plateau of North Punjab ranges over the administrative districts of Mianwali,Chakwal,Khushab,and Attock. The Potohar-Plateau of North Punjab is located between 32° 40° 30° North and 72° 47° 35° East. Potohar Plateau of North Punjab is situated in the subtropical region with height ranging from 250 to 1520m (Chaudhary et.,al, 1969). Potohar Plateau of North Punjab extends from the Jhelum river in the Barkala ridges and Tilla jogian which runs southwards and cross the Indus river near Kalabagh (Ahmad and Waseem, 2006).

The Potohar-Plateau contains two distinctive hill tracts present in the east to south west direction separated by about 5 miles distance. The mountains of Potohar-Plateau running parallel to each other intervened by spurs,ridges, and hills with various lakes and valleys of the various phytogeographic and agroclimatic authorities. The three prominent lakes include Uchali lake, Kallar kahar and Khabikki lake. The Potohar-Plateau in the northern part of the Punjab Province includes great mines of Khewra, that yield great supplies of salt.

# 2.3 Climate of the Study Area

Potohar-Plateau of North Punjab has low annual precipitation of about 50cm. Rainfall is mostly confined to the months of june,July and August. The hills of Soon Sakesar in the salt range receive maximum rainfall due to their height. The study areas are warm temperate in nature. Extreme temperature is noticed between winter and summer. The average maximum temperature is 40 c(July) and the average minimum temperature is 10 C(January). Hot dry winds are common and drought continues for a long time in the area. The long winter season exists with frost (Ahmad et al., 2007).

# 2.4 Schematic Sketch of Present Study

The overall design for current research study is presented in given Figure 2.1

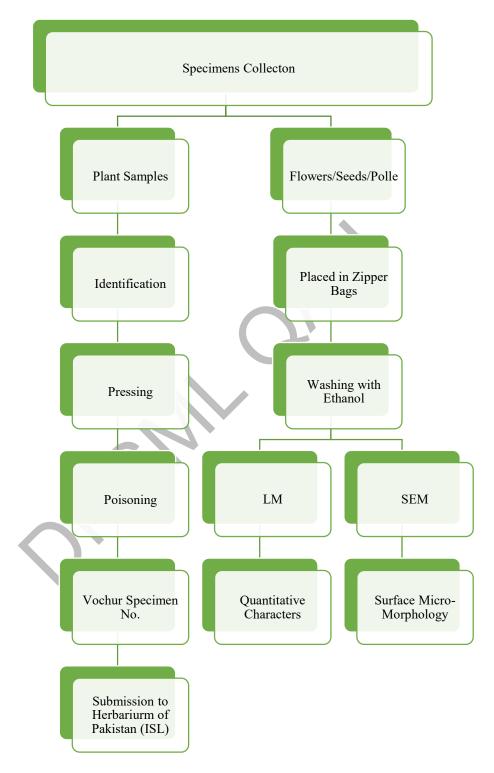


Figure 2: Schematic Sketch of present project

# 2.5 Field survey and Plants Collection

The Study was confined to the Plant Morphology, Palynology and seed Documentation of Floral Diversity in Potohar Plateau Pakistan. The Study Sites of Potohar Plateau includes Attock, Chakwal, Mianwali, Soon sakasar, Miani Village, Uchali Lake, Kabbaki Lake, Kalla Chitta Hills, Rawalpindi and Islamabad (Map). A number of trips to the fields were carried out during various blossoming seasons in the years (2020-2022). Plants with floral parts were collected, pressed, dried and placed in blotting papers and newspapers. Field Data was recorded in the field notebook regarding date of collection, field number, locality, habit, Habitat, Phytography, Flower color and Flowering Period. The particulars of source and collection with GPS data of the collected plant species are presented in Table 1.

# 2.6 Herbarium Preparation and Plant Identification

The collected plants were pressed, dried, poisoned, labelled and mounted on herbarium sheets and deposit in Herbarium of Pakistan (ISL), Quaid-i-Azam University Islamabad for future reference. In case of any ambiguities voucher specimens were compared with type specimens available at Herbarium of Pakistan (ISL), QAU Islamabad. Voucher specimens of all the documented species were properly processed and submitted to Herbarium of Pakistan (ISL), QAU Islamabad. The collected plants were identified based on their different morphological characteristics and cross verified with the samples already record documented in the herbarium and consulting Flora of Pakistan given by Ali and Qaiser, (1993-2010).

# 2.7 Systematics Studies of Floral Diversity of Potohar-Plateau

### 2.7.1 Palynological Studies (LM & SEM)

Palynological studies were carried out by adopting following procedures.

### a) Pollen Material

The pollen material used in this research study was obtained from fresh collection containing flowers collected from various regions of Potohar-Plateau Pakistan. The details and origin of studied plant species have been presented in Table 2.1. In order to separate the pollen grains from anthers, stereo microscope was used.

### b) Glycerin Jelly Preparation

Preparation of glycerin jelly for palynological studies was carried out by using the methodology of Meo and Khan (2005). For glycerin jelly preparation about 30 gm of gelatin was boiled in 50 ml of distilled water. After boiling for <sup>1</sup>/<sub>2</sub> an hour, 40 gm of glycerin and few drops of phenol crystals were added to the mixture. For pink color 0.1 % safranin was added and then mixture was stabilized at room temperature for later use.

### c) Light Microscopic (LM) analysis of Pollen

Mature flowers were used for pollen morphological studies. The anthers were acetolyzed with 99.9% pure acetic acid then cursed to release pollen from anthers and debris was removed. Then drop of glycerin jelly was put on acetolysed pollen and slides were covered with cover slips and after these slides were permanently sealed with transparent nail paint. Tagging tape was used for giving slide number or name (Khan et al., 2020). Prepared slides were observed under light microscope (Meiji Techno MT4300H) and microphotography was carried out using Leica microscope fitted camera Meiji Infinity 1. Both qualitative and quantitative features were studied (Ahmad et al., 2013; Bahadur et al., 2018; Khan et al., 2021).Terminologies for pollen morphology were described according to Barthlott (1984), Erdtman (1960) and Ronald (2000).

#### d) Scanning Electron Microscopic Analysis (SEM) of Pollen

For SEM studies mature anthers were placed in the middle of clean glass slide, 1-2 drops of acetic acid were added and left for a minute. Anthers were then crushed to release the pollen from anthers, debris was removed and extracted pollen were transferred to stubs. Coating with gold was done in sputtering chamber using SPI-MODEL<sup>TM</sup>. After gold coating, stubs were placed in Jeol Vacuum –evaporator for 15 minutes and observed under Scanning Electron Microscope (Model JEOL JSM- 5910) (Butt et al., 2018; Hussain et al., 2019). Pollen exine ornamentation was studied following the terminally of Bahadur et al., (2018).

### e) Pollen Fertility and Sterility Estimation

Estimation of pollen fertility and sterility percentage was calculated by using formula adapted by Ullah et al., (2018).

Percentage of fertility = Fertility /F + SX 100Percentage of Sterlity = Sterility /S + FX 100

Where F represents fertile pollen numbers and S represents sterile pollen numbers on slide.

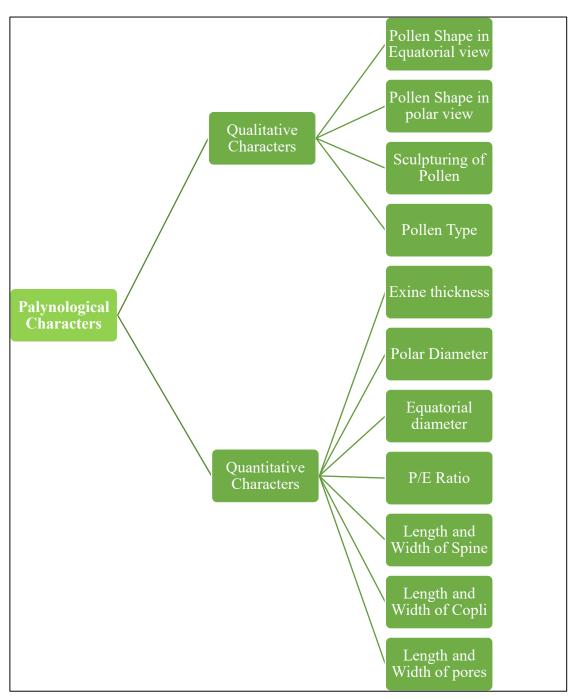


Figure 3: Schematic Representation of Pollen Parameters Characterization

# 2.7.2 Seed Morphological Studies

#### a) Taxon Sampling

In order to obtain seeds, mature seeds of plants were removed from fruits collected during field visits in fruiting season.

#### b) Light Microscopic Studies of Seed Morphology

Mature seeds of plants were washed through different grades of ethanol (60-80%) for 1 to 3 minutes to remove the debris on their surface. External color, Shape, Size (length & width), Hilum were observed using Binocular Dissecting microscope, placed in plant anatomy Lab Department of Plant sciences Quaid-i-Azam University, Islamabad, Pakistan.

#### c) Scanning Electron Microscopic Studies of Seed Morphology

For SEM studies seeds were immersed in absolute ethanol and left for a minute to completely dry. This step was carried out for detailed study of invasive seed morphology. Then seeds were mounted on stubs in different positions and gold coating was performed in sputtering chamber using SPI-MODEL<sup>TM.</sup> In all cases seeds of at least 10 samples of each species were analyzed, characterized and photographed with a Scan Electron Microscope (Model JEOL JSM- 5910). The seed characters observed were length of main axis, position of hilum, surface sculpture.



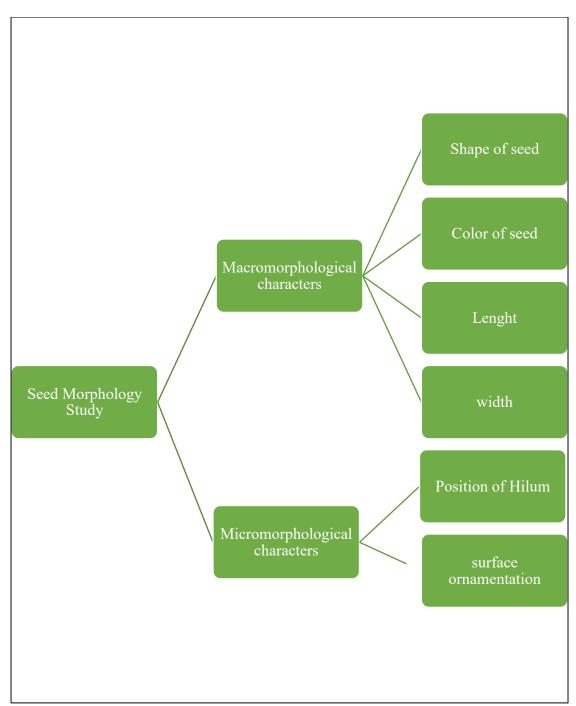


Figure 4: Parameters studied for seed morphological characters.



Plate 5: Panoramic view of Kallar Kahar



Plate 6: View of agricultural fields of Miani village



Plate 7: Diversity of plants near Uchali Lake



Plate 8: Pictorial view of Khabeki Lake



Plate 9: Wild flora near Numl Lake



Plate 10: Pictorial view of Vegetation on Kala Chitta Hills



Plate 11: Paranomic view of Soan Sakaser Valley



Plate 12: Diversity of plants of Khewra Salt Range



Plate 13: Plant collection during field surveys



Plate 14: Experimental work in laboratory to study pollen morphology



Plate 15: Quantitative data analysis of pollen using Microscope



Plate 16: Qualitative data analysis by studying morphological characters of plants



Plate 17: Light microscopic study of pollen grains



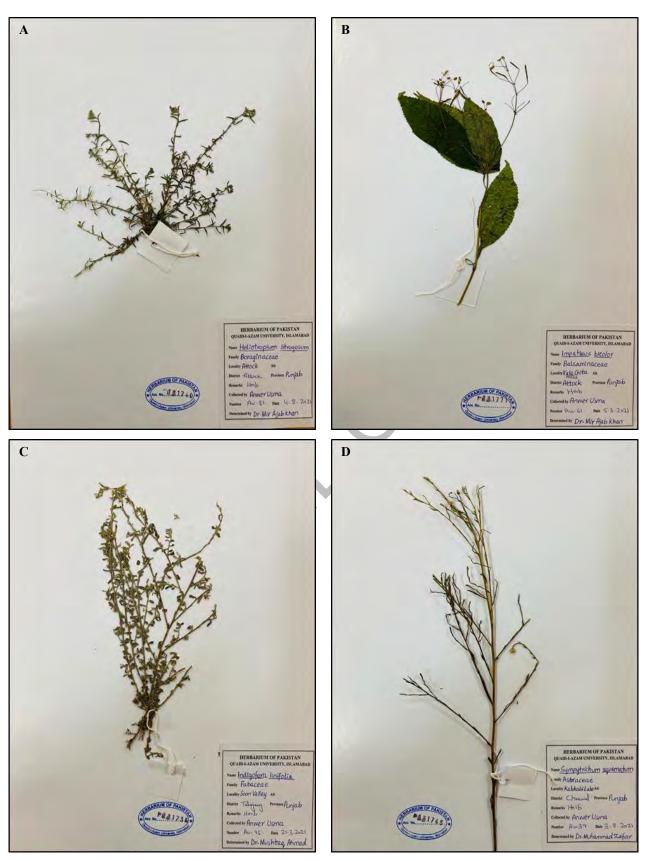
Plate 18: Morphological study of plants using stereo microscope



Plate 19: Preservation of plant specimen



Plate 20: Mounting of plants on herbarium sheets



**Plate 21:** Herbarium specimen photographs of (A) *Heliotropium strigosum*, (B) *Impatiens bicolor*, (C) *Indigofera linifolia*, (D) *Symphyotrichum squamatum* 



**Plate 22**: Herbarium specimen photographs of (A) *Dactyloctenium scindicum*, (B) *Croton bonplandianum*, (C) *Chrozophora plicata*, (D) *Cleome brachycarpa* 

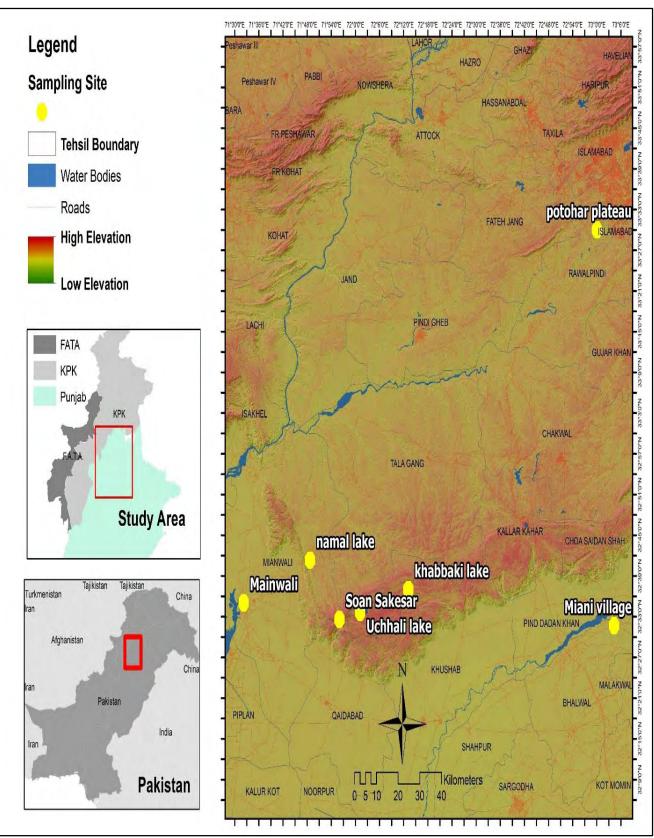


Figure 5: Map of study area showing collection sites.

Sr.No	Plant Taxa	Family	Locality	Voucher No	Accession No
1.	Ageratum conyzoides (L.) L.	Asteraceae	Khallar Kahar	AU-51	ACC-131695
2.	Artemisia annua L.	Asteraceae	Kalla chitta hills	AU-67	ACC-131696
3.	Aster squamatus (Spreng.) Hieron.	Asteraceae	Rawalpindi	AU-63	ACC-131697
4.	Bidens bipinnata L.	Asteraceae	Attock	AU-81	ACC-131698
5.	Bidens pilosa L.	Asteraceae	Attock	AU-23	ACC-131699
6.	Calendula arvensis M.Bieb.	Asteraceae	Uchali lake	AU-97	ACC-131700
7.	Calendula officinalis L.	Asteraceae	Soon sa Qaisar	AU-53	ACC-131701
8.	Carthamus lanatus L.	Asteraceae	Islamabad	AU-95	ACC-131702
9.	Carthamus oxyacantha M.Bieb.	Asteraceae	Numl Lake	AU-35	ACC-131703
10.	Cirsium arvense (L.) Scop.	Asteraceae	Soon Valley	AU-29	ACC-131704
11.	Cousinia prolifera Jaub. & Spach	Asteraceae	Chakwal	AU-25	ACC-131705
12.	Eclipta prostrata (L.) L.	Asteraceae	Moosa Khail Mianwali	AU-41	ACC-131706
13.	Erigeron bonariensis L.	Asteraceae	Islamabad	AU-91	ACC-131707
14.	Erigeron canadensis L.	Asteraceae	Islamabad	AU-39	ACC-131708
15.	Ixeris polycephala Cass.	Asteraceae	Kalla chita Hills	Au-51	ACC-131709
16.	Lactuca sativa L.	Asteraceae	Margalla Hills	Au-53	ACC-131710
17.	Lactuca serriola L.	Asteraceae	Soon Valley	Au-57	ACC-131711
18.	Launea naudicaulis Hook.f.	Asteraceae	Khallar kahar	Au-61	ACC-131712
19.	Launea procumbens Roxb.	Asteraceae	Margalla Hills	Au-65	ACC-131713
20.	Parthenium hysterophorus L.	Asteraceae	Islamabad	AU-13	ACC-131714
21.	Silybum marianum (L.) Gaertn.	Asteraceae	Islamabad	AU-107	ACC-131715
22.	Sonchus arvensis L.	Asteraceae	Mianwali	Au-67	ACC-131716
23.	Sonchus asper (L.) Hill.	Asteraceae	Margalla Hills	Au-71	ACC-131717
24.	Sonchus oleraceous (L.) L.	Asteraceae	Soon Valley	Au-79	ACC-131718
25.	Sphagneticola trilobata (L.) Pruski	Asteraceae	Chakwal	AU-15	ACC-131719
26.	Taraxacum offinale (L.) Weber ex F.H.Wigg.	Asteraceae	Chakwal	Au-81	ACC-131720
27.	Taraxacum platycarpum Dahlst.	Asteraceae	Soon valley	Au-85	ACC-131721

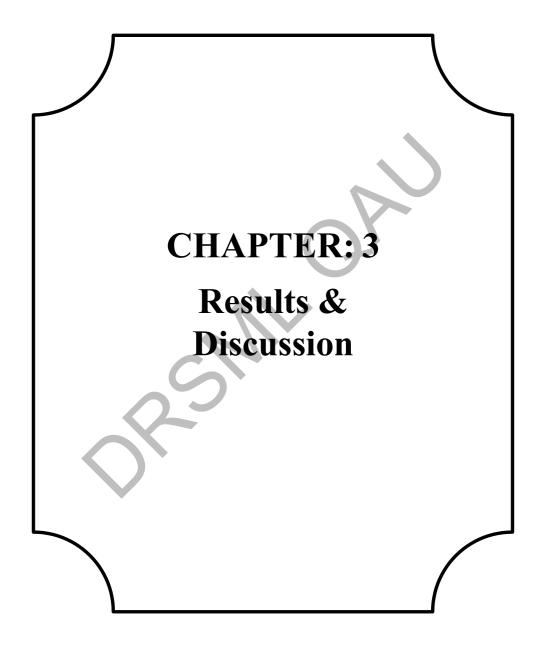
 Table 1:Check List of Floral Diversity of Potohar-Plateau with Collection Site.

28.	Verbesina helianthoides Michx.	Asteraceae	Mianwali	AU-109	ACC-131722
29.	Zinnia elegans L.	Asteraceae	Salt range Chakwal	AU-118	ACC-131723
30.	Achyranthus aspera L	Amaranthaceae	Islamabad	Au-135	ACC-131724
31.	Aerva javanica Hill	Amaranthaceae	Salt Range, Chakwal	Au-37	ACC-131725
32.	Amaranthus gracizens Linn	Amaranthaceae	Salt Range, Chakwal	Au-95	ACC-131726
33.	Amaranthus hybridus L.	Amaranthaceae	Salt Range Chakwal	Au-91	ACC-131727
34.	Amaranthus spinosus L.	Amaranthaceae	Salt Range Chakwal	Au-137	ACC-131728
35.	Amaranthus viridis L	Amaranthaceae	Salt Range Chakwal	Au-39	ACC-131729
36.	Atriplex stocksii (Forssk.) Stapf.	Amaranthaceae	Salt Range, Chakwal	Au-179	ACC-131730
37.	Celosia argentia L	Amaranthaceae	Salt Range, Chakwal	Au-55	ACC-131731
38.	Chenopodium album (L.) Stapf.	Amaranthaceae	Islamabad	Au-66	ACC-131732
39.	Chenopodium murale (L.) P.Beauv.	Amaranthaceae	Chakwal	Au-83	ACC-131733
40.	Chenopodium ambrosioides L.	Amaranthaceae	Islamabad	Au-61	ACC-131734
41.	Digeria muricata L	Amaranthaceae	Rawalpindi	Au-59	ACC-131735
42.	Kochia indica L	Amaranthaceae	Salt Range, Chakwal	Au-231	ACC-131736
43.	Kochia lonifolia Linn	Amaranthaceae	Salt Range, Chakwal	Au-107	ACC-131737
44.	Pupalia lappaceae L	Amaranthaceae	Salt Range, Chakwal	Au-91	ACC-131738
45.	Salsola lubricate L	Amaranthaceae	Miani Village Chakwal	Au-65	ACC-131739
46.	Sauda fruitcosa L	Amaranthaceae	Salt Range, Chakwal	Au-111	ACC-131740
47.	Acacia nilotica L	Fabaceae	Islamabad	Au-211	ACC-131741
48.	Accacia modesta L	Fabaceae	Kabekhi Lake	AU-212	ACC-131742
49.	Arachis hypagae L	Fabaceae	Numl Lake	AU-205	ACC-131743
50.	Astragalus psilocentros L	Fabaceae	Soon valley	AU-206	ACC-131744
51.	Astragalus scoripiurus L	Fabaceae	Mianwali	AU-207	ACC-131745
52.	Cassia fistula L	Fabaceae	Salt range Chakwal	AU-168	ACC-131746
53.	Cassia italic L.(Pers)	Fabaceae	Islamabad	AU-169	ACC-131747
54.	Cassia occidentalis Linn	Fabaceae	Salt Range, Chakwal	AU-51	ACC-131748
55.	Crotolaria burhia (Jones) schult	Fabaceae	Salt Range, Chakwal	AU-53	ACC-131749
56.	Dilbergia sisso Jaub. & Spach	Fabaceae	Salt Range Chakwal	AU-61	ACC-131750
57.	Lathyrus aphaca L	Fabaceae	Salt Range Chakwal	AU-251	ACC-131751
58.	Lespedeza juncea L	Fabaceae	Salt Range Chakwal	AU-253	ACC-131752
59.	Leucaena leucocephalla L	Fabaceae	Salt Range, Chakwal	AU-191	ACC-131753
60.	Melilotus indicus L	Fabaceae	Kalla-Chitta Hills	AU-193	ACC-131754
61.	Pigomia pinnata L	Fabaceae	Qau, Islamabad	AU-45	ACC-131755

62.	Prosopis cineraria L	Fabaceae	Mianwali	AU-47	ACC-131756
63.	Prosopis juliflora Linn	Fabaceae	Mianwali	AU-59	ACC-131757
64.	Pueraria tuberosa L	Fabaceae	Soon Valley	AU-241	ACC-131758
65.	Rhynchosia capitata L	Fabaceae	Chakwal	AU-117	ACC-131759
66.	Sophora mollis (Jones) schult	Fabaceae	Chakwal	AU-119	ACC-131760
67.	Tephrosia purpurea (Jones) schult	Fabaceae	Salt Range, Chakwal	AU-207	ACC-131761
68.	Vicia angustifolia (Jones) schult	Fabaceae	Khallar Kahar	AU-09	ACC-131762
69.	Brachiaria brizantha L	Poaceae	Margalla Hills	Au-11	ACC-131763
70.	Cenchrus biflorus L	Poaceae	Soon Valley	Au-15	ACC-131764
71.	Cenchrus ciliaris L	Poaceae	Khallar kahar	Au-17	ACC-131765
72.	Cenchrus pennisetiformis L	Poaceae	Margalla Hills	Au-21	ACC-131766
73.	Cymbopogon jwarancusa L	Poaceae	Islamabad	Au-19	ACC-131767
74.	Cynodon dactylon L	Poaceae	Islamabad	Au-12	ACC-131768
75.	Dactyloctenium aegyptium L	Poaceae	Mianwali	Au-23	ACC-131769
76.	Dichanthium annulatum L	Poaceae	Margalla Hills	Au-25	ACC-131770
77.	Eleusine indica Linn	Poaceae	Soon Valley	Au-27	ACC-131771
78.	Pennisetum glaucum L	Poaceae	Chakwal	Au-29	ACC-131772
79.	Pennisetum typhoides L	Poaceae	Chakwal	Au-31	ACC-131773
80.	Poa annua (L.) P.Beauv.	Poaceae	Soon valley	Au-33	ACC-131774
81.	Saccharum spontaneum L.	Poaceae	Mianwali	Au-51	ACC-131775
82.	Steria viridis (L.) P.Beauv.	Poaceae	Salt range Chakwal	Au-9	ACC-131776
83.	Zea mays L.	Poaceae	Islamabad	Au-7	ACC-131777
84.	Anagallis arvensis L	Primroses	Salt Range Chakwal	AU-107	ACC-131778
85.	Asphodelus tenuifolius Cav.	Asphodelacea	Miani Village Talagang	AU-113	ACC-131779
86.	Boerhivia procumbense L	Nyctaginaceae	Khewra mines	AU-115	ACC-131780
87.	Brachychiton acerifolius (A.Cunn. ex	Malvaceae	Uchali Lake	AU-99	ACC-131781
	G.Don)				
88.	Broussonetia papyrifera (L.) L'Hér. ex	Moraceae	Uchali Lake	AU-77	ACC-131782
	Vent.				
89.	Bryophyllum pinnatum (Lam.) Oken	Crassulaceae	Rawalpindi	AU-65	ACC-131783
90.	Capparis decidua (Forssk.)edgeW.	Capparaceae	Musa khel Mianwali	AU-39	ACC-131784
91.	Chorchorus depressus L	Tiliaceae	Miani Village	AU-55	ACC-131785
92.	Cissampelos pareira L.	Menispermaceae	Kabkbi Lake	AU-110	ACC-131786
93.	Citharexylum spinosum L.	Verbenaceae	Islamabad	AU-205	ACC-131787

94.	Cleome brachycarpa (Forssk.) Vahl ex	Capparaceae	Salt Range	AU-209	ACC-131788
	DC				
95.	Cleome viscosa L.	Capparceae	Attock	AU-211	ACC-131789
96.	Commelina benghalensis L.	Commelinaceae	Salt Range	AU-215	ACC-131790
97.	Convolvulus arvensis L.	Convolvulaceae	Islamabad	AU-217	ACC-131791
98.	Cucumis melo sub agerestis L.	Cucurbitaceae	Miani village Talagang	AU-31	ACC-131792
99.	Datura innoxia Mill.	Solanaceae	Miani village Talagang	AU-45	ACC-131793
100.	Euphorbia granulata L.	Euphorbiaceae	Miani village Talagang	AU-53	ACC-131794
101.	Oxalis corniculata L.	Oxalidaceae	Thoha Bahadur	AU-69	ACC-131795
			Chakwal		
102.	Solanum surattense Mill	Solanaceae	Miani Village Chakwal	AU-51	ACC-131796
103.	Trianthema portulastrum L	Aizoaceae	Salt Range Chakwal	AU-57	ACC-131797

sait Range Chakwal A



#### **Summary**

The Floral Diversity of Potohar Plateau, Pakistan has been bestowed with rich floristic diversity. The current research was conducted in the selected areas of Potohar Plateau i.e. Attock, Chakwal, Mianwali, Soon Sakasar, Miani Village, Uchali Lake, Kabbaki Lake, Kalla Chitta Hills, Numl Lake. These areas were explored first time systematically. Previous work was fragmented but the present research project is comprehensive and includes the Taxonomic study of 103 Plants belonging to Fifteen Families. Dominant families of my research project include Asteraceae, Amaranthaceae, Fabaceae and Poaceae. The results are compiled and presented separately in two sections.

**Section 1:** Qualitative and Quantitative pollen micro-morphological characters. Qualitative Characters include Pollen type, Pollen shape, Exine sculpturing, Orientation of Pores while Quantitative characters include Polar Diameter, Equatorial Diameter, Colpi Length, Colpi Width, Length and Width of Pores, P/E ratio and Exine thickness, Pollen fertility and sterility, Length and Width of Spines.

Section 2: Micromorphological seed features such as shape, color, size, surface texture, apex, hilum position, Anticlinal wall, Periclinal wall, cell outline, cell arrangement, Projections.

## **SECTION 1:**

# Palynology (LM & SEM)

#### 3.2 Pollen Micro-morphological Studies of Floral Diversity of Potohar Plateau

Here we describe in this section, the detailed Pollen micro-morphology of 103 Plants belonging to Fifteen Families using scanning electron microscopy and Light microscopy. Among them, Dominant families include Asteraceae (29 Species), Amaranthaceae (17 Species), Fabaceae (22 Species), Poaceae (15 Species) and remaining species belonging to different families including Cucubitaceae, Capparaceae, Brasicaceae, Solanaceae, Menispermaceae, Euphorbiaceae, Chenopodiaceae, Tiliaceae (each with one and two species).

Pollen micro-morphological characters of Plants analyzed here include Pollen type, shape, size, Sculpturing, apertural membrane, Spines, Lacunar shape, Echini shape, Echini arrangement, Polar Diameter, Equatorial Diameter, P/E ratio, Exine thickness, Length and width of Spines, Length and width of Colpi, Length and Width of Pores. It also includes Pollen fertility and sterility percentages.

In this section include five parts

- 3.1.1 Asteraceous Pollen Micro-Morphology
- 3.1.2 Amaranthaceous Pollen Micro-Morphology
- 3.1.3 Fabaceous Pollen Micro-Morphology
- 3.1.4 Poaceous Pollen Micro-Morphology
- 3.1.5 Weeds Pollen Micro- Morphology

	Taxa	Shape	Polar	Pollen Type	Polar shape/	Pollen	Echini	Lacunae	Lacunar	Echini	Inter-
No			View		Orientation	ornamentation	arrangement	sculpturing	shape	Shape	lacunar
											Gap
	Ageratum	Oblate-spheroidal	Circular	Tricolporate	Elliptical/sunken	Echinate	Regular	Absent	Absent	-	Absent
6	conyzoides										
<b>2.</b>	Artemisia	Prolate-spheroidal	Angular	Trizonocolporate	Elliptical/sunken	Echinate	Regular	Absent	Absent	-	Absent
C	absinthimum										
3. 4	Aster squamatus	Prolate-spheroidal	Triangular	Tricolporate	Elliptical/slightly	Echinate	Rather regular	Absent	Absent	-	Absent
					sunken						
<b>4.</b> <i>1</i>	Bidens bipinnata	Prolate-spheroidal	Triangular	Tricolporate	Rounded/elliptic	Echino-lophate	Regular	Fenestrated	Pentagonal	-	Slightly
					al sunken	perforate		perforate			narrow
<b>5</b> . <i>1</i>	Bidens pilosa	Spheroidal	Circular	Tricolporate	Rounded/elliptic	Echino-lophate	Regular	Fenestrated	Hexagonal	-	Narrow
					al sunken	perforate		perforate			
6. (	Calendula	Oblate-spheroidal	Circular	Tricolporate	Rounded/sunken	Echinate	Regular	Absent	Absent	-	Absent
6	arvensis										
7. (	Calendula	Oblate-spheroidal	Semi	Trizonocolporate	Elliptical/sunken	Echinate	Rather Regular	Absent	Absent	-	Absent
0	officinalis		angular								
8. (	Carthamus	Sub-oblate	Triangular	Tricolporate	Elliptical/sunken	Coarsely regulate	Rather regular	Perforate	Pentagonal	-	Narrow
0	oxyacantha					echinate					
						perforated					
9. (	Carthamus	Oblate-spheroidal	Triangular	Trizonocolporate	Rounded sunken	Echino-lophate	Rather regular	Fenestrated	Pentagonal	-	Narrow
l	lanatus					perforate		perforate	to		
				~					hexagonal		
10. (	Causinia prolifera	Sub-Oblate	Circular	Tricolporate	Rounded sunken	Micro-reticulate	Regular	Absent	Absent	-	Absent
						echinate					
11. (	Cirsium arvense	Oblate-spheroidal	Circular	Tricolporate	Rounded/sunken	Echinate with	Regular	Fenestrates	Tetragonal	-	Slightly
						broad spines		perforate	to		Narrow
									pentagonal		

## Table 2: Qualitative data of pollen of family Asteraceae members from Potohar region of Pakistan

12.	Eclipta prostrata	Prolate-spheroidal	Triangular	Tricolporate	Rounded elliptical/sunken	Echino-lophate	Regular	Fenestrated	Pentagonal	-	Slightly narrow
13.	Erigeron bonariensis	Prolate-spheroidal	Circular	Tetrazonocolpora te	Rounded	Echinate	Regular	Fenestrated	Hexagonal	-	Narrow
14.	Erigeron Canadensis	Prolate-spheroidal	Circular	Tricolporate	Rounded/slightly sunken	Echinate	Rather regular	Absent	Absent	-	Absent
15.	Ixeris polycephala	Oblate-spheroidal	Circular	Tricolporate	Rounded/ Sunken	Echinate	Regular	Absent	Pentagonal	Spines short with narrow bases and surrounded by lacunas	Absent
16.	Lactuca sativa	Prolate-spheroidal	Circular	Tri-tetra Zonocolporate	Rounded	Echinate	Regular	Fenestrated perforate	Tetragonal	Spines long with narrow bases and pointed tips	Narrow
17.	Lactuca serriola	Oblate-spheroidal	Circular to rectangular	Tetracolporate	Ellipitical/ Slightly sunken	Echinate	Regular	Absent	Absent	Spines short with broad bases and around the lacunar	Slightly narrow
18.	Launea naudicaulis	Spheroidal	Semi Angular	Tetracolporate	Rounded/ sunken	Echinate	Regular	Perforate	Hexagonal	Short with narrow bases and mucronate tips	Narrow
19.	Launea procumbens	Oblate-spheroidal	Circular	Tricolporate	Ellipitical/ Slightly sunken	Micro-reticulate Echinate	Regular	Absent	Pentagonal	Long spines with broad bases	Narrow

20. 21.	Parthenium hysterophorus Silybum marianum	Oblate-spheroidal Prolate-spheroidal	Rounded triangular Circular	Tetracolporate Tricolporate	Rounded/sunken Absent	Echino-lophate Microechinate	Rather-regular Regular	Fenestrated perforate Absent	Tetragonal- pentagonal Absent	and sharp narrow pointed tips	Slightly narrow Absent
22.	Sonchus arvensis	Prolate-spheroidal	Cicular	Tetracolporate	Ellipitical/ Slightly sunken	Echinate perforate-tectate	Regular	Perforate	Pentagonal	Short spine with very broad bases and small narrow tips	Slightly narrow
23.	Sonchus asper	Sub-oblate	Triangular	Tricolporate	Rounded/ Sunken	Psilate	Regular	Fenestrated perforate	Pentagonal	Spines with broad bases and surrounded by lacunas	Absent
24.	Sonchus oleraceous	Oblate-spheroidal	Circular	Tetracolporate	Ellipitical/ Sunken	Echino-lophate perforate	Regular	Perforate	Hexagonal	Absent	Narrow
25.	Sphagneticola trilobata	Oblate-spheroidal	Circular	Trizonocolporate	Elliptical/sunken	Echinate-perforate	Regular	Fenestrated perforate	Pentagonal		Narrow
26.	Taraxacum offinale	Prolate-spheroidal	Irregular	Trizonocolporate	Rounded /sunken	Echino-lophate perforate	Regular	Absent	Pentagonal to Hexagonal	Short spines with broad bases	Slightly Narrow
27.	Taraxacum platycarpum	Sub-oblate	Circular	Tricolporate	Rounded/ Sunken	Echinate	Regular	Absent	Pentagonal	Spine have broad bases and sharp pointed mucronate	Narrow

										tips	
28.	Verbisinia eleusoides	Prolate spheroidal	Hexagonal rounded	Tricolporate	Rounded/sunken	Echino-lophate	Regular	Fenestrated	Pentagonal	-	Narrow
29.	Zinnia elegans	Prolate spheroidal	Circular	Tricolporate	Rounded/elliptic al	Echinate with pointed spines	Regular	Fenestrated	Hexagonal	-	Slightly narrow
					SN						

Plant name	Exine thickness Mean (Min-Max) S.E μm*	Polar Diameter Mean (Min-Max) S.E μm	Equatorial Diameter Mean (Min-Max) S.E μm	P/E** ratio	No of Colpi	No of Pores	No of Spines	Spines Length Mean (Min-Max) S.E μm	Spines Width Mean (Min-Max) S.E µm	Colpi Length Mean (Min-Max) S.E μm	Colpi Width Mean (Min-Max) S.E µm	Pores Length Mean (Min-Max) S.E μm	Pores Width Mean (Min-Max) S.E μm
Ageratum	4.8(4.2-6.2)	39.6(31.7-46.5)	40.3(28.2-47.7)	0.9	3	3	35	7.15(3.5-9.2)	2.6(1.7-4)	9.2(5.7-13.7)	10.7(4-17.7)	11.1(7.2-16)	6.8(4.25-
conyzoides	±0.3	±2.6	±3.49	0.9	5	5	55	$\pm 1.0$	±0.42	$\pm 0.4$	±2.40	±1.5	$8.75) \pm 0.78$
Artemisia	3.3(3-3.7)	49.3(42.7-60.5)	47.0(38-50.5)	1.04	3	2	23	2.75(2-3.7)	4.9(4.2-5.5)	7.9(7.2-8.2)	10.1(8.7-	10(9.7-12.2)	12.30(11.2
absinthimum	$\pm 0.1$	±3.23	±2.31	1.04	5	2	23	$\pm 0.30$	$\pm 0.21$	$\pm 0.18$	$11.2) \pm 0.41$	±0.41	5-13) ±0.30
Aster	4.2(3.2-5.2)	22.2(20.5-23.7)	21.4(19.5-23.5)	1.03	3	3	27	2.6(1.7-3.7)	1.6(1.2-2)	5.0(4.2-6) ±0.34	3.0(2.5-3.7)	3(2.7-3.5)	2.45(1.75-
squamatus	$\pm 0.3$	$\pm 0.57$	$\pm 0.74$	1.05	5	5	27	±0.42	±0.12	5.0(4.2-0) ±0.54	±0.21	±0.14	3.25) ±0.28
Biden pilosa	5.0(4.0-6.0) ±0.37	48.2(41.7-53.5) ±2.05	48.0(41.5-58.5) ±3.4	1.00	3	3	32	3.6(2.7-4.7) ±0.34	2.3(1.7-3.0) ±0.23	12.1(8.0-15.7) ±1.42	9.9 (7-14.2) ±1.21	9.0(4.5-12) ±1.31	7.60(3.5- 12.75) ±1.48
Bidens bipinnata	2.2(1.7-2.7) ±0.16	33.4(30.5-35.7) ±0.92	30.9(29.7-31.7) ±0.33	1.07	3	0	0	-	-	8.5(5.5-10.2) ±0.8	10.0(9.2- 10.5) ±0.24	11.1(9.7-13) ±0.57	9.75(9- 10.25) ±0.23
Calendula	2.8(2.5-3)	15.8(14.7-17.2)	16.6(15.2-18)	0.95	3	3	17	2.8(2.2-3.2)	3.5(2.2-4.2)	3.5(2.7-4.7)	3.1(2.75-	3(2.5-3.5)	3.6(3.25-4)
arvensis	$\pm 0.09$	$\pm 0.41$	±0.47	0.95	3	3	1 /	±0.16	$\pm 0.34$	±0.36	$3.50) \pm 0.12$	±0.16	±0.12
Calendula	5.0(4.7-5.2)	51.8(38-56.2)	51.9(38-56.7)	0.99	3	3	16	4.9(4.2-5.2)	7.8(7.2-8.2)	17.6(10.5-21.2)	9.50(8-	12(10.2-13.7)	12(11.25-
officinalis	±0.12	$\pm 3.49$	$\pm 3.52$	0.99	3	3	10	$\pm 0.2$	±0.16	$\pm 1.9$	$10.25) \pm 0.47$	$\pm 0.68$	13) ±0.29
Carthamus	1.9(1.5-2.2)	22.3(22-22.7)	29(27.5-30.5)	0.76	3	3	21	4.3(3.5-4.5)	3.4(3.2-3.7)	3.1(3-3.25)	5.6(5.2-5.6)	4.7(3.5-6.25)	4.7(3.50-
oxyacantha	$\pm 0.14$	$\pm 0.18$	$\pm 0.57$	0.70	3	3	21	$\pm 0.20$	$\pm 0.09$	$\pm 0.06$	±0.23	±0.45	$5.50) \pm 0.40$
Carthamus	5.5(3.5-7.2)	42.6(29.0-54.0)	44.2(39.5-51.7)	0.96	3	3	28	3.7(2.7-4.5)	2.1(1.5-3.2)	7.5(3.5-11.5)	12.1(2.0-	11.6(6.0-	10.35(7.0-
lanatus	$\pm 0.66$	±4.35	±2.12	0.90	3	3	20	$\pm 0.28$	$\pm 0.30$	±1.29	$19.0) \pm 2.89$	$16.0) \pm 1.76$	14.5) ±1.25
Causinia	4.5(3.5-5.2)	41.9(40.7-43.7)	48.2(45.2-50.7)	0.87	3	2	14	5.7(5.2-6.5)	4.6(4.2-4.7)	9.6(8.7-10.2)	7(6.50-7.25)	10.5(10.2-	10.00(9.25-
prolifera	$\pm 0.28$	±0.51	±1.17	0.87	3	Z	14	±0.23	$\pm 0.10$	±0.29	±0.13	$10.7) \pm 0.11$	10.75)

 Table 3: Quantitative data of pollen of family Asteraceae members from Potohar region of Pakistan

													10.050
													±0.250
Cirsium	4.0(3.2-4.7)	43.9(41.2-45.7)	44.6(41.2-51.2)					5.5(5.2-5.7)	4.6(2.7-5.2)	11.2(8.6-15.5)	8.5(5.5-	11.7(10.2-	9.5(8.25-
arvense	±0.26	±0.90	±1.84	0.98	3	1	19	±0.11	±0.48	±1.42	10.7) ±0.99	$12.7) \pm 0.41$	10.50)
											,		±0.40
Eclipta	3.1(2.7-3.7)	40.9(34.7-47.5)	36.9(29.2-44.7)	1.10	3	3	29	2.8(2.5-3.0)	2.6(2.2-3.2)	5.3(5.0-6.0)	8.10(7.0-	9.35(7.25-	8.80(7.75-
prostrata	±0.16	$\pm 2.0$	$\pm 2.88$		-	-		±0.09	±0.20	±0.18	9.0) ±0.34	$10.50) \pm 0.56$	$11.0) \pm 0.60$
Erigeron	1.2(0.7-1.5)	27.5(26.7-28.2)	25.6(25.2-26)	1.07	4	1	29	2.2(2-2.5)	1.8(1.5-2)	11.4(10.75-12)	8.4(8-9)	9.9(9.5-	8.1(7.75-
bonariensis	±0.12	±0.25	±0.12	1.07		1	2)	$\pm 0.07$	±0.10	±0.21	±0.16	$10.50) \pm 0.18$	8.50) ±0.12
Erigeron	2(1.7-2.2)	25.6(24.7-26.7)	23.8(23-24.7)	1.07	3	1	26	1.8(1.5-2)	1.2(1-1.5)	10.1(8.5-12.2)	7.4(7-7.75)	8.70(7.25-	7.60(7.25-
canadensis	$\pm 0.07$	$\pm 0.33$	$\pm 0.32$	1.07	5	1	20	±0.09	$\pm 0.07$	$\pm 0.69$	±0.15	$9.75)\pm0.47$	8) ±0.15
Ixeris	2.5(2.2-	33.2(30.5-35.2)	32.8(29.7-34.7)	1.01	3	4	27	4.4(3.7-5.2)	3.7(3-4.5)	$9.3(7.7-10.5) \pm$	7.4(6.5-8.7)	7.7(6.5-9.7)	6.3(4.7-7.2)
polycephala	2.7)± 0.12	±0.9	$\pm 0.9$					±0.24	±0.3	0.6	±0.4	$\pm 0.5$	$\pm 0.4$
Lactuca sativa	4.05(2.5-	36.7(32-44.7)	38.40(33-45.7)	0.95	3	3	33	6.3(4.5-7.7)	4(2.2-5.7)	8.55(7.2-10)	9.7(6.2-	8.5(6.7-10.7)	6.3(4.5-10)
	5.2) ±0.45	±2.2	$\pm 2.30$					±0.57	$\pm 0.70$	$\pm 0.46$	$13.2) \pm 1.2$	±0.73	$\pm 0.98$
Lactuca	4.1(3.50-	39.6(33.7-45.7)	40.6(34.75-49)	0.97	4	1	31	2.9(2.7-3.2)	1.7(1.2-2.2)	0.8(5.7-10.2)	7(4-9.5)	7.5(6.2-9.7)	8(5.7-11)
serriola	$5.25) \pm 0.30$	±2.24	±2.4					$\pm 0.09$	$\pm 0.17$	$\pm 0.84$	$\pm 1.05$	±0.73	$\pm 0.97$
Launea	4.40(4-	40.9(37.2-40.9)	40.8(34-43.25)	1.00	4	1	29	3.4(3.2-3.7)	1.9(1.5-2.2)	9.1(8.2-9.7)	6.5(5.7-7.2)	8.6(7.7-9.7)	6.9(6.5-7.2)
naudicaulis	4.75) ±0.12	$\pm 1.6$	$\pm 1.71$					$\pm 0.09$	$\pm 0.14$	±0.25	$\pm 0.2$	$\pm 0.38$	±0.12
Launea	4.90(3.50-	61.1(59.2-63.0)	67.1(64.7-71.2)	0.91	3	2	31	2.8(2.2-3.5)	3.7(2.7-4.7)	7.8(7-8.7) ±0.28	9.6(8-11.0)	17.6(13.7-	15.5(13.2-
procumbens	5.75) ±0.38	$\pm 0.67$	$\pm 1.1$					±0.25	±0.35		±0.51	$19.7) \pm 1.07$	17.7) ±0.9
Parthenium	4.6(2.2-6.2)	36.9(30.0-47.0)	40.9(34.2-48.7)	0.00		2	21	4.0(3.2-5.0)	2.9(2.0-4.0)	5.30(3.50-7.0)	7.80(5.50-	6.8(4.75-	5.5(4.25-
hysterophorus	$\pm 0.67$	±2.84	±2.81	0.90	4	3	31	±0.32	±0.35	±0.62	9.2) ±0.6	9.25) ±0.75	7.0) ±0.52
Silybum	4.2(3.7-4.7)	26.8(25.2-28.7)	24.5(22.2-27)	1.00	2	0	27	1.4(0.7-2)	0.8(0.7-1)	6.50(5.75-7.25)	5.4(4.75-		
marianum	±0.16	±0.68	±0.86	1.09	3	0	27	±0.21	±0.05	±0.25	6.2) ±0.25	-	-
Sonchus	3.30(2.75-	38.1(32.2-52.7)	35.5(30.7-38.2)	1.07	3	1	19	5.4(4.7-5.7)	4(2.7-4.7)	9.8(9.2-10.2)	7.4(6.7-8.2)	7.8(6.5-9.2)	7.8(6.5-9.2)
arvensis	4.25) ±0.25	±3.7	±1.3					±0.18	±0.4	±0.2	±0.26	±0.4	±0.4
Sonchus asper	3.95(3.25-	39.6(38.2-40.7)	45.3(44-46.7)	0.87	3	0	0	-	-	8.4(7.7-9) ±0.21	9.1(8.2-9.7)	-	-
1	4.50) ±0.21	±0.4	±0.4								±0.24		
Sonchus	4.40(3.50-	34.4(24.0-47.2)	38.1(28.7-46.5)	0.90	3	3	27	3.6(3.0-4.2)	2.1(1.2-3.0)	7.4(3.5-12.0)	8.1(3.7-	4.7(2.7-8.5)	5.5(4.0-7.2)
oleraceous	5.75) ±0.73	±3.8	±3.0					±0.23	±0.30	±1.74	14.5) ±1.90	±0.99	±0.53
L	,										/		

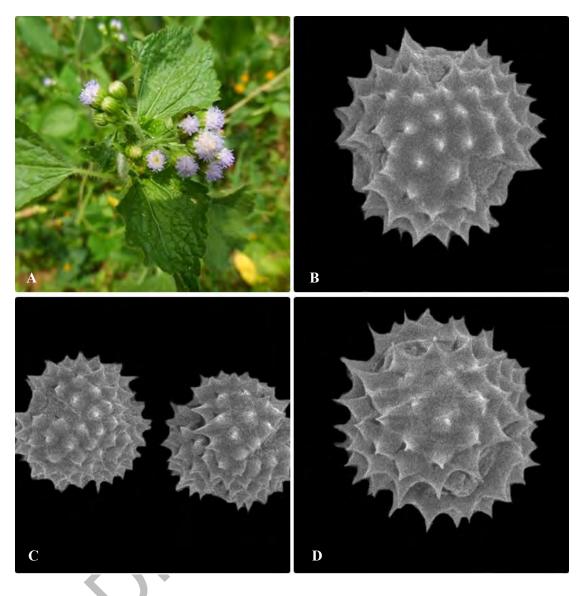
Sphagneticola	4.1(3.5-5.0)	39.6(33.7-45.7)	40.6(34.7-49.0)	0.97	2	2	32	2.9(2.5-3.2)	1.7(1.2-2.2)	7.95(5.75-10.2)	7.0(4.0-	7.50(6.25-	8.0(5.75-
trilobata	$\pm 0.25$	$\pm 2.24$	±2.45	0.97	3	3	32	$\pm 0.12$	$\pm 0.17$	$\pm 0.84$	$9.50) \pm 1.50$	$9.75) \pm 0.73$	$11.0) \pm 0.91$
Taraxacum	4.9(3.0-	33.6(29.0-39.0)	36.4(29.7-41.5)	0.92	3	3	29	2.3(1.2-3.2)	1.6(1.0-2.5)	6.8(4.2-9.5)	12.6(8.0-	7.0(3.7-9.5)	6.2(4.0-8.7)
offinale	$6.25) \pm 5.5$	$\pm 1.85$	±2.1					$\pm 0.3$	±0.25	$\pm 0.94$	$16.7) \pm 1.4$	$\pm 0.98$	$\pm 0.86$
Taraxacum	3.35(3.0-	34.4(30.5-36.7)	32.9(28.7-36.2)	1.04	3	3	31	3.1(2.7-3.5)	2.3(2.5-2.5)	6.3(5.2-7.2)	9.5(9.0-	7.9(7.0-8.7)	7.7(7.2-8.0)
platycarpum	$3.75)\pm0.28$	$\pm 2.5$	$\pm 3.08$					±0.28	$\pm 0.11$	$\pm 0.7$	$10.0)\pm\!0.39$	$\pm 0.69$	$\pm 0.32$
Verbisinia	4.2(3.5-5.2)	36.7(31.0-46.5)	36.2(30.5-41.5)	1.01	2	2	28	2.0(1.5-2.7)	1.9(1.2-2.5)	6.90(3.5-10.5)	6.2(3.5-	3.50(2.75-	4.20(3.25-
eleusoides	$\pm 0.32$	$\pm 2.71$	$\pm 1.78$	1.01	3	3	20	±0.21	±0.23	±1.33	$9.25) \pm 1.0$	$4.0) \pm 0.26$	$5.25) \pm 0.37$
Zinnia elegans	3.5(2.2-4.2)	32.2(28.5-34.5)	31.8(26.5-36.8)	1.01	2	2	20	6.3(5.2-7.2)	2.7(2-3.5)	7.45(6.25-8.25)	4.6(3-6)	3.9(2.75-	2.6(2-3.25)
	±0.33	$\pm 1$	$\pm 1.84$	1.01	3	3	20	±0.35	$\pm 0.28$	±0.33	±0.53	$5.25) \pm 0.41$	±0.23
*		**D/E_ D-1 1	iomoton/Equatorial	1:									

\* $\mu$ m= Micrometer, \*\*P/E= Polar diameter/Equatorial diameter

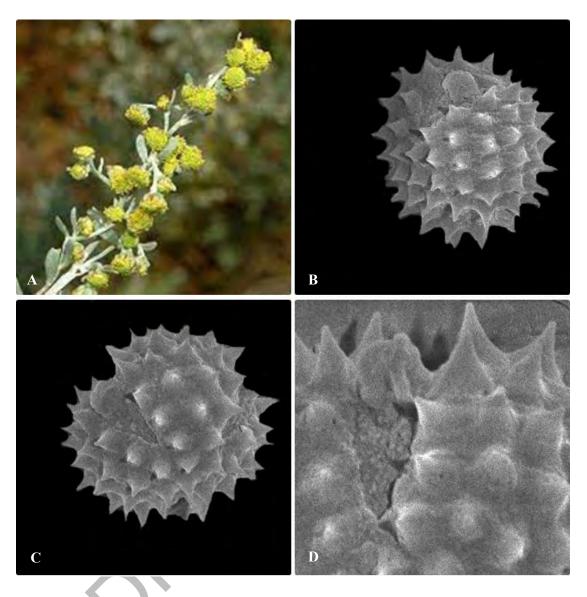
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Sr. No	Taxa	No of Fertile Pollen	No of Sterile Pollen	Fertility %	Sterility %
1.	Ageratum conyzoides	149	28	84.18	15.82
2.	Artemisia absinthimum	117	18	86.66	13.34
3.	Aster squamatus	158	43	78.60	21.4
4.	Bidens bipinnata	183	27	87.14	12.86
5.	Bidens pilosa	92	11	89.32	10.68
6.	Calendula arvensis	47	13	78.33	21.67
7.	Calendula officinalis	55	8	87.30	12.7
8.	Carthamus lanatus	127	19	86.98	13.02
9.	Carthamus oxyacantha	150	14	91.46	8.54
10.	Cirsium arvense	120	17	87.59	12.41
11.	Cousinia prolifera	164	36	<b>8</b> 2	18
12.	Eclipta prostrata	215	35	86	14
13.	Erigeron bonariensis	64	10	86.48	13.52
14.	Erigeron canadensis	203	31	86.75	13.25
15.	Ixeris polycephala	106	27	79.69	20.31
16.	Lactuca sativa	179	33	84.43	15.57
17.	Lactuca serriola	69	11	86.25	13.75
18.	Launea naudicaulis	132	26	83.54	16.46
19.	Launea procumbens	83	15	84.69	15.31
20.	Parthenium hysterophorus	98	11	89.90	10.1
21.	Silybum marianum	215	19	91.88	8.12
22.	Sonchus arvensis	76	16	82.60	17.4
23.	Sonchus asper	120	41	74.53	25.47
24.	Sonchus oleraceous	123	22	84.82	15.18
25.	Sphagneticola trilobata	138	48	74.19	25.81
26.	Taraxacum offinale	87	7	92.55	7.45
27.	Taraxacum platycarpum	156	17	90.17	9.83
28.	Verbesina helianthoides	151	9	94.37	5.63
29.	Zinnia elegans	107	14	89.16	10.84

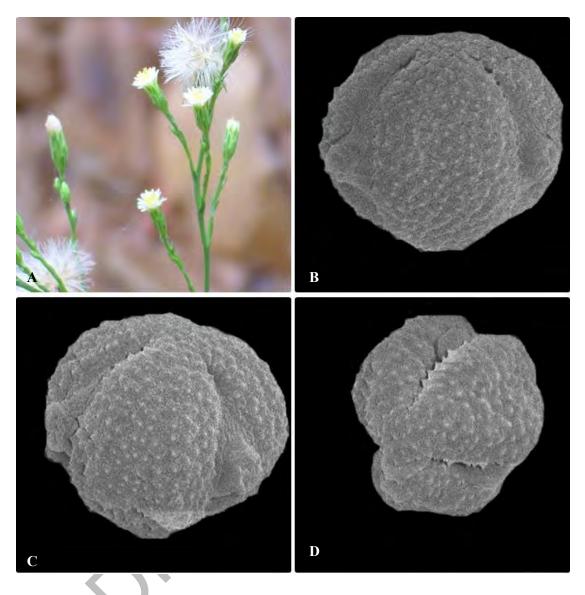
### Table 4: Fertility and Sterility percentages of examined taxa



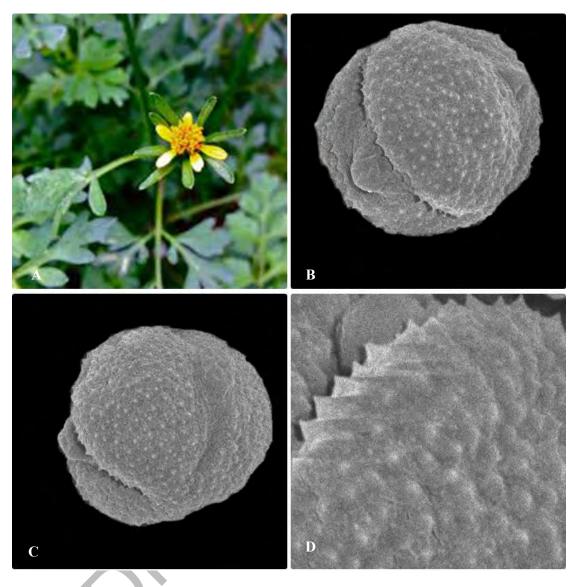
**Plate 23:** Ageratum conyzoides (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Echinate Exine Sculpturing



**Plate 24:** *Artemisia absinthimum* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Surface Sculpturing



**Plate 25:** *Aster squamatus* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Orientation (D) Echinate Exine Sculpturing



**Plate 26:** *Bidens bipinnata* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Perforate Ornamentation (D) Echinate Exine Sculpturing

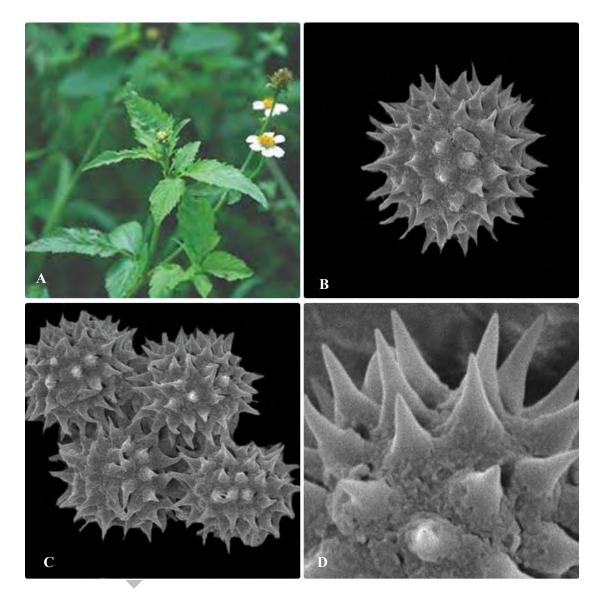
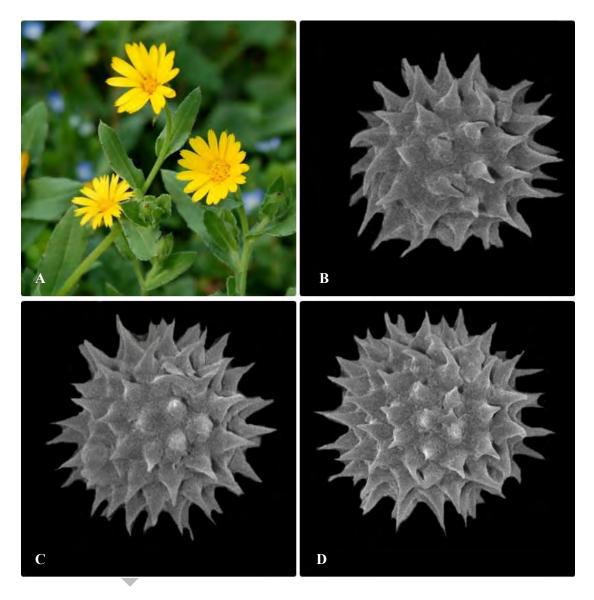
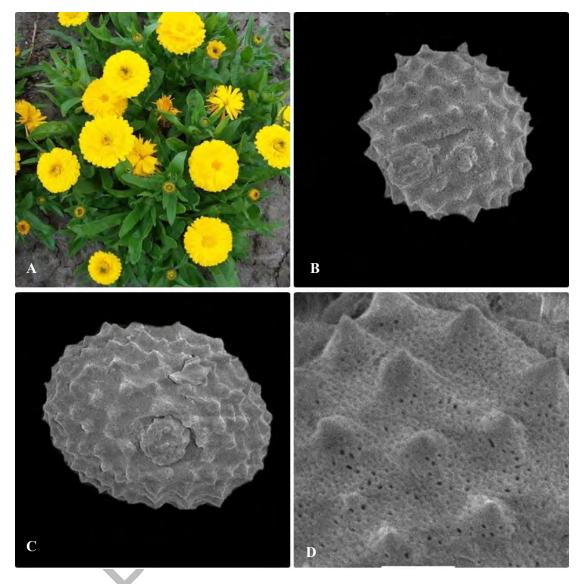


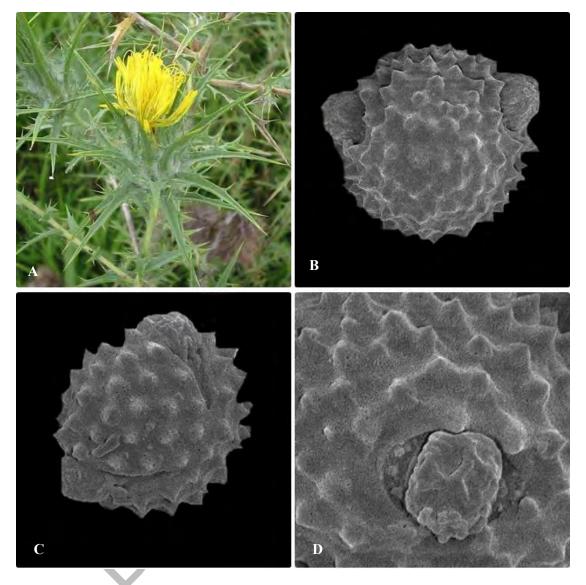
Plate 27: *Bidens pilosa* (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Elliptical sunken orientation (D) Echinate Exine Sculpturing



**Plate 28:** *Calendula arvensis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken orientation (D) Exine Sculpturing



**Plate 29:** *Calendula officinalis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Surface Sculpturing



**Plate 30:** *Carthamus oxyacantha* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Surface Sculpturing

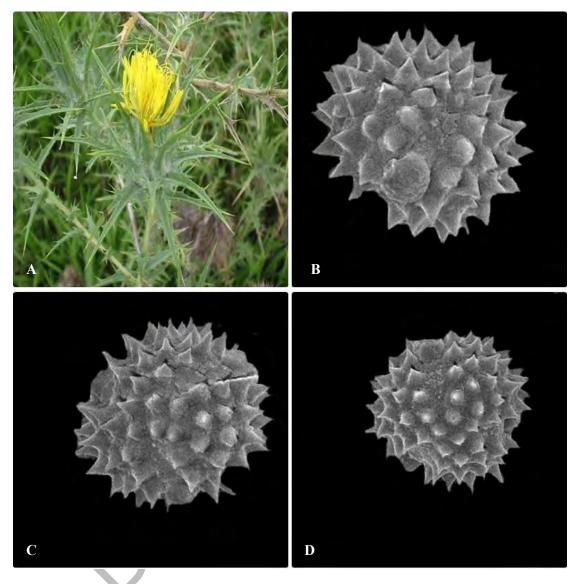


Plate 31: Carthamus lanatus (A) Floral Parts, Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Echinate Exine Sculpturing

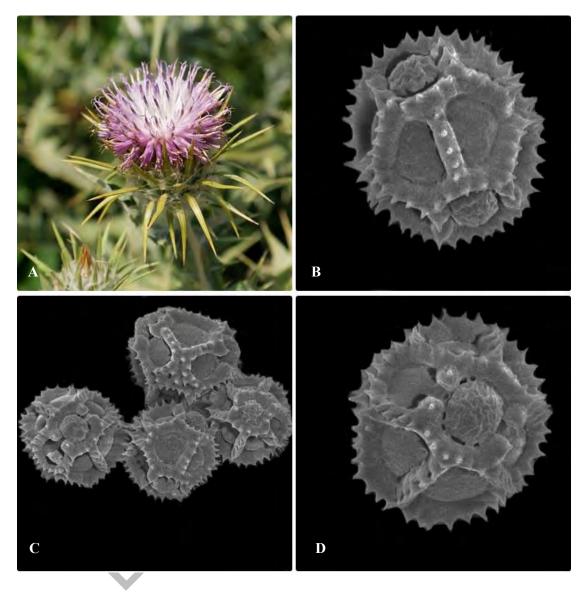


Plate 32: Causinia prolifera (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Echinate Exine Sculpturing

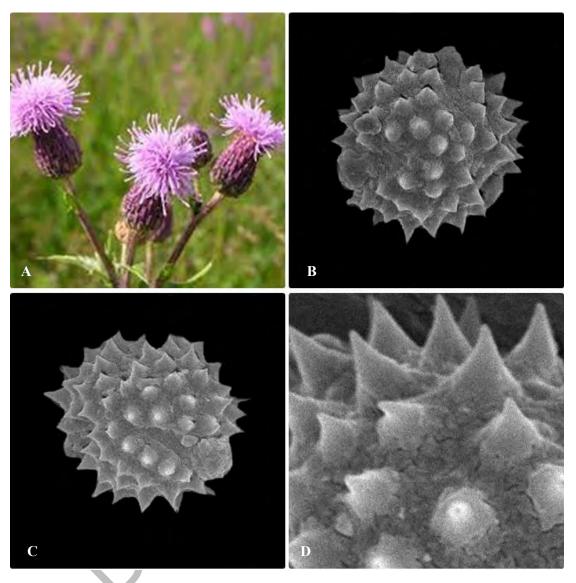
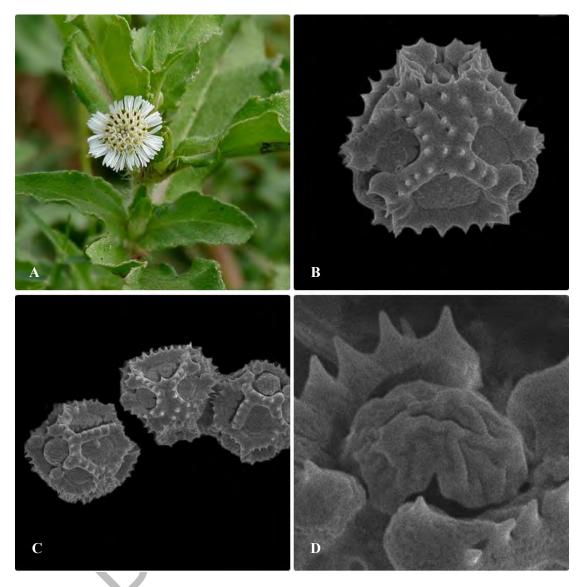
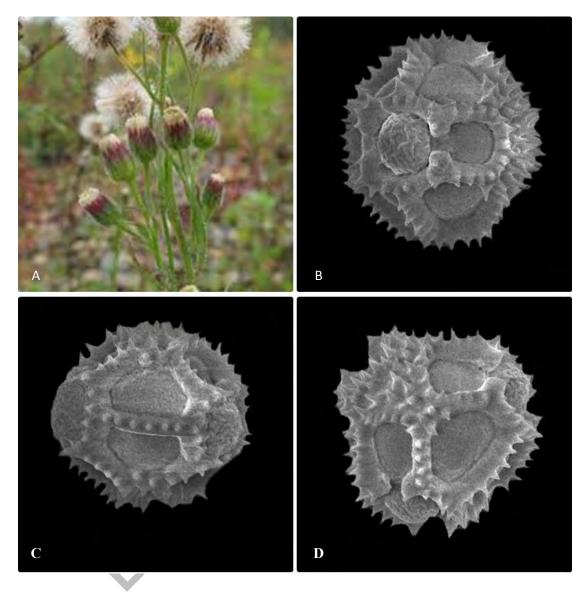


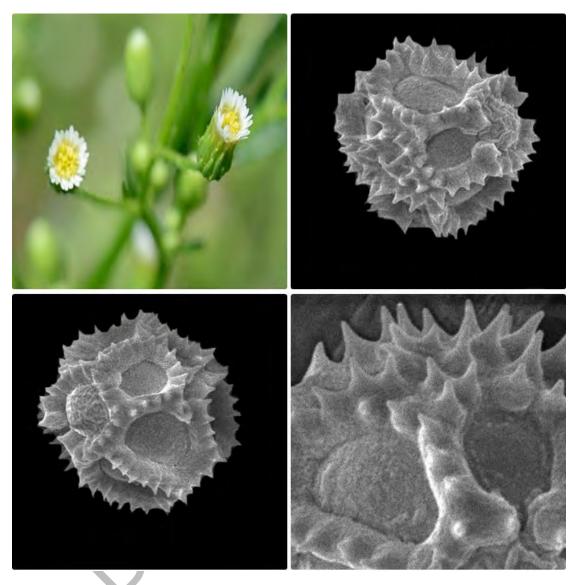
Plate 33: *Cirsium arvense* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Surface Sculpturing



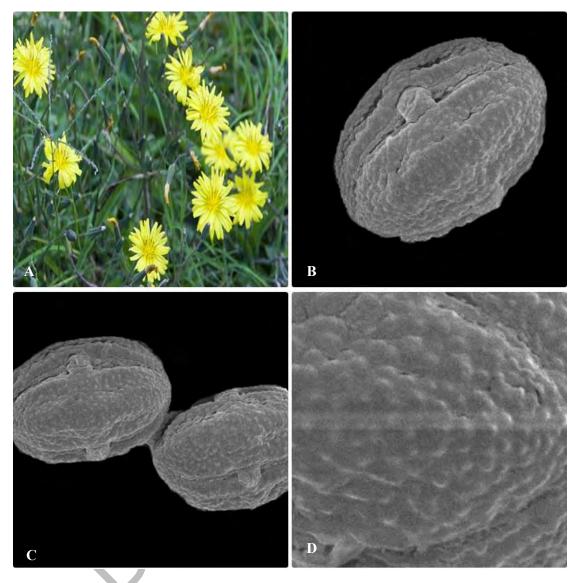
**Plate 34:** *Eclipta prostrate* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Echino-Lophate Ornamentation (D) Surface Sculpturing



**Plate 35:** *Erigeron bonariensis* (A)Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded orientation (D) Echinate Exine Sculpturing



**Plate 36:** *Erigeron Canadensis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Surface Sculpturing



**Plate 37:***Ixeris polycephala* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Echinate Pollen Ornamentation (D) Surface Sculpturing

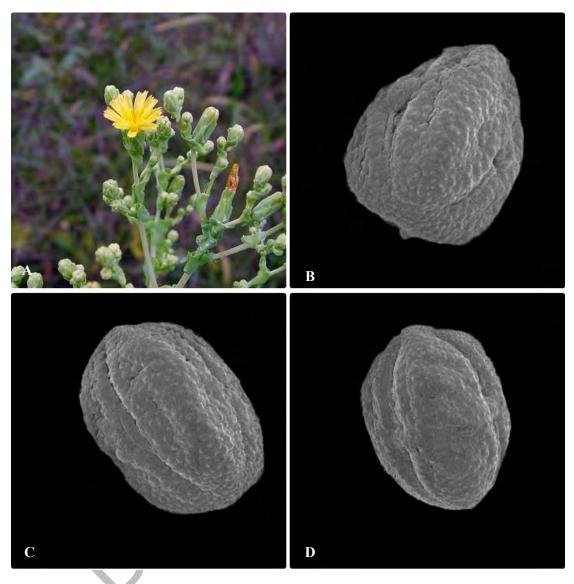


Plate 38:*Lactuca sativa* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Echinate Sculpturing

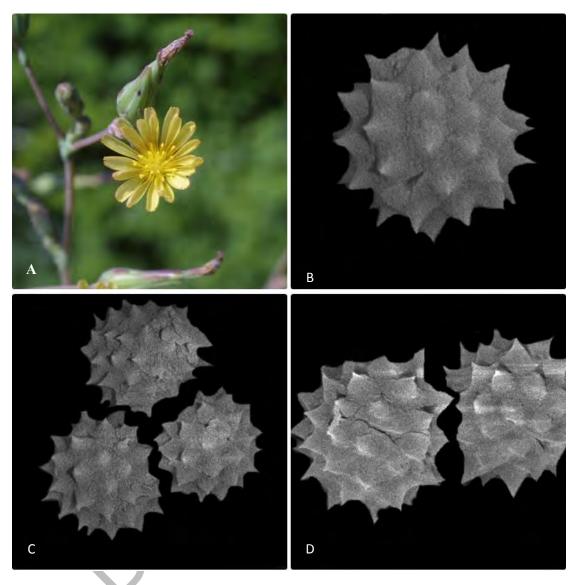
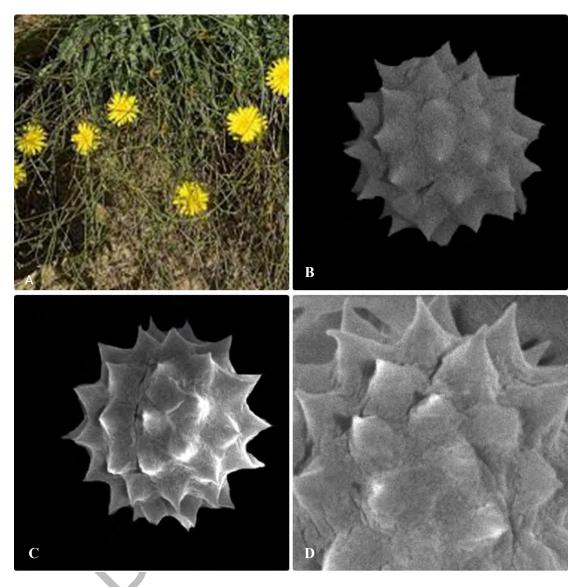
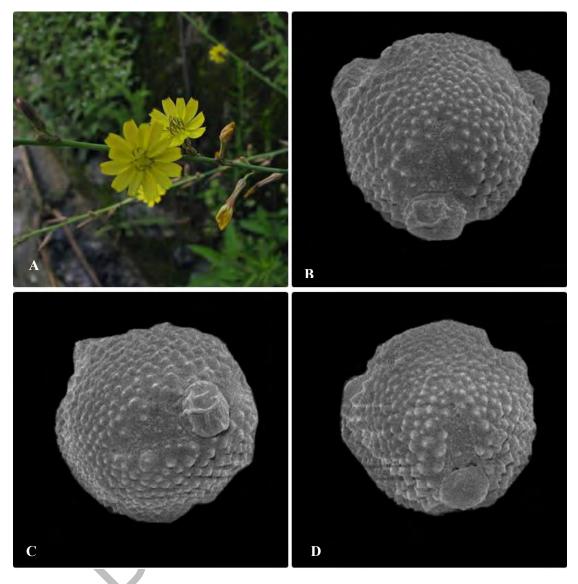


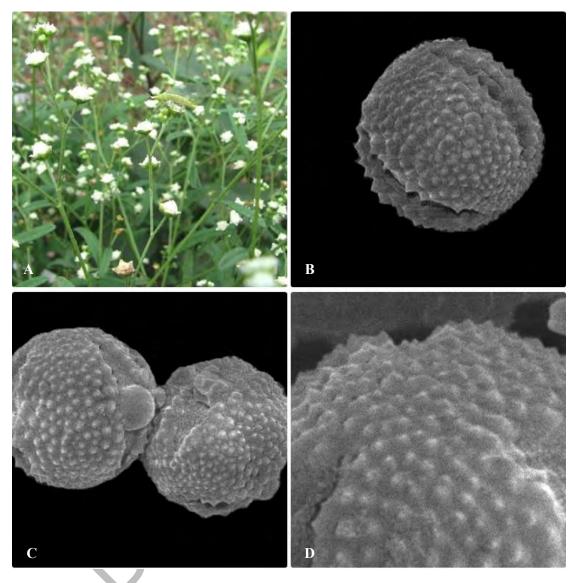
Plate 39:Lactuca serriola (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Elliptical sunken orientation (D) Echinate Exine Sculpturing



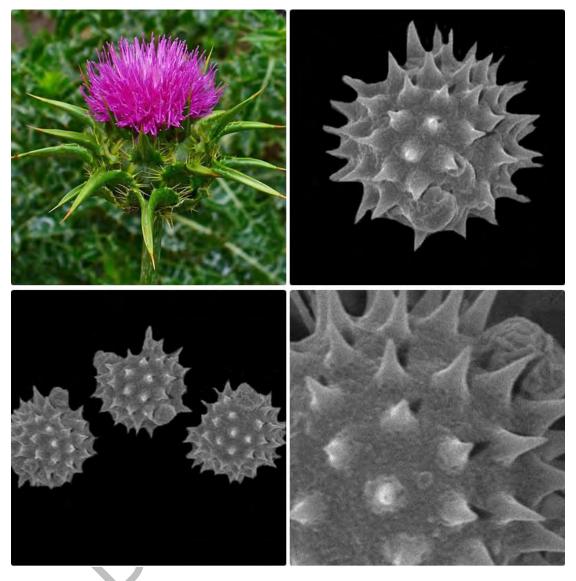
**Plate 40:***Launea naudicaulis* (A)Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Surface Sculpturing



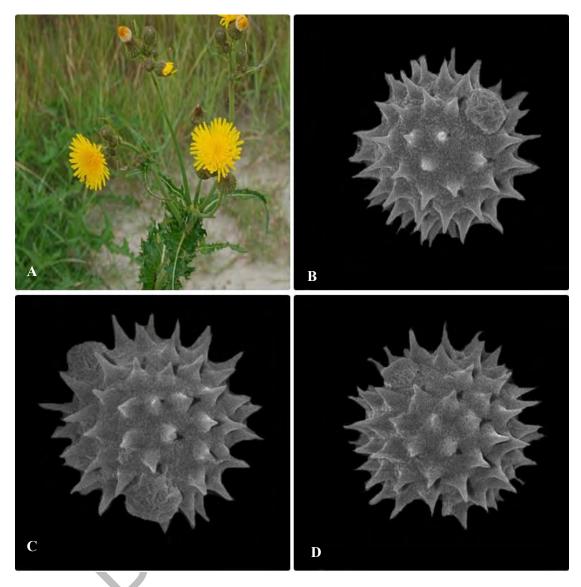
**Plate 41:** *Launea procumbens* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Micro-reticulate Echinate Exine Sculpturing



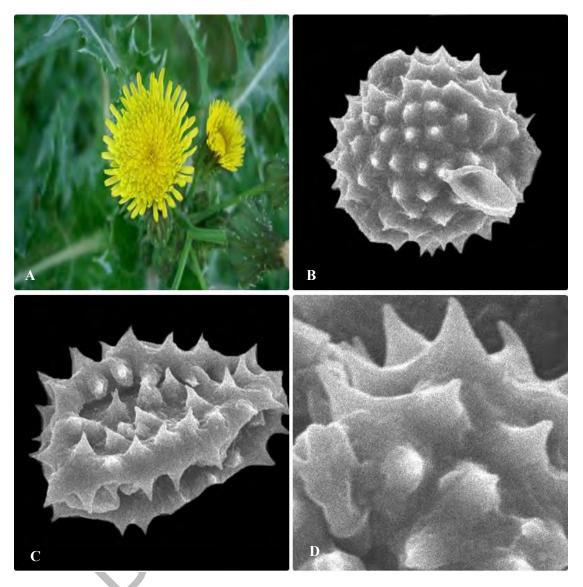
**Plate 42:***Parthenium hysterophorus* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Surface Sculpturing



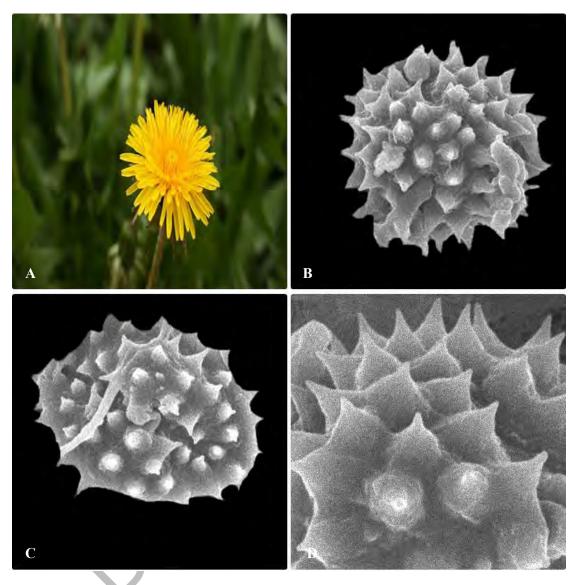
**Plate 43:** *Silybum marianum* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Echinate Pollen Orientation (D) Surface Sculpturing



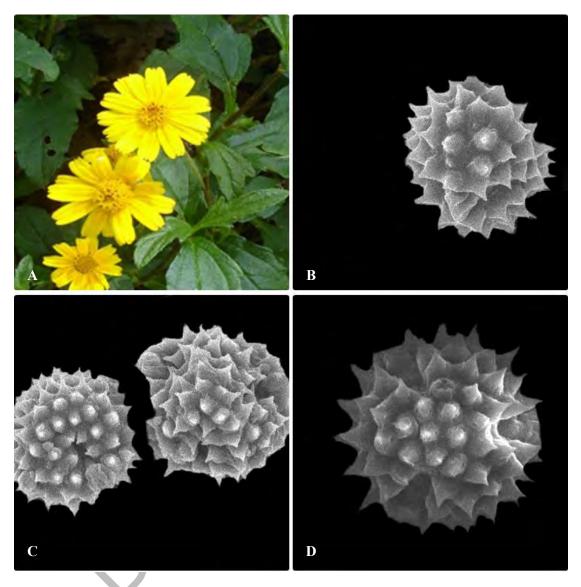
**Plate 44:** *Sonchus arvensis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Echinate perforate- tectate Exine Sculpturing



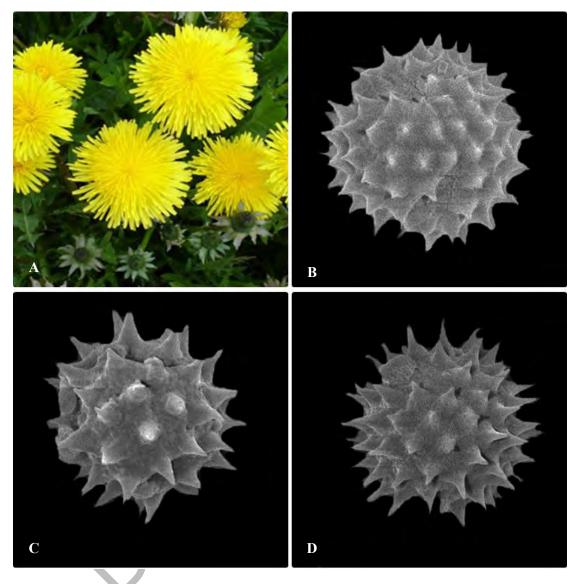
**Plate 45:***Sonchus asper* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical Sunken Orientation (D) Echinate Exine Sculpturing



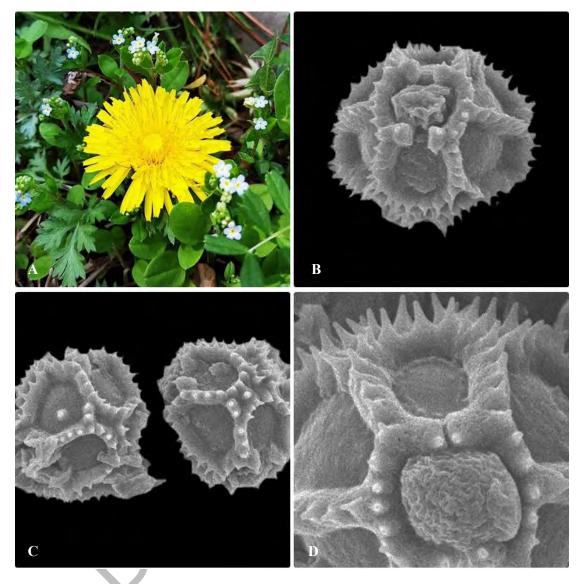
**Plate 46:***Sonchus oleraceous* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Surface Sculpturing



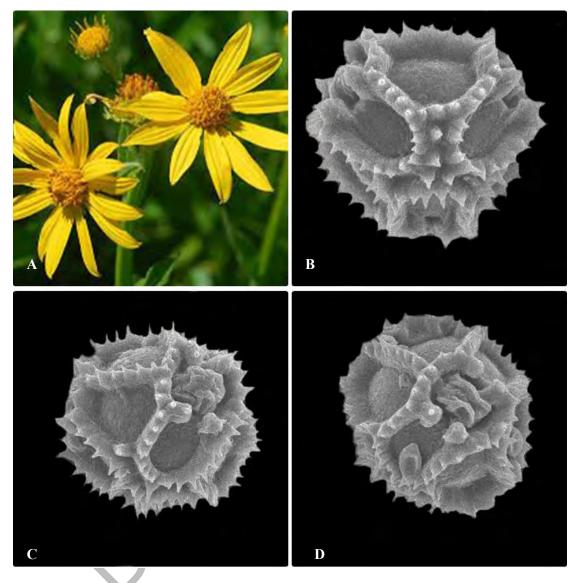
**Plate 47:***Sphagneticola trilobata* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Elliptical sunken orientation (D) Echinate Exine Sculpturing



**Plate 48:***Taraxacum offinale* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Echino-lophate perforate Exine Sculpturing



**Plate 49:***Taraxacum platycarpum* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Echinate Surface Sculpturing



**Plate 50:***Verbisinia eleusoides* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded sunken orientation (D) Echino-lophate Exine Sculpturing

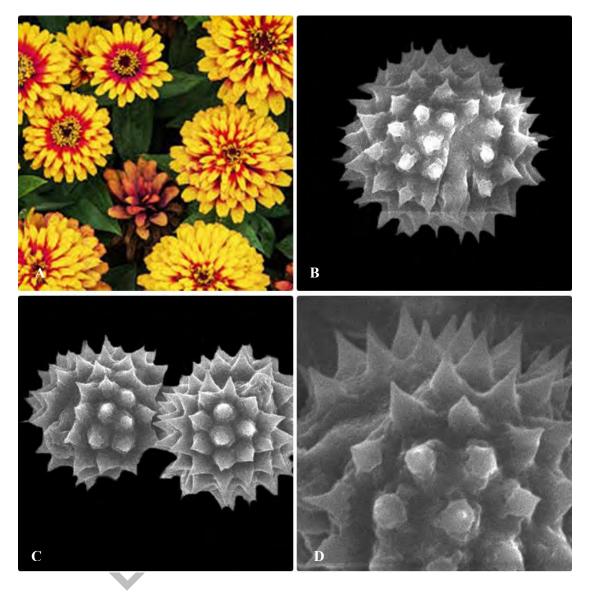


Plate 51: Zinnia elegans (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Echinate with pointed spines ornamentation (D) SurfaceSculpturing

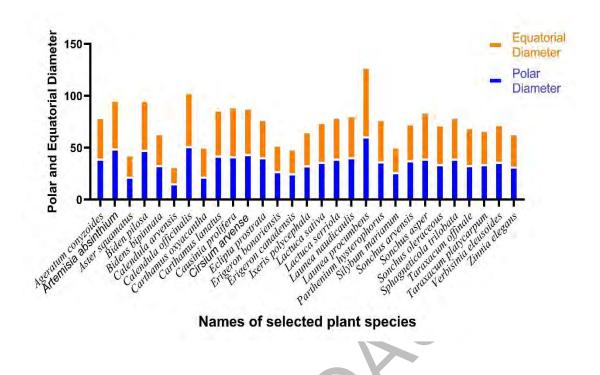


Figure 6: Mean values of Pollen diameter among Asterecious taxa

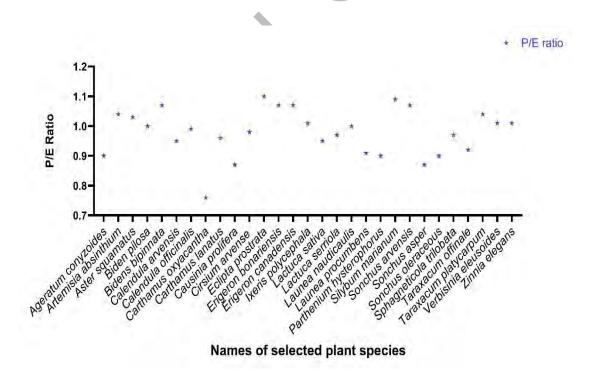


Figure 7: Variation in P/E ratio index of Asterecious taxa

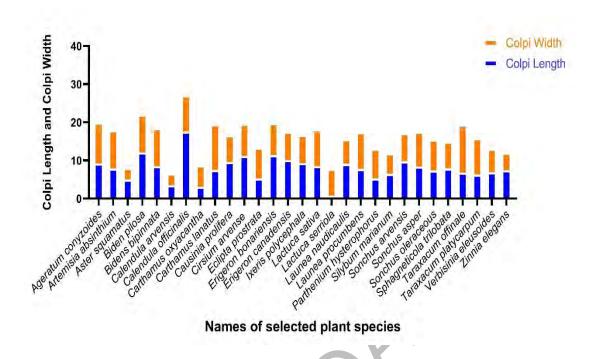


Figure 8: Mean data variations of colpi length and width in Asteracious taxa

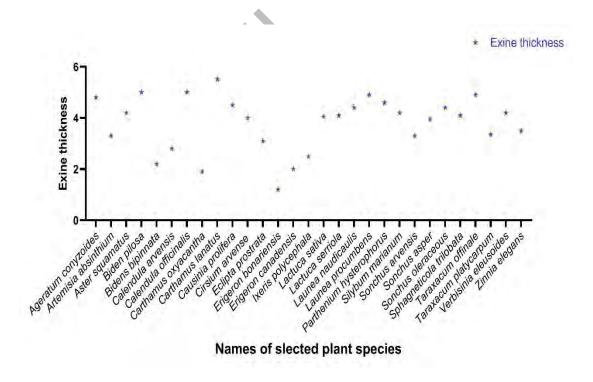


Figure 9: Mean exine thickness in Asteraceous Species.

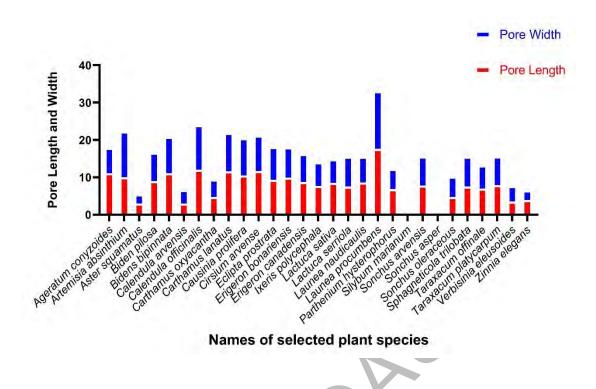


Figure 10: Data variations of pore length and width in Asteraceous species

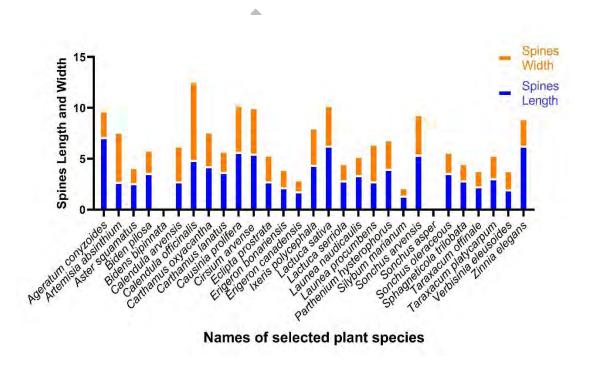


Figure 11: Variations in mean values of Spines Length & Width among Asteraceous taxa

# **Taxonomic Key based on Asteraceaous Pollen** 1b. Lacunae present ...... 12 2a. Pollen type tetra colporate ...... Launea serriola 3a. Pores absent ...... Silvbum marianum 5a. Polar view is semi angular ...... Calendula officinalis 6a. Micro-reticulate pollen ornamentation ...... Launea procumbens 7a. Spine length is above 7µm ...... Ageratum conyzoides 7b. Spine length is below 7 µm ......8 8a. Polar view is irregular ..... *Taraxacum officinale* 9a. Spine length is below 2 µm ..... Erigeron canadensis 9b. Spine length is above 2 µm..... 10 10a. Colpi width is above 10 µm ......Artemesia annua 10 b. Colpi width is below 10 μm...... 11 11 a. Pore length is below 7 µm ..... Calendula arvensis 11b. Pore length is above 7 μm ..... *Taraxacum polycarpum*

12a. Pollen ornamentation is Psilate Sonchus asper
12b. Pollen ornamentation is echinate or rugulate
13a. Pollen type is tri-tetrazonocolporate Lactuca sativa
13b. Pollen type is tri and tetrazonocoplorate 14
14a. Polar view is hexagonal roundedVerbesinia eleusoides
14b. Polar view is circular or triangular 15
15a. Semi angular pollen Launea naudicaultis
15b. Triangular or circular pollen 16
16a. Tetrazonocolporate pollen Erigeron canadensis
16b. Tri or tetra colporate pollens17
17a. Pollen shape is sub-oblate Carthamus oxyacantha
17b. Pollen shape is spheroidal
18a. Pollen ornamentation is tectate
18b. Pollen ornamentation is echinate 19
19a. Spines absent Bidens bipinnata
19b. Spines present 20
20a. Pore width is below 3 μm Zinnia elegans
20b. Pore width is above3 μm 21
21a. Polar diameter is above 45 µm Bidens pilosa
21b. Polar diameter is below 45 μm 22
22a. Rounded triangular pollen Parthenium hysterophores
22b. Polar view is circular or triangular 23
23a. Echini arrangement is rather regular Carthamus lanatus

23b. Regularly arranged Echini 24
24a. Pollen shape is prolate spheroidal <i>Eclipta prostrata</i>
24b. Pollen shape is oblate spheroidal 25
25a. Pollen type is tetra colporate Sonchus oleraceous
25b. Pollen type is tri or tetrazonocolporate
26a. Lacunar shape is tetragonal Cirsium arvensis
26b. Lacunar shape is pentagonal 27
27a. Pollen ornamentation is Echinate perforate Sphagneticola trilobata
27b. Pollen ornamentation is echinate
28a. Polar view circular Ixeris polycephala
28b. Polar view is circular Aster squamatus

### **3.1.1Asteraceaous Species**

In Current Study 29 species of family Asteraceae were investigated using light and scanning electron microscopy. These micro morphological characters of pollens plays vital role in classification of taxa of Asteraceae family. Two types of pollen class have been observed in present study that is Dizonocolporate and Trizonocolporate. Two types of apertures have been seen among the 19 Asteraceae species that is Lacunae and non-lacunae. Different shapes of the pollens have been observed base on the measurement of P.E ratios (Mbagwu et al., 2009). Pollen character is considered as additional information during systematic studies by (Garnatje and Martin, 2007; Meo and Khan, 2004b). In his work he observed that all the species of genus *Echinops* contains same characters except one species *E. strigosus* on the basis of dissimilarity in pollen characters it is placed on other genus of the family Asteraceae.

(Meo and Khan, 2003) studied the pollen morphology of 2 species of Achellia genus i-e A. millefolium and A. santolina. Variations were observed in counting the number of spines between the coli A. mellifolium contains 5-6 rows of spines and A. santilina contains 4-5 rows of spines between each colpi. Compare the result of this study with (Dauti et al., 2017) A. mellifolium contains 4-5 rows of spines present between the colpi.

# 1 P/E Ratio

In the present work the smaller P/E ratio was found in *Carthamus oxyacantha* (0.7  $\mu$ m) while the greater value was found in *Eclipta prostrata* (1.1  $\mu$ m). Their polar and equatorial ratio, that is, P/E ratio varied between different taxa. Mbagwu et al., (2009) documented that palynological application is miscellaneous and multidisciplinary. Conversely, the role of pollen morphology is of significance in taxonomic argue for classification. The pollen characters are used for the organization of interspecies relationships among species of Asteraceae. From the present study it was concluded that the differences and similarities in pollen morphology are significant and could be subject for biosystematics purpose. Our results are in accordance with the findings of Wodehouse (1935) who recorded echinate and spheroidal pollens in *Berkheya heterophylla* (Arctotidae— Compositae). A similar result was found in the work of Mbagwu et al., (2007), who proposed that the pollens of *G. nigritiana* were spheroidal in shape and their general appearance was circular. According to the present study

echinate ornamentation was found approximately in all members of family Asteraceae.

The large sized pollen grains were found in family Asteraceae members such as G. regins, Tagetus erectus and C. officinale and small pollen in H. annuus which is in line with the findings of Osman (2011) who studied small pollen in Lasiopogon muscoides (19-23 µm) and large in T. minuta (42-45 µm). The pollen grains were less or more similar in shape that is, spheroidal, oblate-spheroidal and prolatespheroidal. Mumtaz et al., (2000) reported a P/E ratio of 1.14 to1.31 in Artemisia species similar to the P/E ratio of C. sulphureus (1.2  $\mu$ m) which is similar with the present study. Spines were minute and rudimentary in two species of Artemisia and Tanacetum gracile, while other species have well developed spines which are not in accordance with the present work because all pollen in the family Asteraceae in the present study possessed spines on pollen surface. Caramiello and Fossa (1994) noted the exine thickness in the range of  $3.1-4.6 \ \mu m$  in four species of Artemisia which agreed with the exine thickness of the present findings. This parameter is useful at the species level, as almost all the species had different exine thickness. Spines are also considered as distinguishing characters in some tribes of family Asteraceae. Usually shorts spines in terms of size are found in A. conyzoides and V. cinerea while long spines are observed in B. pilosa. Walls of the pollen grains of mentioned species are thin same result was also reported by the study conducted by Perveen (1999) for the family Asteraceae.

(Wodehouse, 1935) works on the evolution of spines in family Asteraceae and proposed a decline in spines from long to short. Pollens with spines were considered to the primitive character in comparison with spineless pollens. Tribe Astereae was studied by (Clark et al., 1980) separates some genus on the basis of size of pollens, spine length and the number of spine among the colpi. Spine length and the number of spines between the colpi was also studied by (Abid and Qaiser, 2004; Tomb et al., 1974) and considered them as distinguishing characters for genus Inula of family Asteraceae. Exine morphology of sub tribe Ecliptinae was studied by (Coutinho et al., 2016). Pollen fertility of some genus was studied by (Qureshi et al., 2002) and it revealed that fertility of pollens plays significant role in cytology studies of chromosomes. Much research work is done on family Asteraceae by (Akyalcin et al., 2011; Dematteis and Pire, 2008; Jafari and Ghanbarian, 2007; Skvarla et al., 1977; Urtubey and Tellería, 1998) and studied different qualitative and qualitative characters of pollens. (Farco and Dematteis, 2017) studied 10 species of genus Campuloclinium in terms of pollen morphology and also worked on diverse mitotic abnormalities. (Tellería, 2017) studied the exine ornamentation of pollen grains of family Asteraceae. (Zafar et al., 2007) studied 7 species of pollens and observed characters that include pollen size shape, exine ornamentation, aperture types, spine length and number of spines between the colpi. The present study could be taxonomically significant for the correct identification and species delimitations of family Asteraceae. Colpi number in pollen plays important role in tracing evolutionary relationship among the reported species of present study. In advanced dicotyledonous plants more number of colpi are present as compared to the primitive ones. Primitive species either contains single colpi (monocolpate) or they are without colpi (acolpate) (Adedeji, 2005).

Apart from generic characters there are some quantitative characters which also help in identification of species. It has been observed in most of the species of this family that the diameter of species is small. Erdtman (1952) categorized pollen according to size into different groups namely perminuta (diameter less than 10 $\mu$ m), minuta (diameter 10-25 $\mu$ m), media (diameter 25-50 $\mu$ m), magna (diameter 50-100 $\mu$ m), permagna (diameter 100-200 $\mu$ m), giganta (diameter greater than 200 $\mu$ m). Keeping in view this classification some species of family Asteraceae belong to the groups' minuta & media. *A. conyzoides* is in the group minuta while the other species belong to the group media which means that the pollen diameter measured is classificatory for others and delimiting for *A. conyzoides*.

# 2 Pollen Shape

In a study conducted by Khan (2006) documented the presence of tetrazonocolporate pollen in *Calendula officinalis* and also studied the size of spines in comparison with other species zcnm\``of this tribe. The result of present study differs from previousfindings as tri- tetracolporate pollen were found with spine ornamentation echinate having broad base. Akinnubi et al., (2014) reported polypantoporate and circular pollen in genus *Bidens*. Zhang et al., (2004) reported trizonocolporate pollen in *Bidens bipinnata*.

Bülbület al., (2013) studied genus *Carthamus* from flora of turkey and stated tricolporate iso-polar echinate, spheroidal or oblate spheroidal pollen. Jafari et al., (2007) observed diverse outlines of pollen surface in family Asteraceae and detected verrucate pattern of exine in *Cousinia*, sub-echinate or verrucate pattern in *Centaurea*, echinate pollen in *Carthamus* and echinate surface with short colpi in *Cirsium*.

Wróblewska et al., (2016) studied pollens of ornamental taxa of family with respect to bees as a source of pollen and reported tricolporate, echinate and oblate-spheroidal pollen in genus *Zinnia*. Bolick (1978) study exine pattern in different tribe of Compositae Heliantheae, Calenduleae, Sececioneae, Eupatorieae and Calenduleae etc with respect to effect of mechanical stresses on exine and its evolution in response to the factor. Bemer (1987) examined cladistics of 27 different tribe of family Asteraceae relating 81 different character and record that Heliantheae is monophyletic tribe and the hypothesis that other tribe of Asteraceae is evolved from this proven wrong.

Although the study of morphological characters of pollen observed through microscope i.e pattern of surface, ornamentation, spines, colpi, pores, size and shape of tribe Cichorieae has revealed prominent diagnostic characters related to the flora, vegetative and chromosomal characters of the tribe, these characters show some variations with the taxonomic classification of the tribe into sub families and sub tribes. On the other hand in some genera surface pattern is unique and some tribes and sub tribes also show similarity in the surface pattern of pollen grains, although other features such as size range and shapes of pollen grains do not differentiate them from other species of the tribe. So it is clear that only morphological characters cannot be used as the base of taxonomic classification of the family or tribe. Though, it is known that these morphological features of pollen demonstrations evolutionary sequences similar to those in other structures, and then it may need to be given as much weight as any other morphological character. On this assumption, in the pollen morphology of species examined, some in consistencies and alternative relationship have been suggested in correlation with taxonomic classification proposed by (Johnson and Briggs 1975). The general features of tribe Cichorieae, taken together are not repeated in other tribes. This gives tribe Cichorieae a unique taxonomic status. Combined efforts of evolutionary ecological and systematic studies are still important to aid in understanding the advancement of this interesting tribe.

Pollen morphological studies have great importance in taxonomic study of plants and the improvement in microscopic technique has led to the utilization of novel morphological features of pollen for taxonomic purposes. The characteristics of pollen not only provide the information but they are also useful in improving the systematic position of their members with respect to their families, sub-families or tribe. Wodehouse (1935) concluded that not only the overall morphological characters are significant in delimitation of species. Two major groups are formed in Asteraceae based on their pollen characters i.e., non-lophate and lophate pollen. The former ones characterize in *Sonchus* and *Lactuca*. Surface ornamentation of exine is echinate in all the members of Lactuceae, also some members are trizonocolporate while some members are also tetrazonocolporate (Qureshi et al., 2002).

### **3** Spines Variation

(Wodehouse, 1935) works on the evolution of spines in family Asteraceae and proposed a decline in spines from long to short. Pollen with spines was considered to the primitive character in comparison with spineless pollen. Tribe Astereae was studied by (Clark et al., 1980) separates some genus on the basis of size of pollens, spine length and the number of spine among the colpi. Spine length and the number of spines between the colpi was also studied by (Abid and Qaiser, 2004; Tomb et al., 1974). In current study all the taxa had echinolophate or echinate ornamentation consist both spine and lophae except for *Sonchus asper* it has psilate type of sculpturing. Lophae are the partitions of pollen surface into small cubicle or circular portion separated by walls known as Muri (Wodehouse, 1935).Some species show very clear recognizable lacuna i.e. genus *Lactuca* while *Taraxacum* and *Launeae* showed less visible lacuna which revealed that the type of lacunae was slightly different from that of *Lactuca*.

#### **4 Exine Sculpturing**

Echinolophate sculpturing in *Lactuca* is considered recent as compare to echinate sculpture in other tribe of Asteraceae or we can say echinate ornamentation is primitive in Asteraceae (Jafari & Ghanbarian, 2007). In genus *Lactuca* entire surface of pollen was covered by spine except the depression known as lacunae, exine seems smooth in these regions but the wall of lacunae were echinate.

Al-Tameme (2014) observed echinolophate pollen in genus *Koelpinia*. Jeffrey et al., (2007) account echinolophate colporate pollen in genus *Lactuca*. Diez et al., (1999) reported echinolophate and trizonocolporate pollen in *Sonchus* and Launaea. Perveen (1999) investigate echinolophate trizonocolporate and oblate pollen in genus *Sonchus*. Blackmore (1981) reported echinolophate ornamentation in his study.

The ornamentation of exine and characters of spines are very significant in the phylogenetic relationship of the family (Pınar and Dönmez, 2000). Variation in exine thickness among the species in this family was also observed previously (Ahmad et al., 2013). All the studied taxa have tricolporate pollen as the number of colpi on pollen grains has been a useful tool in tracing evolutionary relationship among the species. The advanced dicotyledons have more colpi than the primitive ones, with either a colpus (monocolpate) or none at all (acolpate) (Akinnubi et al., 2014 and Adedeji, 2005). The dendrogram showing the similarity index of the species on bases of the studied taxa of family Asteraceae. Highest similarity found between *T. platycarpum* and *I. polycephala* opposite to the one between *L. sativa* and *S. arvensis* which were distantly related. *S. oleraceous* and *T. officinale* show more similarity index as compared to *L. naudicaulis*, *L.sativa* and *S. arvensis*.

Sr.	Taxa	Pollen type	Shape	Pollen	Pore
No				ornamentation	ornamentation
1.	Achyranthus aspera	Apolar	Spheroidal	Scabrate	Sunken
2.	Aerva javanica	Apolar	Spheroidal	Microechinate	Sunken
3.	Amaranthus gracizens	Apolar	Spheroidal	Scabrate	Sunken
4.	Amaranthus hybridus	Apolar	Spheroidal	Microechinate	Sunken
5.	Amaranthus spinosus	Apolar	Spheroidal	Microechinate	Sunken
6.	Amaranthus viridis	Apolar	Spheroidal	Microechinate	Sunken
7.	Atriplex stoeksii	Apolar	Spheroidal	Scabrate	Sunken
8.	Celosia Argentia	Apolar	Spheroidal	Microechinate	Sunken
9.	Chenopodium album	Apolar	Spheroidal	Scabrate	Sunken
10.	Chenopodium ambrosioides	-	Spheroidal	Microechinate	Sunken
11.	Chenopodium murale	Apolar	Spheroidal	Microechinate	Sunken
12.	Digeria muricata	Apolar	Spheroidal	Scabrate	Sunken
13.	Kochia indica	Apolar	Spheroidal	Scabrate	Sunken
14.	Kochia lonifolia	Apolar	Spheroidal	Scabrate	Sunken
15.	Pupalia lappaceae	Apolar	Spheroidal	Microechinate	Sunken
16.	Salsola lubricate	Apolar	Spheroidal	Scabrate	Sunken
17.	Sauda fruitcosa	Apolar	Spheroidal	Microechinate	Sunken

**Table 5:** Qualitative data of pollen of family Amaranthaceae members from Potohar

 region of Pakistan

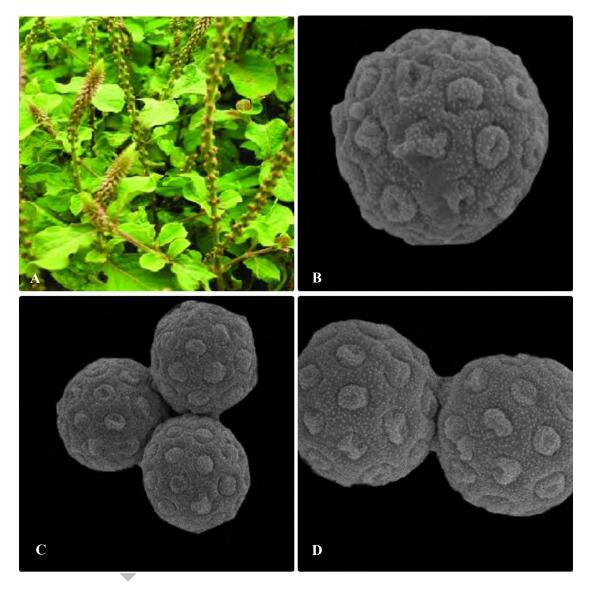
Sr.	Taxa	Polar diameter (µm) min	Equatorial diameter	P/E	Exine thickness (µm)	Pore length (µm) min	Pore width (µm) min
No		(mean ± Std error) max	(µm) min (mean ± Std	ratio	min (mean ± Std	(mean ± Std error)	(mean ± Std error)
			error) max	(µm)	error) max	max	max
1	Achyranthus aspera	$22.75~(23.95\pm0.52)~25.5$	$22.25\;(23.90\pm0.72)\;25.5$	1.00	1.0 (1.55±0.16) 2.0	$1.0\;(1.85\pm0.23)\;2.25$	$0.75~(1.2\pm0.14)~1.5$
2	Aerva javanica	$18.25\ (20.2\pm0.50)\ 21.0$	$19.0\ (20.1\pm 0.32)\ 20.75$	1.00	1.0 (1.2 ± 0.09) 1.5	$1.5 (1.70 \pm 0.09) 2.0$	$1.5 (1.75 \pm 0.11) 2.0$
3	Amaranthus gracizens	$12.75 (16.2 \pm 1.02) 18.25$	$12.85\ (18.87\pm1.9)\ 22.75$	0.85	1.0 (1.3 ± 0.09) 1.5	$0.02\;(0.04\pm 0.01)$	$0.25\;(0.55\pm0.09)\;0.75$
4	Amaranthus hybridus	23.25 (26.25-21.2)±2.09	27.25 (31.25-23.7)±2.85	0.85	4.55 (5.25-4)±0.48	3(2.5-3.5) ±0.16	3.6(3.25-4) ±0.12
5	Amaranthus spinosus	54.25(51.25-56.25)±0.88	49.6(38.75-60.50)±0.29	1.09	5.20(4.50-6.0)±0.28	12(10.2-13.7) ±0.68	12(11.25-13) ±0.29
6	Amaranthus viridis	27.8(26.5-29.7)±0.68	28.5(26.2-31)±0.78	0.97	4.11(2.75-6)±0.72	4.7(3.5-6.25) ±0.45	4.7(3.50-5.50) ±0.40
7	Atriplex stoeksii	$18.0\ (20.0\pm0.84)\ 22.75$	$17.75 (18.85 \pm 0.49) 20.2$	1.07	$1.0 (1.6 \pm 0.23) 2.25$	$0.75~(1.20\pm0.27)~2.25$	$0.5 \ (0.75 \pm 0.79) \ 1.0$
8	Celosia Argentia	$13.50\ (16.2\pm0.85)\ 18.0$	13.25 (15.65 ± 1.0) 18.50	1.03	$1.5 (2.15 \pm 0.3) 3.0$	$0.75 (1.05 \pm 0.09) 1.25$	0.5 (0.7±0.09) 1.0
9	Chenopodium album	27.1(25.5-29.2)±0.71	25.8(22.7-31.5)±1.49	1.05	4.40(2.75-6)±0.65	4.75 (3.75-6.00) ±0.93	3.00 (2.25-3.75) ±0.28
10	Chenopodium ambrosioides	7.75(11.00-5.25)±0.88	7.67(10.75-5.50)±0.78	1.01	.88(1.2550)0±.11	3.85 (2.50-4.50) ±0.38	2.75 (2.25-3.25) ±0.18
11	Chenopodium murale	12.75 (14.75-9.50)±2.06	10.75 (12.5-9.75)±1.04	1.18	2.00 (2.75-1.25)±0.5	4.20 (3.75-4.75) ±0.20	3.30 (2.25-4.25) ±0.34
		$\bigcirc$					

 Table 6: Quantitative Data of Amaranthaceae members from Potohar Region of Pakistan

12	Digeria muricata	$12.75(16.85 \pm 1.73)20.75$	16.25 (17.85±0.53) 19.5	0.96	$1.75(2.0\pm0.11)2.25$	$0.5\;(0.6\pm0.06)\;0.75$	$0.25~(0.50\pm0.07)~0.75$
13	Kochia indica	$12.0\ (13.95\pm0.63)\ 15.50$	$12.0 (13.45 \pm 0.70) 15.50$	0.99	$0.75\;(0.95\pm0.09)\;1.25$	$0.75~(1.1\pm0.23)~2.0$	0.75 (1.0±0.13) 1.5
14	Kochia lonifolia	$13.0\ (15.25\pm1.21)\ 19.75$	$13.5 (15.50 \pm 0.75) 17.25$	0.98	$0.75~(1.05\pm0.09)~1.25$	$1.75 (1.95 \pm 0.09) 2.25$	1.5 (1.9 ± 0.12) 2.25
15	Pupalia lappaceae	$13.5(14.25\pm0.31)15.0$	11.5 (13.35±0.61) 15.25	1.06	1.5 (2.5 ± 0.34) 3.5	$0.06\ (0.06\pm0.00)\ 0.06$	$0.50~(0.55\pm0.05)~0.75$
16	Salsola lubricate	$16.0(17.4\pm0.45)18.5$	13.5 (16.7 ± 0.81) 18.0	1.06	1.25 (1.8 ± 0.2) 2.25	$1.5(1.95\pm0.14)2.25$	1.75 (2.1±0.10) 2.25
17	Sauda fruitcosa	$13.75(14.9 \pm 0.35)15.75$	14.75 (16.8±0.67) 18.25	0.90	$1.0(1.5\pm0.17)2.0$	$0.25~(1.05\pm0.44)~2.25$	$0.25~(1.55\pm0.34)~2.25$

Sr.	Taxa	No of Fertile Pollen	No of Sterile	Fertility	Sterility
No			Pollen	%	%
1	Achyranthus aspera	87	12	87.87	12.12
2	Aerva javanica	67	6	91.78	8.21
3	Amaranthus gracizens	82	9	90.10	9.89
4	Amaranthus hybridus	150	14	91.46	8.54
5	Amaranthus spinosus	120	17	87.59	12.41
6	Amaranthus viridis	164	36	82	18
7	Atriplex stoeksii	71	13	84.52	15.47
8	Celosia Argentia	109	5	95.61	4.32
9	Chenopodium album	315	88	78.16	21.84
10	Chenopodium ambrosioides	373	58	86.54	13.46
11	Chenopodium murale	430	62	87.39	12.60
12	Digeria muricata	87	11	88.77	11.22
13	Kochia indica	105	21	83.33	16.66
14	Kochia lonifolia	98	26	79.03	20.96
15	Pupalia lappaceae	135	33	80.35	19.64
16	Salsola lubricate	126	24	84	16
17	Sauda fruitcosa	86	8	91.48	8.51
	Res				

# Table 7: Fertility and Sterility percentages of examined taxa



**Plate 52:** *Achyranthus aspera* (A) Floral Parts .Scanning Electron Micrographs (B) Polar View (C) Sunken Pore orientation (D) Micro-Echinate Exine Sculpturing

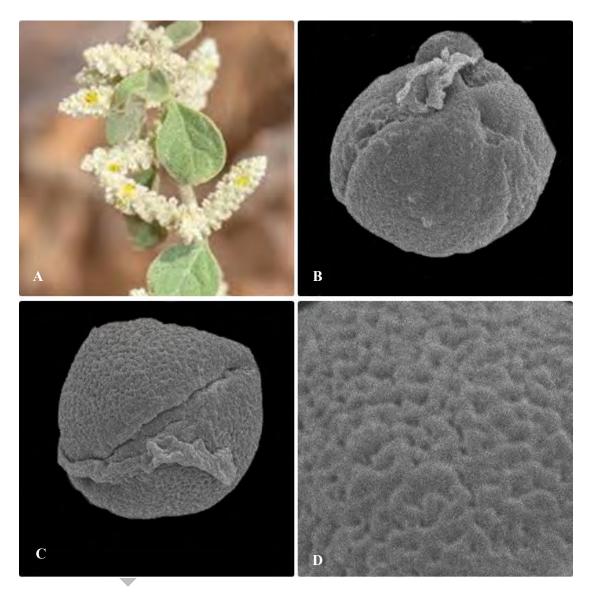
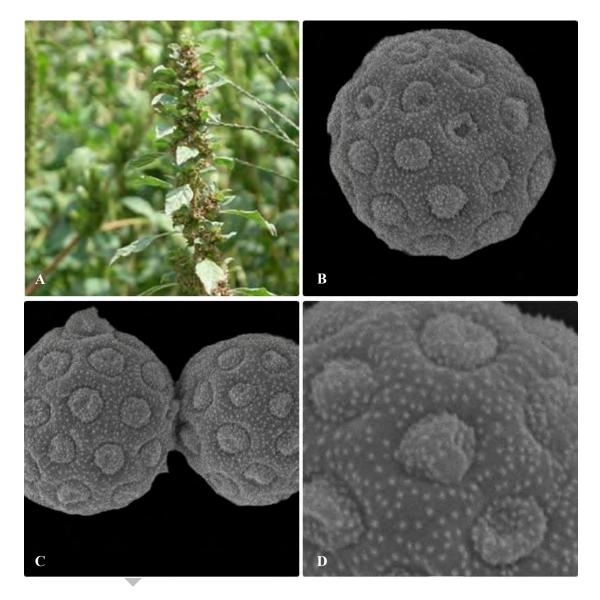
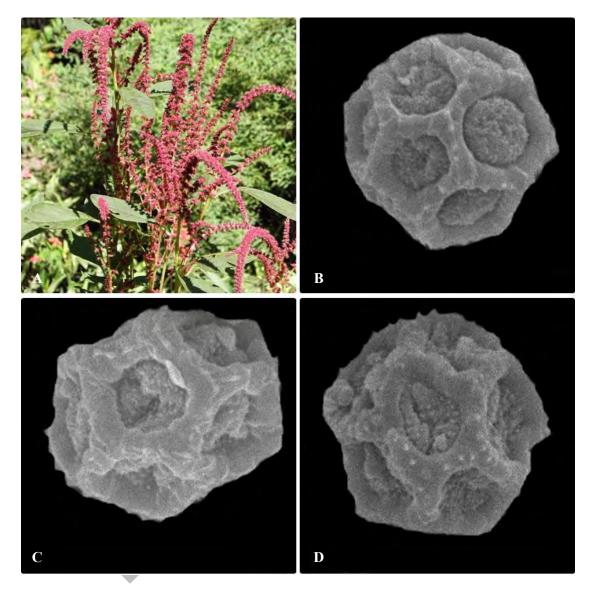


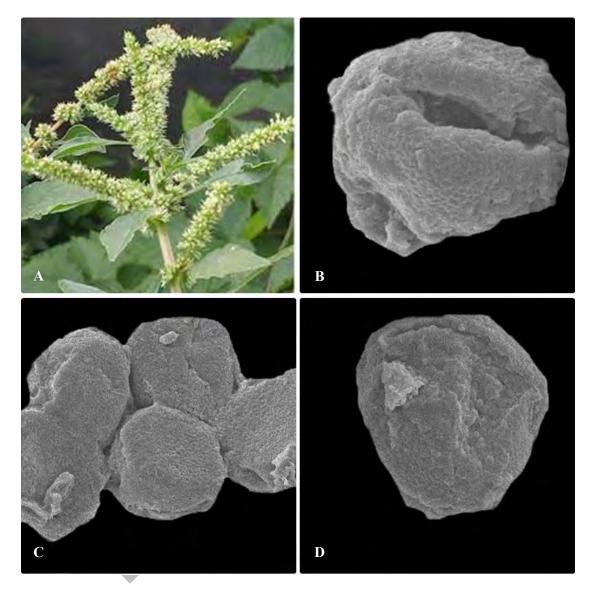
Plate 53: Aerva javanica (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Equatorial View (D) Surface Sculpturing



**Plate 54:** *Amaranthus gracizens* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore Ornamentation (D) Surface Sculpturing



**Plate 55:** *Amaranthus hybridus* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Micro-Echinate Exine Sculpturing



**Plate 56:** *Amaranthus spinosus* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Equatorial View (C) Polar View (D) Scabrate Exine Sculpturing

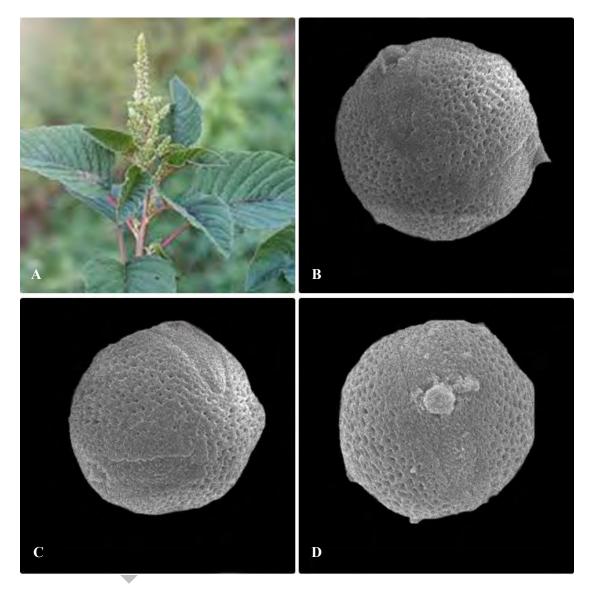


Plate 57: Amaranthus viridis (A) Floral Parts, Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Micro-Echinate Exine Sculpturing

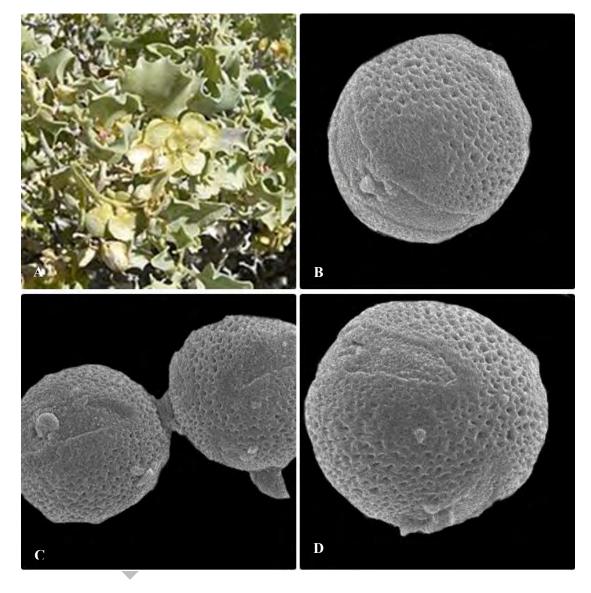
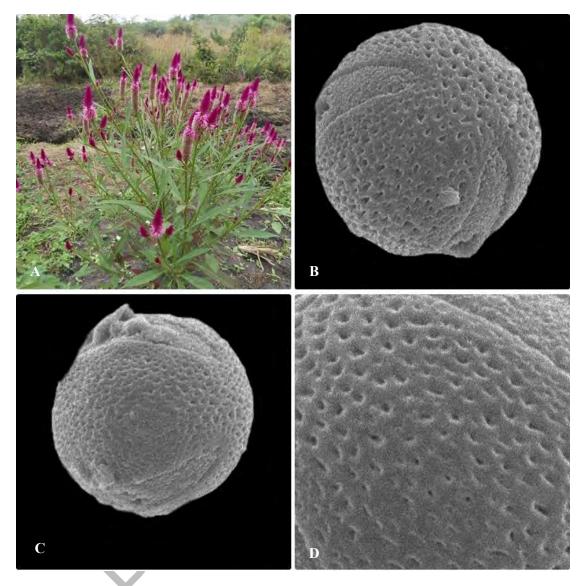
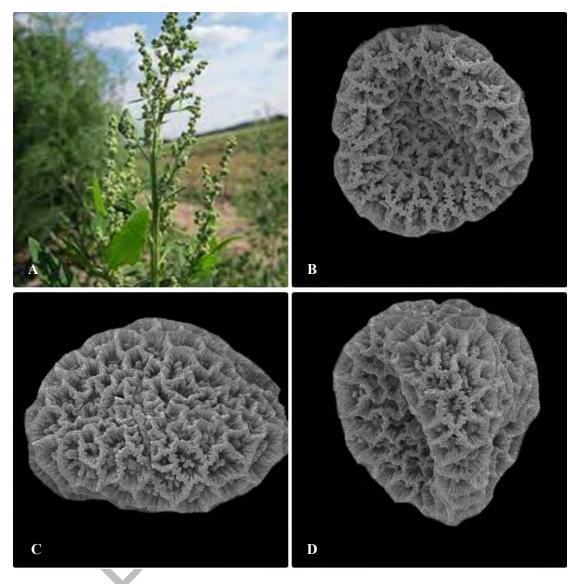


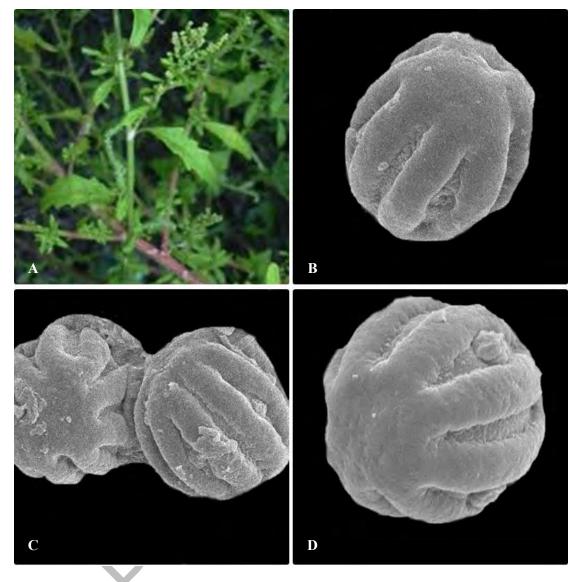
Plate 58: Atriplex stoeksii (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Sunken Pore orientation (D) Scabrate Exine Sculpturing



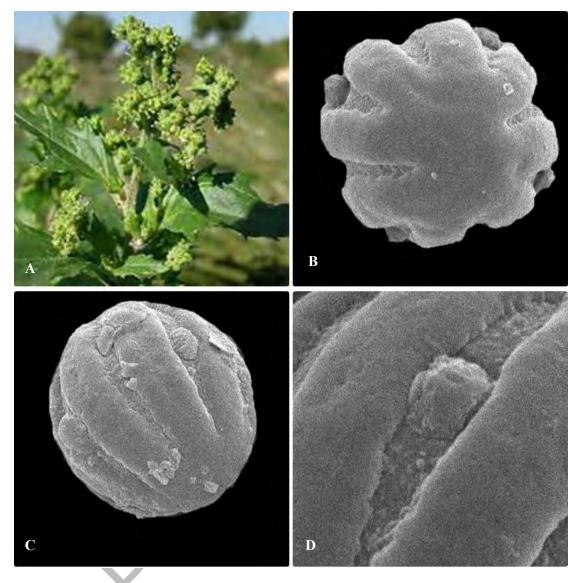
**Plate 59:** *Celosia Argentia* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Surface Sculpturing



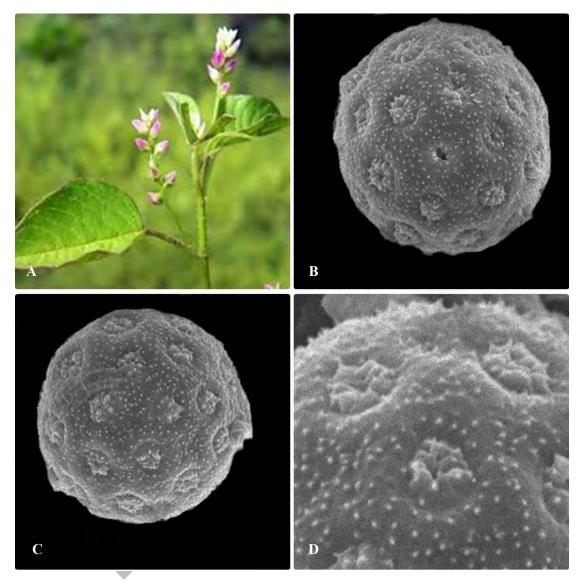
**Plate 60:** *Chenopodium album* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Scabrate Exine Sculpturing



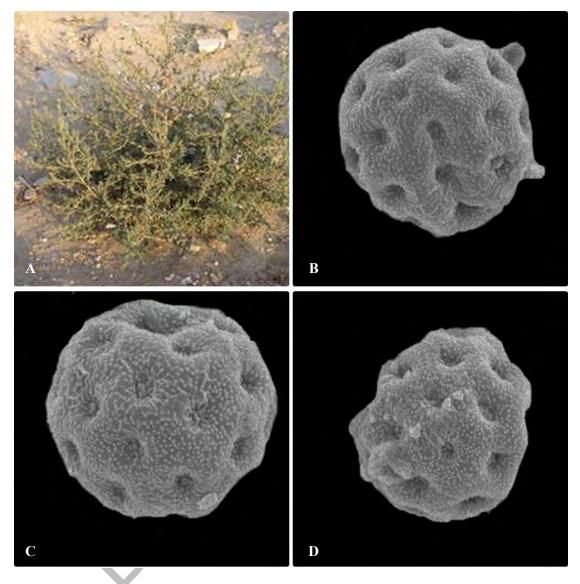
**Plate 61:** *Chenopodium ambrosioides* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken orientation (D) Micro- Echinate Exine Sculpturing



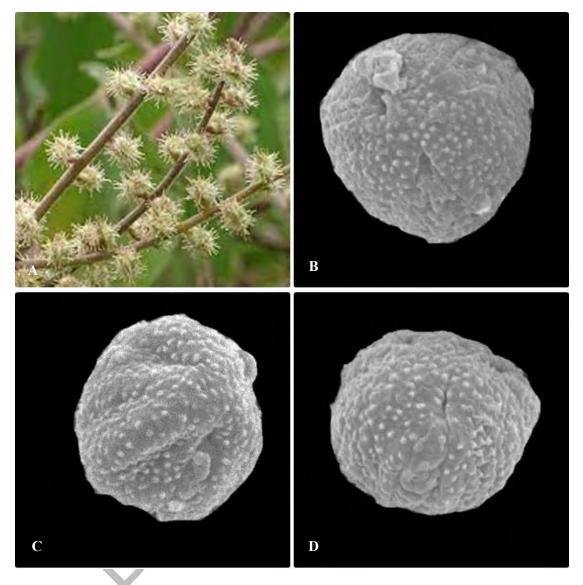
**Plate 62:** *Chenopodium murale* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing



**Plate 63:** *Digeria muricata* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B)Polar View (C) Sunken Pore orientation (D) Surface Sculpturing



**Plate 64:***Kochia indica* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Scabrate Exine Sculpturing



**Plate 65:** *Pupalia lappaceae* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Micro-Echinate Exine Sculpturing

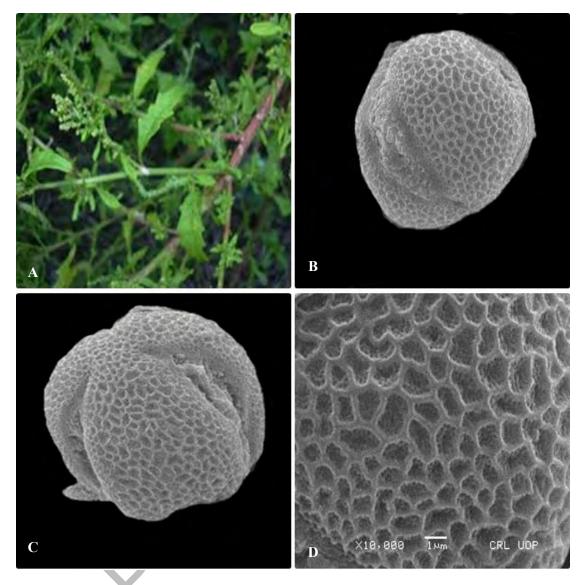


Plate 66: Sauda fruitcosa (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Sunken Pore orientation (D) Surface Sculpturing

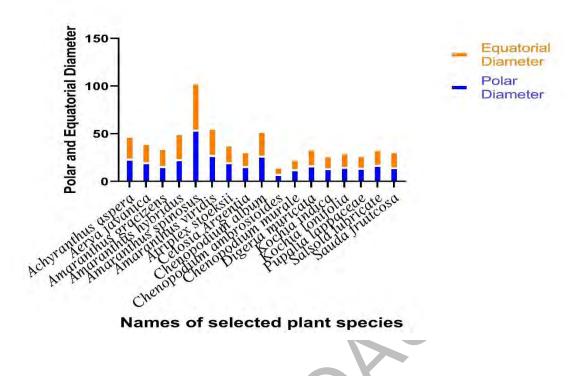


Figure 12: Mean values of Pollen diameter among Amaranthaceous taxa

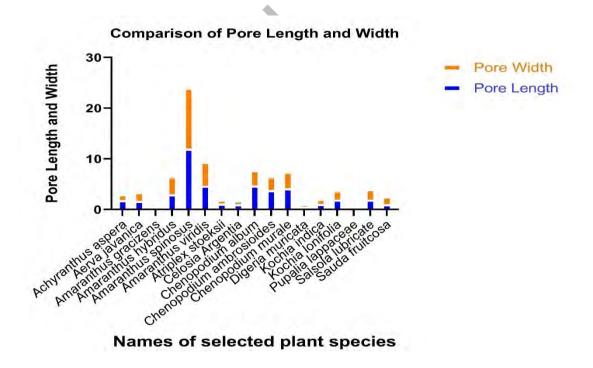


Figure 13: Data variations of pore length and width in Amaranthaceous species

Phytodiversity using Microscopic Characterization in Potohar Plateau-Pakistan

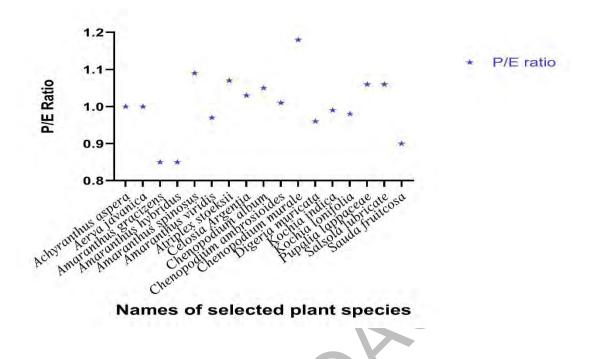


Figure 14: Variation in P/E ratio index of Amaranthaceous taxa

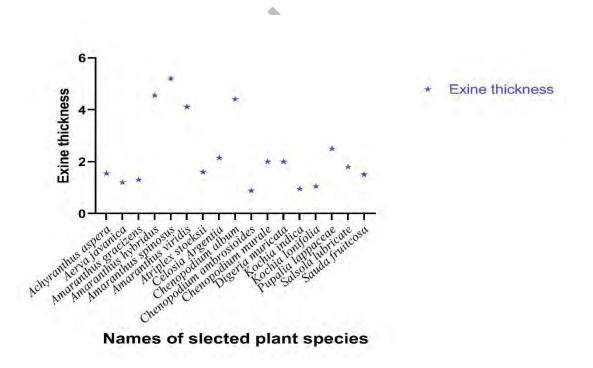


Figure 15: Mean exine thickness in Amaranthaceous Species.

Phytodiversity using Microscopic Characterization in Potohar Plateau-Pakistan

# Taxonomic key of Amaranthaceae Taxa

1	+	Polar diameter 22.75 to 25.5, P/E ratio 1Achyranthus aspera
	-	Polar diameter 12.75 to 18.25, P/E ratio 0.852
2	+	exine thickness 1 to 1.5, polar length 0.02 to 0.04Amaranthus
grcize	ens	
	-	exine thickness 1 to 1.5, polar length 1.5-1.7
3	+	polar diameter 18.25 to 21, equatorial diameter 19 to 20.75Aerva
javan	іса	
	-	polar diameter 21.2 to 26.25, equatorial diameter 27.25 to 31.254
4	+	P/E ratio 0.85, pore width 3.6Amaranthus hybridus
	-	P/E ratio 1.09, pore width 125
5	+	polar diameter 54.25 to 56.25, Equatorial diameter 49.6 to 60.5A.
spino	sus	
	-	polar diameter 27.8 to 29.7, equatorial diameter 28.5 to 316
6	+	P/E ratio 0.97, exine thickness 4.11A. viridis
	-	pore length 0.75, and pore width 0.57
7 stoeks	+ sii	polar diameter 18 to 22.75, equatorial diameter 17.75 to 18.85Atriplex
SIOCAL	511	
	-	polar diameter 13.5 to 18, equatorial diameter 13.25 to 8.58
8	+	pore length 0.75, pore width 0.5Celosia argentia
	-	pore length 4.75, pore width 39
9	+	polar diameter 27.1 to 29.2, P/E ratio 1.05Chenopodium album
	-	polar diameter 7.75 to 11, P/E ratio 1.0110
10	+	equatorial diameter 6.67 to 10.75, pore length 3.85C. ambrosioides
	-	equatorial diameter 10.75 to 12.5, pore length 4.211
11	+	polar diameter 12.75 to 14.75C. murale
	-	polar diameter 12.75 to 20.7512

12	+	equatorial diameter 16.25 to 19.5, polar length 0.5Digeria
muric	ata	
	-	equatorial diameter 12 to 15.5, P/E ratio 0.9913
13	+	pore length 0.75, pore width 0.75Kochia indica
	-	pore length 1.75, pore width 1.514
14 lonifo	+ lia	polar diameter 13 to 19.75, equatorial diameter 13.5 to 17.25K.
	-	polar diameter 13.5 to 15, equatorial diameter 11.5 to 15.2515
15	+	pore width 0.06 pore length 0.50Pupalia lappaceae
	-	pore length 1.5, pore width 1.7516
16	+	polar diameter 16 to 18.5, equatorial diameterSalsola lubricate
18.2	-	Polar diameter 13.75 to 15.75, equatorial diameter 14.75 toSauda fruitcosa

### 3.1.2 Amaranthaceous Species

Palynological features of Amaranthaceae family was investigated for qualitative and quantitative characters by utilizing Light microscopy and scanning electron microscopy. In the current research, 17 species belonging to 11 genera were studied for examining pollen characters. All the pollens have the same shape category i-e spheroidal. All pollens of studied species are porate with sunken apertures. Pore number exhibit variations. Plants with triporate to polyporate structure were observed. Variations were observed in terms of pollen ornamentation. 8 species has scabrate pollens while 9 studied species has micro-echinate pollen grains. The fertility and sterility ratio has been estimated for each species. Maximum fertility was observed in *Celotia argentia* (95.61 %) while maximum sterility percentage was observed in *Chenopodium album* (21.84 %). The percentage illustrate the pollens were collected during the mature flowering season. Micrographs (LM and SEM) are also illustrated.

## Qualitative and Quantitaive Characters of Pollen and P/E ratio

All the studied species belongs to spheroidal shape group. Maximum polar diameter 54.25  $\mu$ m (51.25-56.25) is observed for *Amaranthus spinosus* while minimum polar diameter 18.25  $\mu$ m (20.2±0.50) 21.0 is observed for *Aerva javanica*. In case of equatorial diameter, minimum mean value 10.75  $\mu$ m (12.5-9.75) is calculated for *Chenopodium murale* while maximum value 49.6  $\mu$ m (38.75-60.50) is studied for *Amaranthus spinosus*. The pollen shape is determined on the basis of P/E ratio which ranges from 0.85 (*Amaranthus gracizens*) to 1.18 (*Chenopodium murale*).

### **Morphology of Pores**

All the studied plant species have pore uniformly present on their surface. The number of pores varies among species. The pore number is recorded from 3 (*Digeria muricate, Amaranthus viridis*) to 15 (*Amaranthus spinosus*). All the studied species have sunken pores. Maximum pore length is observed for *Chenopodium murale* while minimum pore length is observed for *Amaranthus grezians*. In case of pore width, maximum pore width value is calculated for *Amaranthus spinosus* while minimum pore width value is studied for *Diigaria muricata*.

### **Exine Thickness**

Exine thickness was noted by using SPSS software. Mean, minimum and maximum values were calculated for each species. Minimum mean values were obtained for *Kochia indica* 0.75  $\mu$ m (0.95±0.09) and *Kochia lonifolia* 0.75  $\mu$ m (1.05±0.09) whereas maximum range was recorded for *Amaranthus spinosus* 5.20  $\mu$ m (4.50-6.0).

In this research work, Light and scanning electron microscope was taken into consideration to detect pollen of family Amaranthaceae from Salt Range Pakistan. The studied species have pantoporate and sunken pores. The pollen grain of family Amarantahceae have spheroidal pollen shape based on P/E ratio. Number of pore are various in different species of the Amaranthus family range from 3 in *Digeria muricate, Amaranthus viridis* to 15 in *Amaranthus viroides* stated.

The exine of pollen of Amarnathaceae were thin walled (Ghazalli., 2022; Nazish et al., 2009), our investigated species exine size was also have thin wall. Spheroidal pollen shape in Caryophyllaceae reported in current study agree with that of previous literature (Nazish et al., 2009, Chin et al., 2011, Pino et al., 2016). In present report, the size of exine of *Sauda fruticosa* was observed slightly larger. Different researcher work on the pollen morphology of Amaranthaceae illustrated the small sized pollen grains 5 to 20 um (Asadi et al., 2016; Angelini et al., 2014 ; Hermin et al., 2012; Nazish et al., 2019). Our results agreed with the above studied, In all reported species of Amaranthaceae zoopoartae pollenss were studied , and they all are sunken. Nazish et al. (2019) and Noor et al., (2021) also described such types of pori in the family Amaranthaceae . Nazish et al. (2019), Asadi Et al., (2016), and Hermin et al (2012) used pori number for the taxonomic identification of species in Amaranthaceae. Zooporate type of pollen stated in this research work, and previous researcher also reported same in Amaranthus family (Borsch 1998; Nazish et al., 2019).

Noteworthy dissimilarities in size and sculpture of pollen displays great taxonomic potential in identification and species delimitation (Borsch 1998). Previous workers has demonstrated Amaranthaceae as significant floral family because of its distinct pollen morphology. Accordance to present research, the studied family is very much similar pollen morphology with previous study. Amaranthacaee family is devoid of echini on the surface of pollen. (Nazish et al., 2019, Asadi et al., 2016). Two types of ornamentation microechinate and scabrate were observed here which are in accordance to previous authors (Brosch 1998; Asadi et al., 2016; Angelini et al., 2014). Palyno-morphological multiplicity delivers indication about exine thickness, pore ornamentation and pollen type. In our finding, pollen type was spheroidal in all studied species. Zooporate pollen was the basic and primitive type in Amaranthaceae (Brosch 1998; Asadi et al., 2016; Nazish et al., 2019). The pollen ornamentation of the 9 species (*Aerva javanica, Amaranthus hybridus, Amaranthus spinosus ,Amaranthus viridis ,Celosia argentia, Chenopodium ambrosioides, Chenopodium murale, Pupalia lappaceae*) exhibited micro-echinate ornamentation while 8 species (*Achyranthus aspera ,Amaranthus gracizens ,Atriplex stoeksii ,Chenopodium album ,Digeria muricate ,Kochia indica ,Kochia lonifolia*) belongs to scabrate ornamentation. The current findings were found to be similar with previous literature (Nazish et al., 2019, Asadi et al., 2016).

The microscopic examination of pollen morphology exemplify, pollen diameter, pore diameter and P/E ratio play important role in the identification of species (Nazish et al., 2009, Chin et al., 2011, Pino et al., 2016). In present study the pollen diameter in equatorial and polar view, number of pore, exine thickness, P/E ratio, length and width of pore, qualitatively shape of pollen and pollen sculpturing as significant taxonomic tools were described.

Sr. No	Taxa	Pollen Size	Symmetry	Aperturation	Sculpture type	Exine Texture
1.	Acacia nilotica	Medium	Radially symmetric	Tricolporate	Reticulate	Thin
2.	Acacia modesta	Medium	Radially symmetric	Tricolporate	Reticulate	Thick
3.	Arachis hypagae	Small	Radially symmetric	Bicolpate	Reticulate	Thin
4.	Astragalus psilocentros	Medium	Radially symmetric	Tricolporate	Reticulate	Moderately thick
5.	Astragalus scoripiurus	Small	Bilitterally symmetric	Bicolpate	Perforate	Moderately thick
6.	Cassia fistula	Medium	Bilitterally symmetric	Tricolporate	Perforate	Moderately thick
7.	Cassia italica	Small	Bilitterally symmetric	Tricolpate	Psilate	Moderately thick
8.	Cassia occidentalis	Small	Radially symmetric	Tetracolpate	Perforate	Moderately thick
9.	Crotolaria burhia	Small	Radially symmetric	Tricolporate	Reticulate	Thin
10.	Dilbergia sisso	Small	Radially symmetric	Bicolpate	Psilate	Thin
11.	Lathyrus aphaca	Medium	Bilitterally symmetric	Bicolporate	Perforate	Thin
12.	Lespedeza juncea	Small	Radially symmetric	Bicolpate	Reticulate-Verrucate	Moderately thick
13.	Leucaena leucocephalla	Medium	Radially symmetric	Bicolpate	Verrucate	Thin
14.	Melilotus indicus	Medium	Bilitterally symmetric	Bicolpate	Reticulate	Thin
15.	Pigomia pinnata	Small	Radially symmetric	Tricolpate	Reticulate-Vrrucate	Moderately Thick
16.	Prosopis cineraria	Small	Bilitterally symmetric	Tricolporate	Perforate	Thin
17.	Prosopis juliflora	Medium	Bilitterally symmetric	Tricolpate	Reticulate-Perforate	Thin
18.	Pueraria tuberosa	Medium	Radially symmetric	Bicolpate	Reticulate	Thick
19.	Rhynchosia capitata	Small	Radially symmetric	Tricolpate	Reticulate-Perforate	Moderately thick
20.	Sophora mollis	Small	Radially symmetric	Tricolporate	Reticulate-Perforate	Thin
21.	Tephrosia purpurea	Small	Bilatterally symmetric	Bi-Tricolporate	Reticulate- Perforate	Thin
22.	Vicia angustifolia	Medium	Radially symmetric	<b>Bi-Tricolporate</b>	Reticulate-Perforate	Thin

**Table 8:** Qualitative data of pollen of family Fabaceae members from Potohar region of Pakistan

Plant name	Polar Diameter	Equatorial Diameter	P/E	Exine Thickness	Colpi Length	Colpi Width	Fertility %
Acacia nilotica	23.25 (26.25-21.2)±2.09	27.25 (31.25-23.7)±2.85	0.85	3.90(3.25-3.75)±0.26	2.45(2.00-3.00)±0.18	3.40(2.75-4.25)±0.26	92.6
Accacia modesta	54.25(51.25-56.25)±0.88	49.60(38.75-60.50)±0.29	1.09	20.2 (19.7-22.0)	3.95 (2.75-4.75)	2.70 (2.00-3.25)	81.2
Arachis hypagae	27.8(26.5-29.7)±0.68	28.5(26.2-31)±0.78	0.97	2.5(2-3.4)±0.22	2.2(0.95-2.8)±0.43	1.1(0.7-1.5)±0.18	60
Astragalus psilocentros	27.1(25.5-29.2)±0.71	25.8(22.7-31.5)±1.49	1.05	0.74(0.5-1.05)±0.35	0.28(0.12-0.56)±0.07	0.16(0.10-0.23)±0.02	59.5
Astragalus scoripiurus	12.75 (14.75-9.50)±2.06	10.75 (12.5-9.75)±1.04	1.18	2.55(1.75-300)±0.24	1.80(0.75-300)±0.39	3.10(2.75-3.75)±0.18	76.2
Cassia fistula	7.75(11.00-5.25)±0.88	7.67(10.75-5.50)±0.78	1.01	3.35(3-3.7)±0.12	4.25(3.75-4.75)±0.17	7.95(7.00-9.75)±0.48	78.16
Cassia italic	19.4(17.7-20.5)±1.14	18.3(16.5-20.5)±1.70	1.06	2.00 (2.75-1.2)±0.55	3.25 (4.00-2.75)±0.46	4.55 (5.50-3.75)±0.77	87.39
Cassia occidentalis	25(22.6-26.7) ±1.49	26.3(21.7-30.1)±2.61	0.95	2.45(2.00-3.00)±0.18	3.65(2.75-4.75)±0.40	6.25(5.25-7.00)±0.30	86.54
Crotolaria burhia	41.2(37.5-43.2)±0.93	36.9(34.7-39.5)±0.77	1.11	4.9(3.25-6.2)±0.52	6.25(4.50-8.00)±0.69	7.10(6.50-8.00)±0.57	83.33
Dilbergia sisso	18.03(16.80-18.6) ±0.34	17.85(15.30-19.65) ±.0.8	1.01	20(21.00-18.7)±0.41	7.75(8.50-7)±0.29	3.30(4.00-2.75)±0.22	84.61
Lathyrus aphaca	20(19.7-20.4)±0.13	19.6(18.2-20)±0.24	1.02	3.35(2.25-4.7)±0.53	2.45(2.00-3.00)±0.18	2.55(2.10-3)±0.18	80.63
Lespedeza juncea	30.8(26.2-35.2)±1.82	24.5(20.2-30.2)±1.96	1.25	3.75(2.75-4.7)±0.35	2.35(1.75-3.00)±0.23	4.85(4.25-5.50)±0.23	89.1
Leucaena leucocephalla	23.3(16.7-30.2) ±3.12	21.2(12.6-27.7) ±2.32	1.09	1.05(0.75-1.2)±0.09	5.2(4.2–6.1)± 0.15	6.4(5.7–7.3)± 0.15	87.6
Melilotus indicus	13.85(15.50-12.00)±0.65	13.9(16.25-11.50)±0.66	0.99	5.35(2.75-6.7)±0.69	6.50(5.25-775)±0.44	7.00(6.00-7.75)±0.37	74.3
Pigomia pinnata	42.4(34.2-52.2)±3.25	41.6(35.5-48.7)±02.52	1.01	0.52(8.75-6.25)±.52	3.20(3.50-3.00)±.09	1.75(2.75-1.25)±0.13	77.8
Prosopis cineraria	33.0(30.2-36.2)±1.10	32.0(28.7-35.2)±1.11	1.03	2.65(2.25-3.0) ±0.12	$4.8(4.4-5.4)\pm 0.29$	2.7(2.4-3.1)± 0.2	85
Prosopis juliflora	20.25 (23.75-18.7)±1.00	27.25 (31.25-23.7)±1.27	1.34	1.80 (3.00-1)±0.34	3.25 (3.75-2.75)±0.17	5.60 (6.25-5.25)±0.18	91.3
Pueraria tuberosa	40.1(39.5-40.7)±0.21	38.4(35.7-43.0)±1.21	1.04	4.6(3.9-5.2)±0.18	1.18(0.75-1.85)±0.18	1.3(0.35-2.1)±0.25	73.4
Rhynchosia capitata	36.6(34.7-40.2)	48.9 (44.7-52.7)	1.25	2.65(2.00-3.2)±0.23	3.70(2.75-5.25)±0.42	7.00(5.25-8.00)±0.48	77.7
Sophora mollis	32.3(19.5-36.10±2.78	22.6(18.2-25.7)±1.09	1.42	3.45(4.7-6.25) ±0.29	5.30(6.23-9.75) ±0.54	$5.80(5.40-7.65)\pm0.42$	81.3
Tephrosia purpurea	35.2(30.3-41.2)±1.94	34.3(32.2-36.7)±0.84	1.02	2.70(2.00-3.5)±0.26	3.60(2.75-4.25)±0.26	4.90(4.50-5.50)±0.20	65.5
Vicia angustifolia	37.6(33.0-42.7)±1.85	29.5(25.2-35.2)±1.64	1.27	4.55 (5.25-4)±0.48	21.9(19.7-23.2)±0.6	4.75 (5.25-4.25)±0.39	83.7

**Table 9:** Quantitative data of pollen of family Fabaceae members from Potohar region of Pakistan

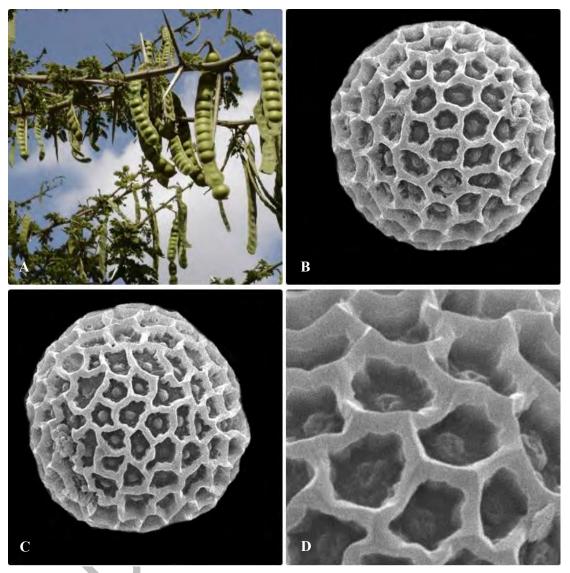


Plate 67: Acacia nilotica (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Reticulate Exine Sculpturing (D) Surface Sculpturing

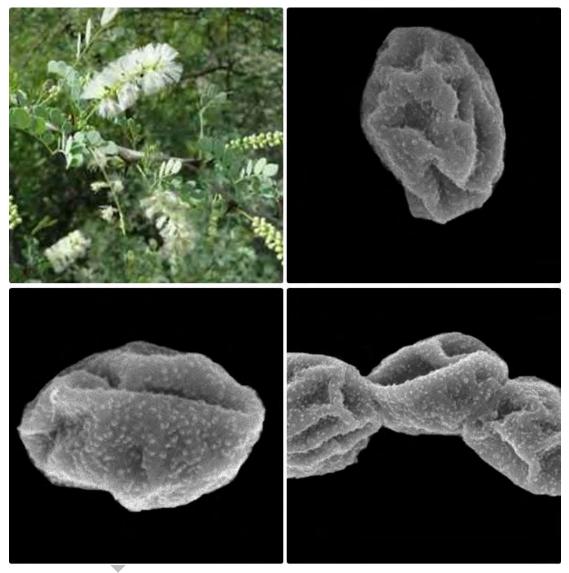
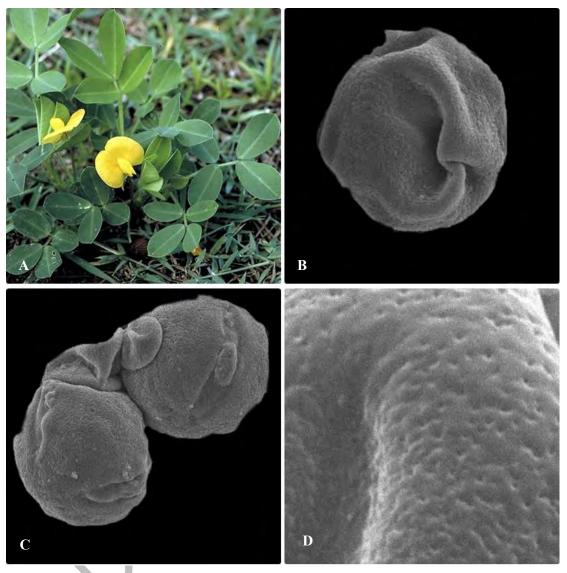
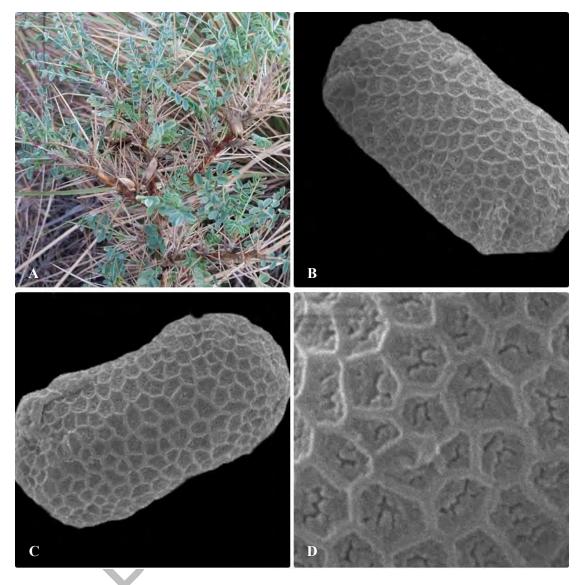


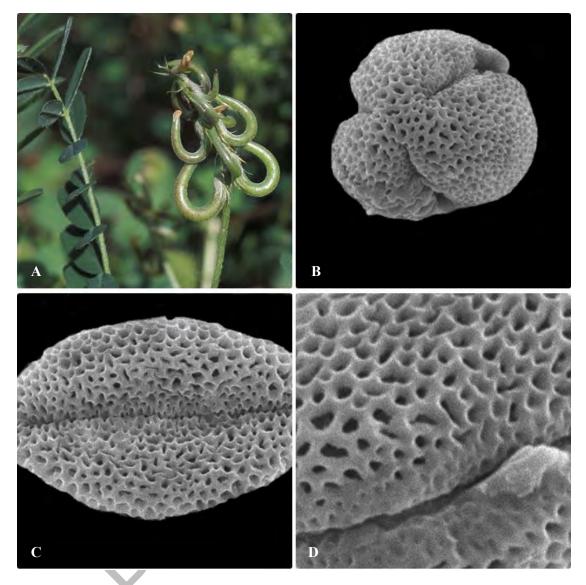
Plate 68: Acacia modesta(A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Reticulate orientation (D) Exine Sculpturing



**Plate 69:** Arachis hypagae (A )Floral Parts , Scanning Electron Micrographs of Pollen (B)Polar View (C) Scabrate Surface (D) Exine Sculpturing



**Plate 70:** *Astragalus psilocentros* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B)Polar View (C) Moderately Thick Exine Texture (D) Surface Sculpturing



**Plate 71:** *Astragalus scoripiurus* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B)Polar View (C) Equatorial View (D) Surface Sculpturing

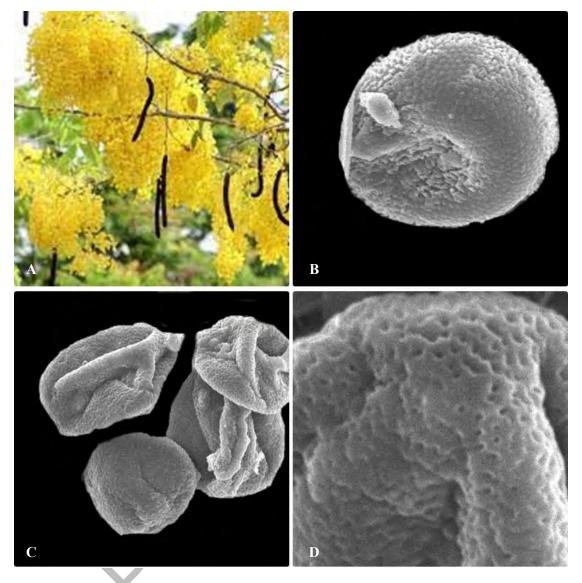
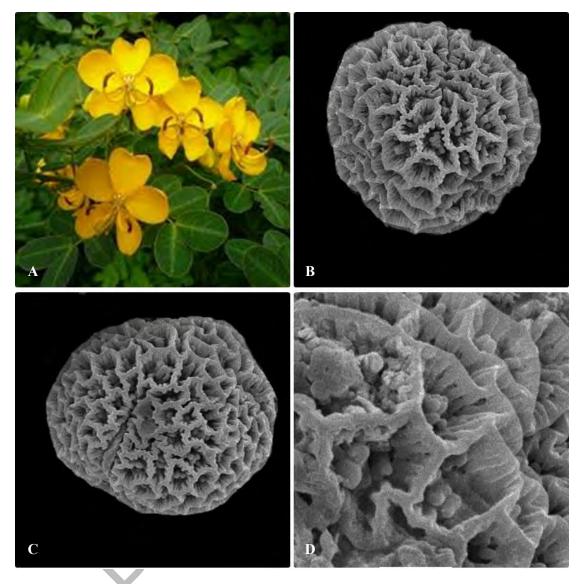
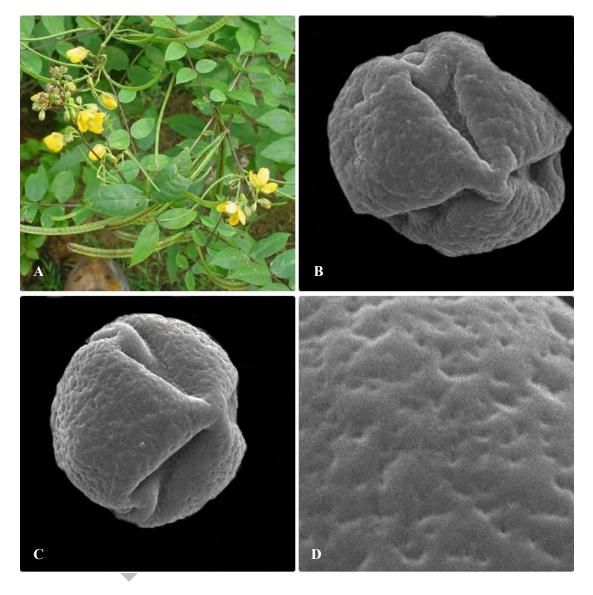


Plate 72: Cassia fistula (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Equatorial View (D) Surface Sculpturing



**Plate 73:** *Cassia italica* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B)Polar View (C) Moderately Thick Exine (D) Surface Sculpturing



**Plate 74:** *Cassia occidentalis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Perforate Sculpturing (D) Surface Sculpturing

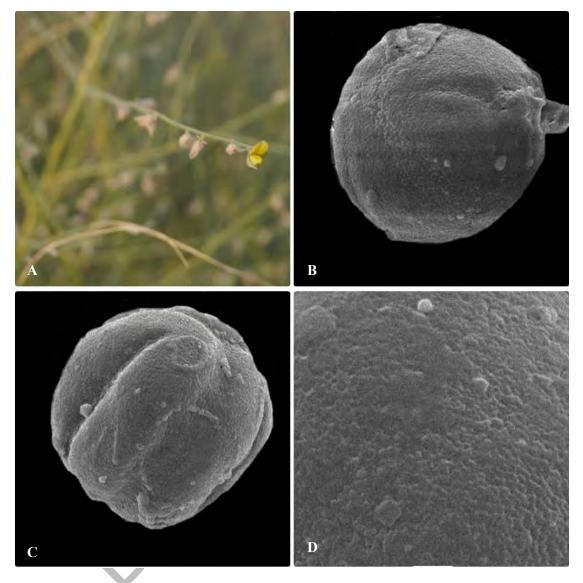


Plate 75: *Crotolaria burhia* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing

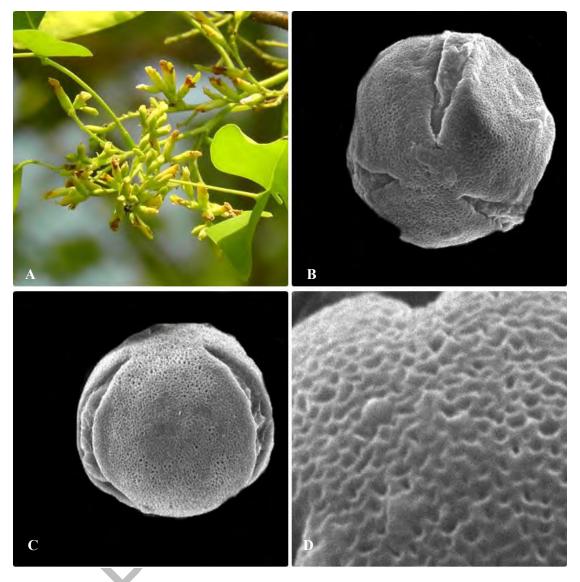


Plate 76: *Dilbergia sisso* (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Thin Exine Texture (D) Surface Sculpturing

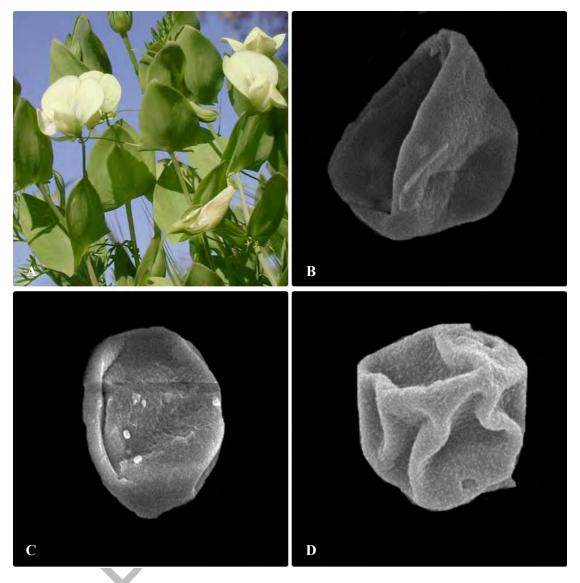
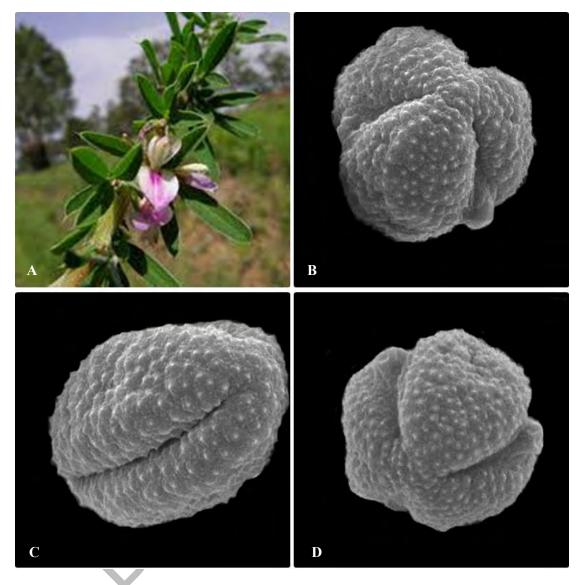
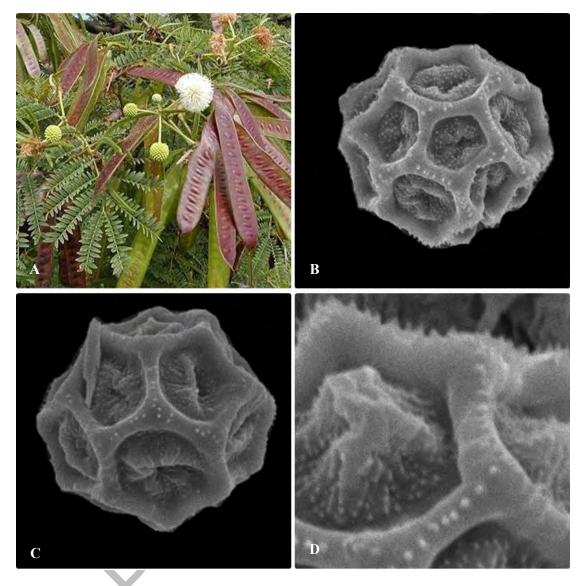


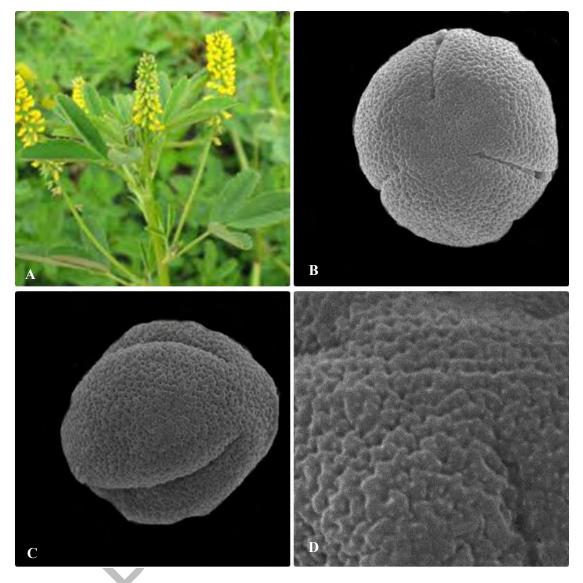
Plate 77: *Lathyrus aphaca* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B)Equatorial View (C) Thin Exine Texture (D) Perforate Sculpturing



**Plate 78:** *Lespedeza juncea* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Moderately Thick Exine Sculpturing



**Plate 79:** *Leucaena leucocephalla* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Verrucate Exine Sculpturing (D) Surface Sculpturing



**Plate 80:** *Melilotus indicus* (A) Floral Parts, Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing

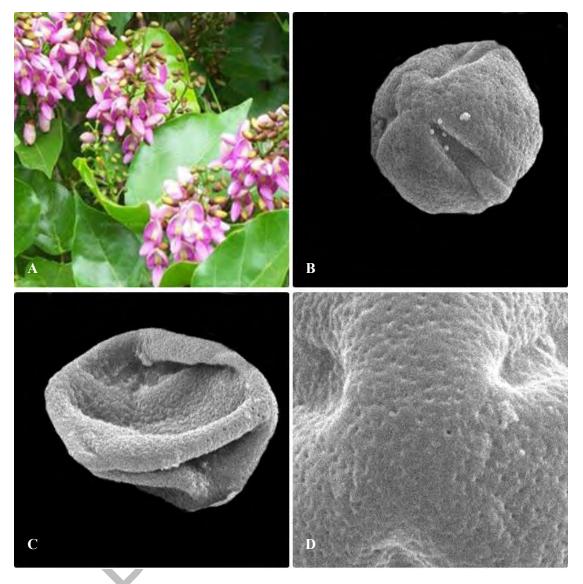
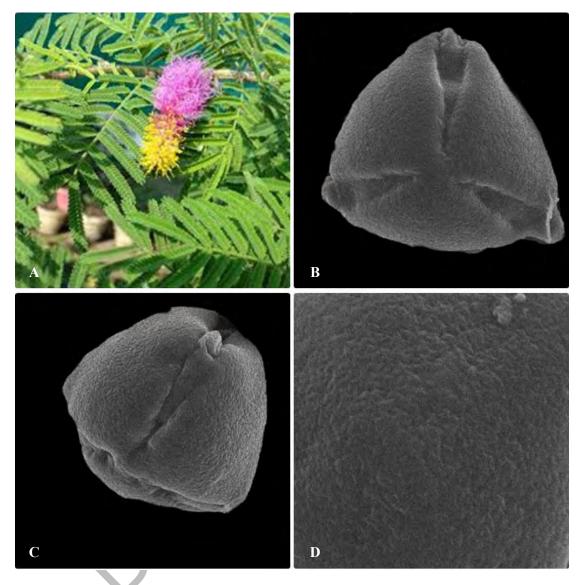
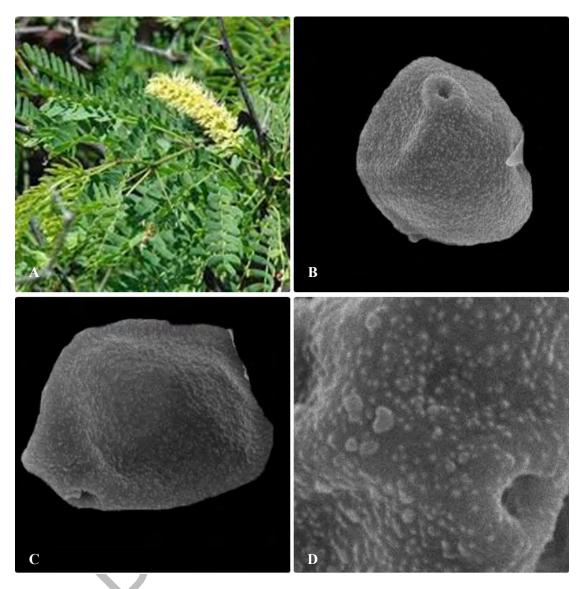


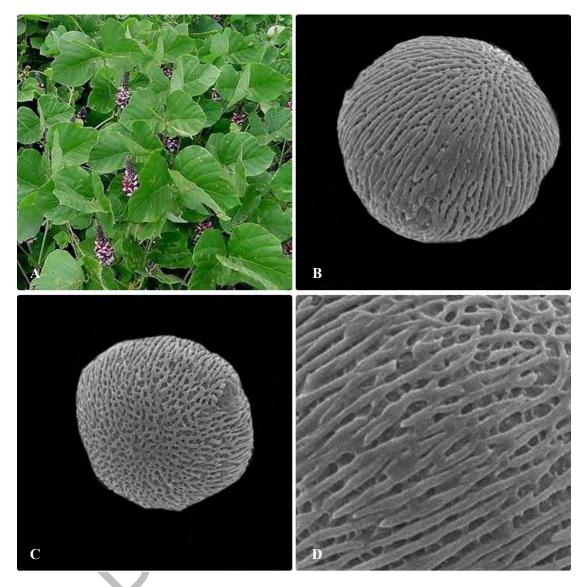
Plate 81: *Pigomia pinnata* (A )Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing



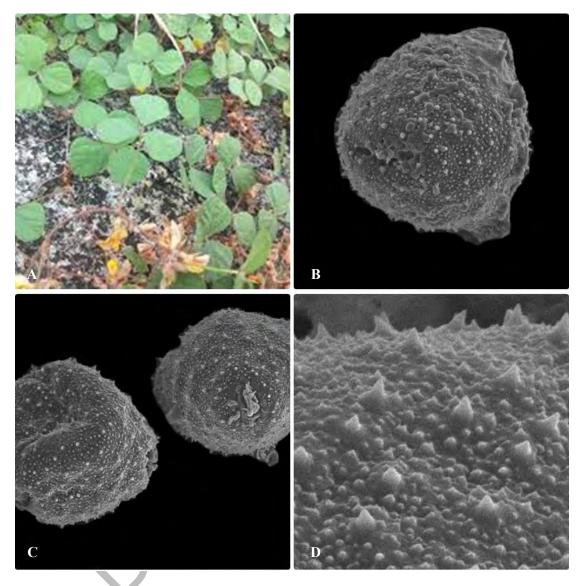
**Plate 82:** *Prosopis cineraria* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing



**Plate 83:** *Prosopis juliflora* (A) Floral Parts, Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing



**Plate 84:** *Pueraria tuberosa* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Reticulate Exine Sculpturing (D) Surface Sculpturing



**Plate 85:** *Rhynchosia capitata* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Reticulate Perforate Exine Texture (D) Surface Sculpturing

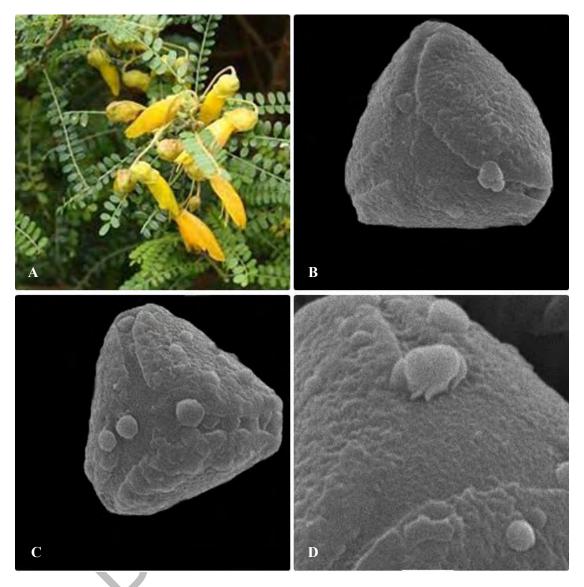
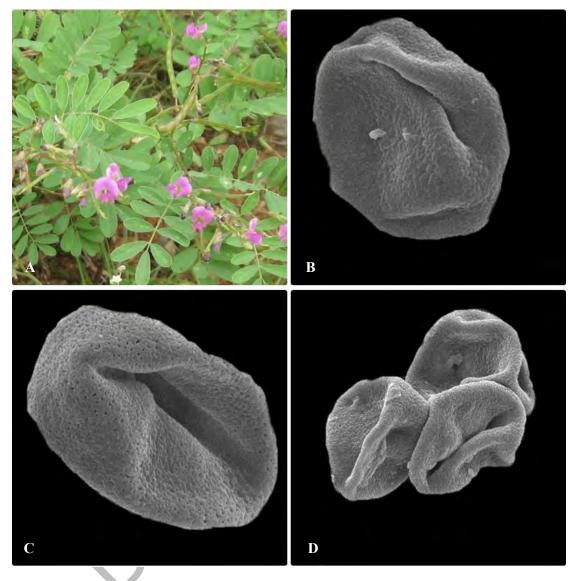
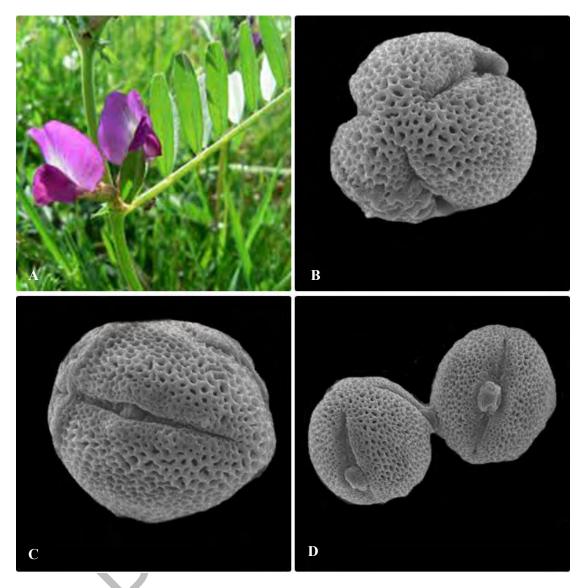


Plate 86: *Sophora mollis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Perforate Exine Sculpturing (D) Surface Sculpturing



**Plate 87:** *Tephrosia purpurea* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Reticulate sculpturing(D) Thin Exine Texture



**Plate 88:** *Vicia angustifolia* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Reticulate-Perforate Sculpturing

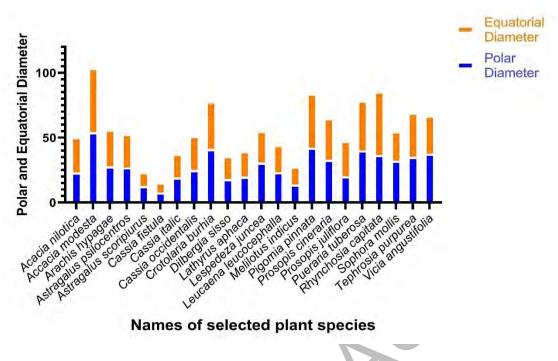


Figure 16: Mean values of Pollen diameter among Fabaceous taxa

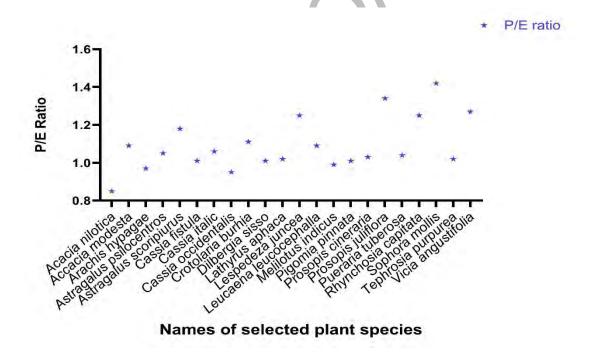


Figure 17: Variation in P/E ratio index of Fabaceous taxa

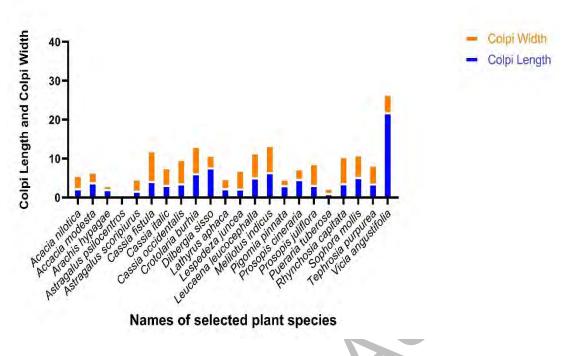


Figure 18: Mean data variations of colpi length and width in Fabaceous taxa

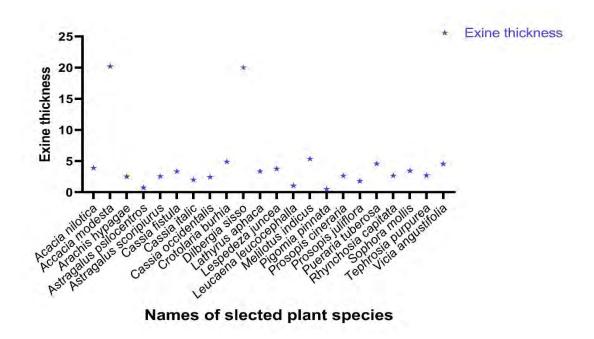


Figure 19: Mean exine thickness in Fabaceous Species.

## Taxonomic Key Based on Fabaceaos Pollen

1a. Pollen size is small   2
1b. Pollen size is medium 10
<ul><li>2a. Pollen aperture is tricolpoate</li></ul>
3a. Exine sculpturing is verrucate Leucaena leucocephalla
3b. Exine sculpturing is reticulate or perforate
4a. Apertural condition is bi-tricolporate
4b. Apertural condition is bicolpate or tricolporate
5a. Pollen sculpture is reticulate-perforate Prosopis juliflora
5b. Pollen exine is reticulate or perforate
6a. Polar diameter is above 50 μm Accacia modesta
6b. Polar diameter is blow 50 μm7
7a. Fertility is below 60% Astragalus psilocentros
7b. Fertility is above 60 %
8a. Exine thickness is moderately thick
8b. Exine thickness is thin
9a. Exine sculpturing is reticulate
9b. Exine sculpturing is perforate Lathyrus aphaca
10a. Tetra colpate pollen Cassia occidentalis
10b. Bicolpate, tricolpate or tricolporate pollen 11
11a. P/E ratio is below 1 Arachis hypagae
11b. P/E ratio is above 1 12

12 a. Colpi width is below 2 μm Pigomia pinnata
12b. Colpi width is above 2 μm
13a. Colpi length is above 7 μm Dilbergia sisso
13b. Colpi length is below 7 μm14
14a. Exine sculpturing is psilate Cassia italica
14 b. Exine sculpturing is not psilate 15
15a. Exine thickness is above 4 μm Crotolaria burhia
15b. Exine thickness is below 4 μm
16 a. Sculpturing of exine is perforate Astragalus scoripiurus
16b. Exine sculpturing is reticulate 17
17a. Bilateral symmetry of seed <i>Tephrosia purpurea</i>
17b. Radial symmetry of seed
18 a. Exine is thin Sophora mollis
18b. Exine is moderately thick 19
19a. Exine sculpture is reticulate verrucate <i>Lespedeza juncea</i>
19b. Eine sculpture is reticulate perforateRhynchosia capitata

## **3.1.3 Fabaceaous Species**

Astragalus is the biggest and one of the problematical genus from taxonomic point of view. The rich variety of the genus has engrossed the consideration of many systematists in various regions worldwide. Pinar et al. (2014), observed pollens with prolate, subprolate, and prolate-spheroidal shapes in *Astragaleae* (genus *Astragalus*). Akan and Aytac (2014), carried out study on pollen morphology of the section Alopecuroidei DC. of genus Astragalus and reported subprolate or prolatespheroid pollen type in the members of this section. The pollen is observed with tricolporate aperturation and the reticulate ornamentation with semiangular amb type (Akan et al., 2005). Oskouian, Ousalou, and Masoumi (2007) examined pollen morphology of 37 Astragalus species in section Malacothrix and its allies. They reported that the most common exine surface sculpturing is reticulate and pollen shape ranges from spheroidal to prolate. Ekici, Yüzbaşıoğlu, and Aytaç (2005) studied pollen characters of Astragalus ovalis and reported prolate pollen with tricolporate aperturation and reticulate sculpturing. Ceter et al. (2013) examined the variations in pollen characters of Astragalus L. section Hololeuce Bunge in Turkey. The study concluded that the pollen grains appeared subprolate and prolate-spheroidal with few prolate shaped pollens having trizonocolporate and operculate pattern of apertures. Colpi were reported to be long and narrow with perforate or reticulate sculpturing pattern. İlçim and Behçet (2016) carried out palynological investigation on Astragalus topalanense and recorded subprolate, tricolporate pollen grains with tricolporate sculpturing pattern. The previous reports on Astragalus corroborate with recent findings in which genus has been reported with prolate, subprolate, and prolatespheroidal pollen grains. Ceter et al. (2013) described pollen morphology of genus Oxytropis and reported isopolar, radially symmetric, prolate, or subprolate pollens with great variation in polar and equatorial diameter. These results are in harmony with the pollen features of genus Oxytropis recorded in this study. The findings of this study are supported by previous literature including; radially symmetrical, isopolar pollen with trizonocolporate aperturation in genus Astragalus (Pinar et al., 2014), three type of apertures observed in members of papilionoidae (Schrire & Sims, 1997), tricolporate and trizonocolporate pollen aperturation pattern in Astragaleae (Oxytropis) (Ceter et al., 2013); (Pavlova, 2013).

The variations in apertures may be due to genetic factors like ploidy level and environmental circumstances such as temperature fluctuation, high or low humidity, altitude, and pollination activity (do Pico & Dematteis, 2010). Earlier researchers reported that variations that exist in aperture types may be due to heteromorphy in pollens (Inceoglu, 1973). Pinar et al. (2014) studied the LM and SEM features of pollens in section *Onobrychoidei* in genus *Astragalus* and distinguished two types of sculpturing patterns i-e microreticulate and reticulate. Ceter et al. (2013) and (Pavlova, 2013) described microreticulate, psilate, and perforate exine sculpturing in genus *Oxytropis*.

The examined pollen grains in *Oxytropis* species appeared to be small having bi-tricolporate apertures and slight disparity or specialization. These characters closely resemble to pollen grain features in Astragalus thus validating previous studies. The tectum was observed to be restricted and primarily reticulate (O. *lapponica*) or reticulate-perforate (O. staintoniana). These findings harmonizes with the phylogenetic trend thus suggesting the specialization within genera and delimiting species as well (Ferguson, 1981). The delimitation of taxa in Oxytropis is difficult on the basis of pollen characters, even though a few apparent grouping of species could be predictable. The aperturation pattern is of huge significance for classification of the pollen type at genus and tribe level and can also predict associations (Ferguson, 1981). The tendency of increasing the aperture number in members of Fabaceae was explained by Ferguson (1981) as well as Guinet and Ferguson (1989). A little information is present in literature to explain the variation in apertures in genus Astragalus (Pinar et al., 2009) and Oxytropis (Pavlova, 2013). The pollen wall stratification is considered as a significant character in classification at tribe level but it has less importance at species due to absence of significant variations (Ferguson, 1981). The investigation of the pollen morphological characters is important for certain taxonomical judgments particularly above the rank of species (Amina et al., 2020; Bano et al., 2020). In general, the data on pollen morphology offer reliable facts for resolving taxonomical problems and confer about resemblance as well as association between species.

Sr.No	Plant taxa	Pollen Size	Pollen Type	Pollen shape	Aperture condition	Pore orientation	Exine sculpturing
1	Brachiaria brizantha	Medium	Monoporate	Subprolate	Monoporate	Prominent	Perforate, scabrate
2	Cenchrus biflorus	Medium	Monoporate	Prolate	Monoporate	Sunken	Scabrate, psilate and sparsely
							verrucate
3	Cenchrus ciliaris	Large	Monoporate	Subprolate	Monoporate	Slightly sunken	Sparsely reticulate scabrate
							and rugulate at the poles
							and rest of the area reticulate
4	Cenchrus pennisetiformis	Large	Monoporate	Prolate	Monoporate	Oriented	Gemmate-verrucate
5	Cymbopogon jwarancusa	Medium	Monoporate	Perprolate	Monoporate	Prominent	Fine gemmate
6	Cynadon dactylon	Medium	Monoporate	Subprolate	Monoporate	-	Scabrate-verrucate
7	Dactyloctenium aegyptium	Medium	Monoporate	Prolate	Monoporate	Sunken	Scabrate, psilate
8	Dichanthium annulatum	Medium	Monoporate	Prolate	Monoporate	Prominent	Gemmate-verrucate
9	Eleusine indica	Medium	Monoporate	Subprolate	Monoporate	Prominent	Sparsely Perforate, psilate and
							scabrate
10	Pennisetum glaucum	Medium	Monoporate	Perprolate	Monoporate	Sunken	Echinate Perforate, scabrate,
							sparsely reticulate and
							verrucate
11	Pennisetum typhoides	Medium	Monoporate	Perprolate	Monoporate	Prominent	Granulate to perforate tectate
12	Poa annua	Medium	Monoporate	Perprolate	Monoporate	-	Scabrate-reugulate
13	Saccharum spontaneum	Medium	Monoporate	Prolate	Monoporate	Sunken	Fine gemmate
14	Steria viridis	Medium	Monoporate	Prolate	Monoporate	Prominent	Gemmate-scabrate
15	Zea mays	Large	Monoporate	Prolate	Monoporate	-	Scabrate, psilate

 Table 10: Qualitative characters of some species of family Poaceae.

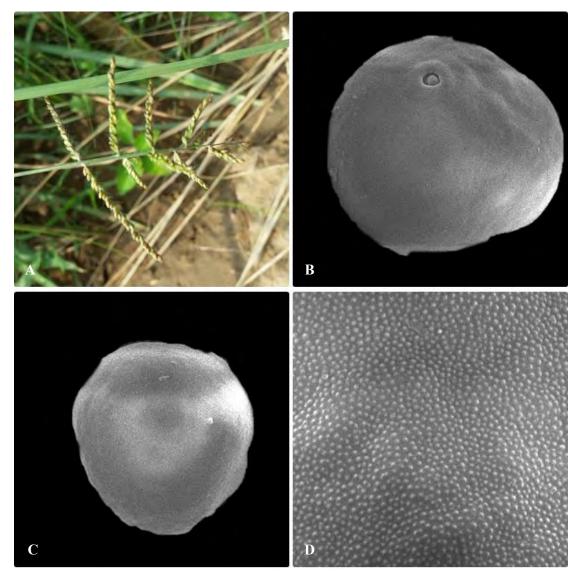
Sr.	Plant Name	Exine Thickness	Polar Diameter	<b>Equatorial Diameter</b>	Pores Length	Pores Width
No		Mean (Min-Max) SE	Mean (Min-Max) SE	Mean (Min-Max) SE	Mean (Min-Max) SE	Mean (Min-Max) SE
		(μm)	(μm)	(μm)	(μm)	(μm)
1	Brachiaria brizantha	1.25 (1.00-1.50) ±0.79	$40.45~(36.50\text{-}45.00)\pm1.43$	40.50 (37.25-43.00) ±1.2	4.20 (3.50-5.00) ±0.27	2.700 (2.25-3.50) ±0.21
2	Cenchrus biflorus	1.50 (1.25-1.75) ±0.11	32.95 (28.25-37.75) ±1.73	32.35 (27.00-40.75) ±2.32	4.45 (3.25-5.75) ±0.46	3.35 (2.75-4.25) ±0.26
3	Cenchrus ciliaris	1.30 (0.75-1.75) ±0.17	63.25 (57.75-71.75) ±2.36	57.25 (53.50-60.00) ±2.36	4.1 (3.25-4.75) ±0.27	3.30 (1.75-4.75) ±0.49
4	Cenchrus pennisetiformis	1.40 (1.00-1.75) ±0.13	55.50 (45.50-65.75) ±3.55	44.74 (33.25-54.25) ±3.63	5.55 (4.00-8.00) ±0.69	3.80 (3.00-5.00) ±0.40
5	Cymbopogon jwarancusa	2.70 (1.00-4.50) ±0.73	37.35 (30.00-41.75) ±2.08	34.90 (29.25 -38.00) ±1.63	4.10 (3.50-5.00) ±0.26	3.00 (2.25-4.00) ±0.30
6	Cynadon dactylon	1.20 (1.00-1.50) ±0.09	37.20 (33.50-41.75) ±1.50	38.90 (35.00-41.75) ±1.50	5.50 (4.25-7.75) ±0.63	3.100 (2.25-3.75) ±0.27
7	Dactyloctenium aegyptium	1.50 (1.00-2.00) ±0.18	39.95 (32.00-47.50) ±2.83	37.00 (32.75-40.00) ±1.35	-	-
8	Dichanthium annulatum	1.45 (1.25-1.75) ±0.93	41.70 (36.25-46.00) ±1.79	40.75 (38.00-42.50) ±0.87	4.75 (3.75-6.00) ±0.93	3.00 (2.25-3.75) ±0.28
9	Eleusine indica	1.20 (0.75-1.75) ±0.16	34.05 (30.75-38.75) ±1.43	32.70 (27.25-37.75) ±1.86	4.20 (3.75-4.75) ±0.20	3.30 (2.25-4.25) ±0.34
10	Pennisetum glaucum	2.20 (1.75-3.00) ±0.24	34.60 (31.75-38.75) ±1.38	37.00 (33.75-40.50) ±2.00	3.85 (2.50-4.50) ±0.38	2.75 (2.25-3.25) ±0.18
11	Pennisetum typhoides	2.30 (1.75-3.00) ±0.21	29.25 (25.25-32.00) ±1.13	32.00 (28.25-37.00) ±1.53	5.95 (4.75-7.50) ±0.50	3.70 (3.00-5.00) ±0.35
12	Poa annua	2.35 (0.75-3.75) ±0.58	30.05 (26.75-37.75) ±2.04	34.55 (29.75-37.74) ±1.46	3.90 (3.25-4.75) ±0.27	2.15 (1.25-3.00) ±0.30
13	Saccharum spontaneum	1.90 (1.25-2.50) ±0.23	37.10 (33.50-46.00) ±2.29	39.15 (32.50-45.00) ±2.49	$7.00(5.25-8.50)\pm0.54$	4.05 (3.00-5.00) ±0.40
14	Steria viridis	1.60 (1.25-2.00) ±0.13	34.65 (30.75-40.00) ±1.75	34.40 (29.25-39.75) ±2.13	5.10 (4.00-6.00) ±0.41	3.55 (2.50-4.50) ±0.34
15	Zea mays	1.35 (1.00-1.75) ±0.13	66.65 (61.00-70.00) ±1.59	62.00 (59.00-65.00) ±1.08	-	-

 Table 11: Quantitative pollen morphological characters of some species of family Poaceae

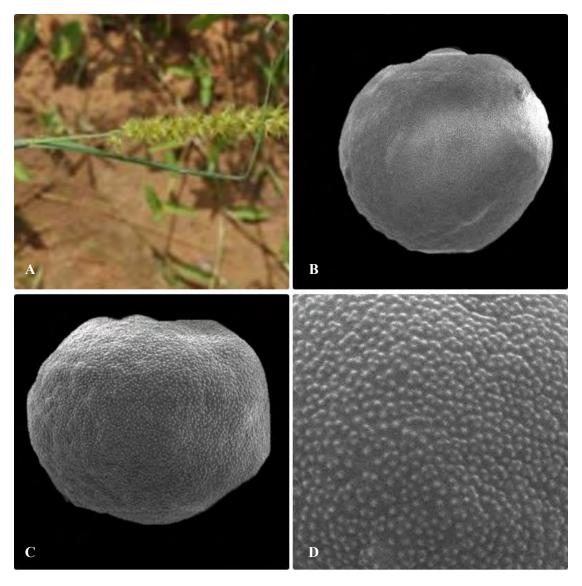
Sr.	Plant Name	No. of fertile	No. of sterile	Fertility	Sterility percentage	
No.		Pollen	pollen	percentage		
1	Brachiaria brizantha	377	66	85.10	14.89	
2	Cenchrus biflorus	407	83	83.06	16.94	
3	Cenchrus ciliaris	289	67	81.18	18.82	
4	Cenchrus pennisetiformis	315	88	78.16	21.84	
5	Cymbopogon jwarancusa	430	62	87.39	12.60	
6	Cynadon dactylon	373	58	86.54	13.46	
7	Dactyloctenium aegyptium	260	52	83.33	16.67	
8	Dichanthium annulatum	363	66	84.61	15.38	
9	Eleusine indica	383	92	80.63	19.37	
10	Pennisetum glaucum	422	69	85.94	14.06	
11	Pennisetum typhoides	283	41	87.34	12.65	
12	Poa annua	377	61	86.07	13.92	
13	Saccharum spontaneum	285	51	84.82	15.18	
14	Steria viridis	636	123	83.79	16.21	
15	Zea mays	283	50	84.98	15.01	

Table 12: Fertility and Sterility percentage of selected species of Poaceae family





**Plate 89:** *Brachiaria brizantha* (A) Floral Parts, Scanning Electron Micrographs of Pollen (B) Prominent Pore Orientation (C) Polar View (D) Surface Sculpturing



**Plate 90:** *Cenchrus biflorus* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Surface Sculpturing

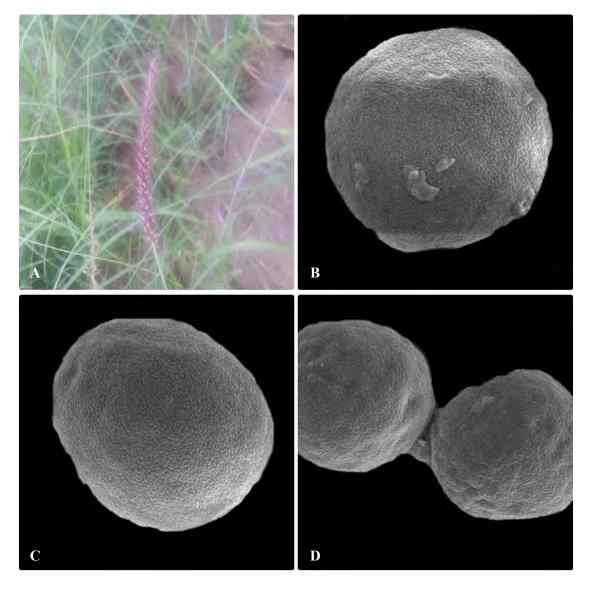
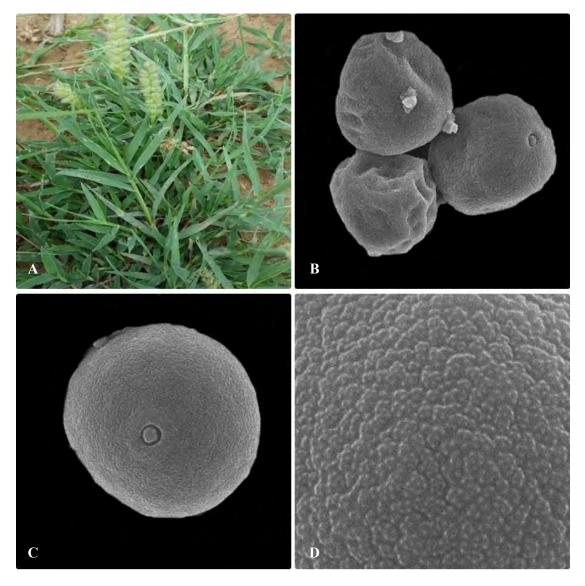
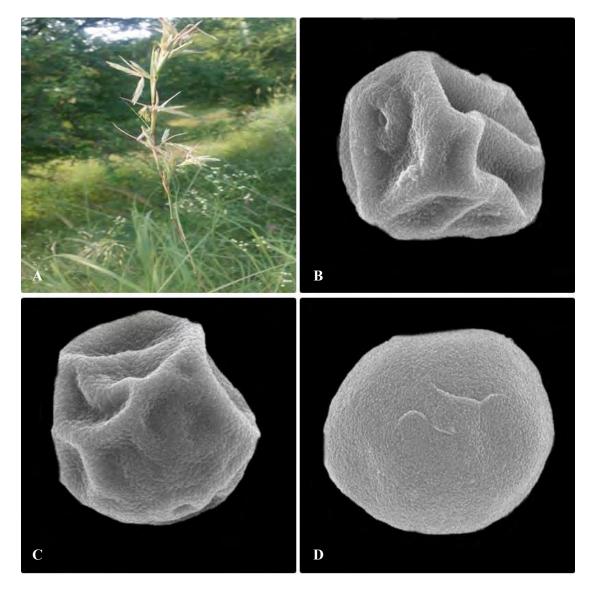


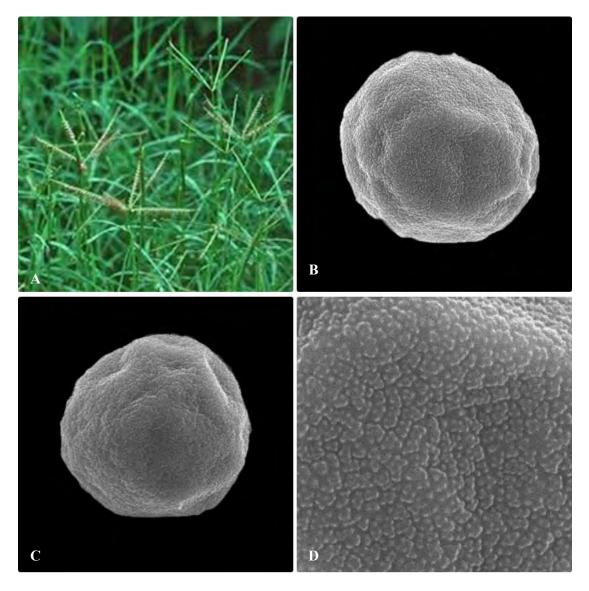
Plate 91: Cenchrus ciliaris (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Slightly sunken Pore orientation (D) Scabrate Exine Sculpturing



**Plate 92:** *Cenchrus pennisetiformis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Oriented Pore orientation (D) Surface Sculpturing



**Plate 93:** *Cymbopogon jwarancusa* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore Orientation (D) Fine gemmate Exine Sculpturing



**Plate 94:** *Cynadon dactylon* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Scabrate-verrucate Exine Sculpturing (D) Surface Sculpturing

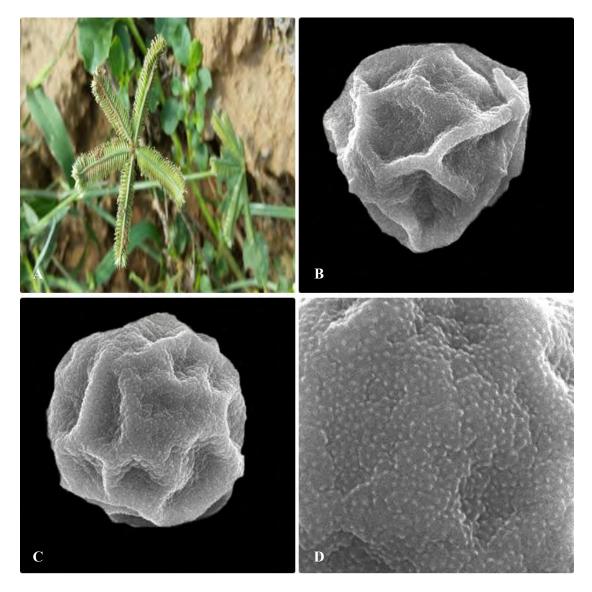
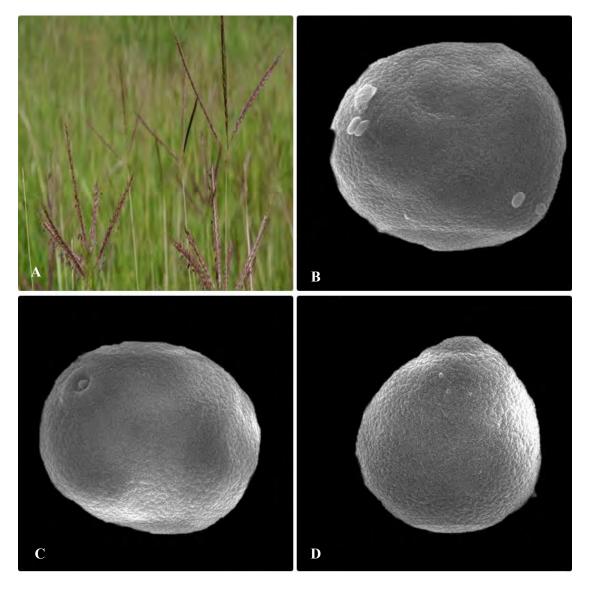


Plate 95: *Dactyloctenium aegyptium* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Surface Sculpturing



**Plate 96:** *Dichanthium annulatum* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Prominent Pore orientation (D) Gemmate–verrucate Exine Sculpturing

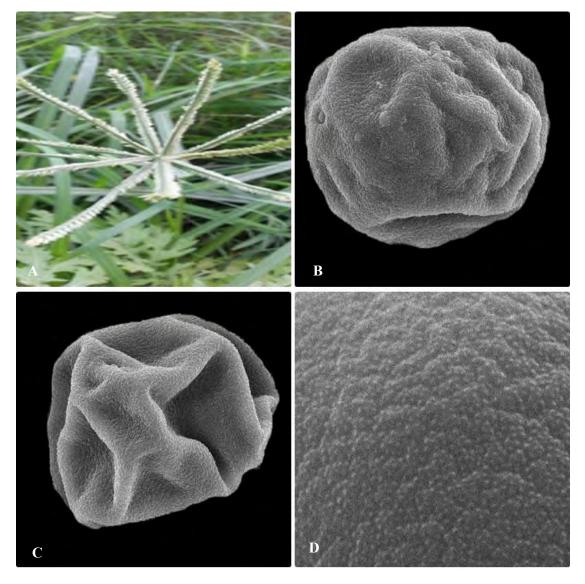
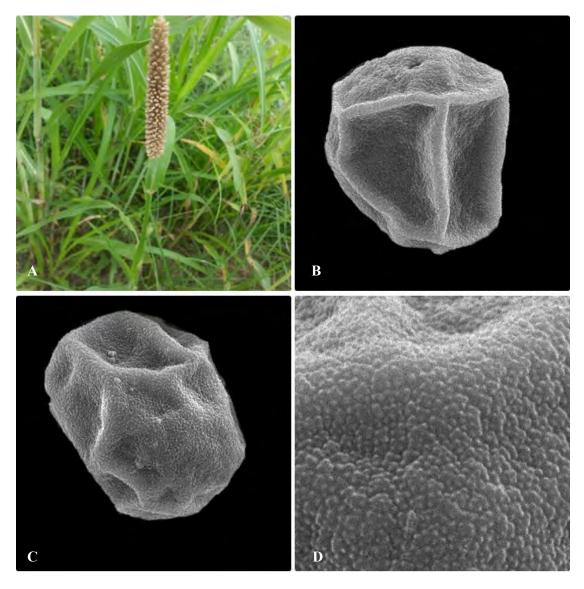
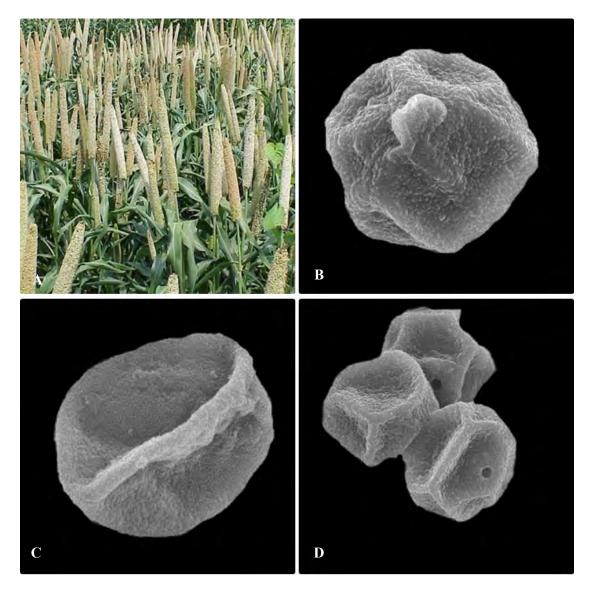


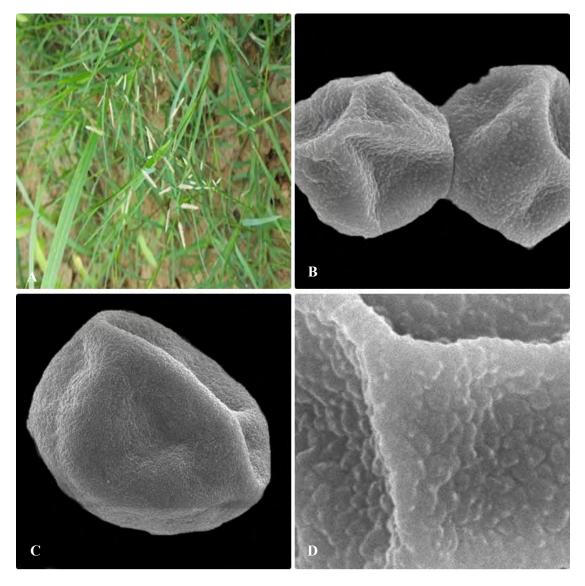
Plate 97: *Eleusine indica* (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Psilate-Scabrate Exine Sculpturing (D) Surface Sculpturing



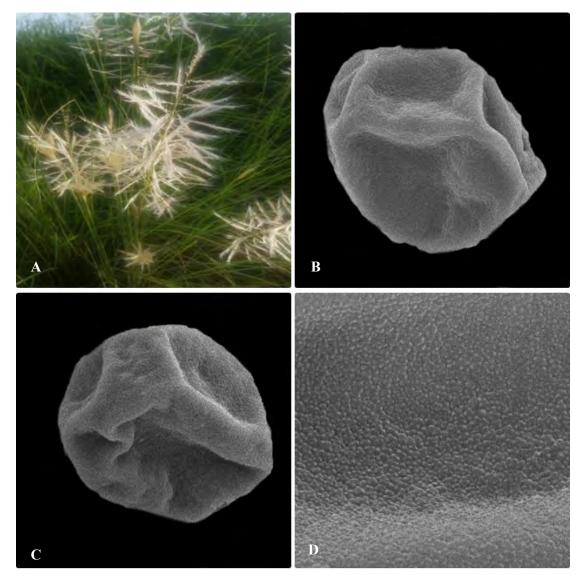
**Plate 98:** *Pennisetum glaucum* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing



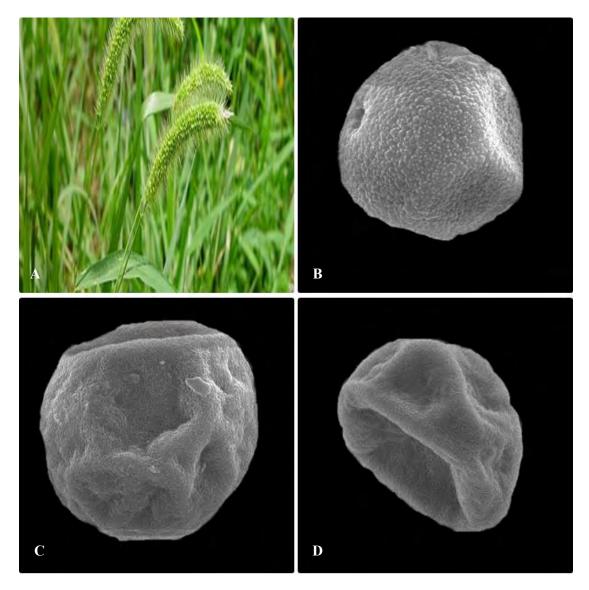
**Plate 99:** *Pennisetum typhoides* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Granulate to perforate tectate Exine Sculpturing



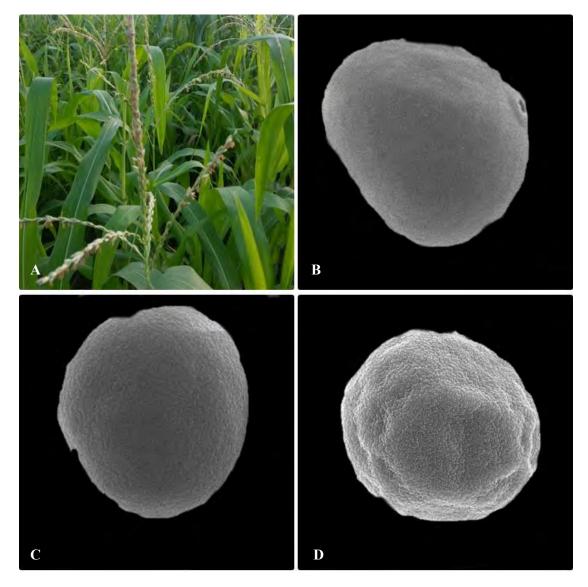
**Plate 100:** *Poa annua* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing



**Plate 101:** *Saccharum spontaneum* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken Pore orientation (D) Surface Sculpturing



**Plate 102:** *Steria viridis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Gemmate–scabrate Exine Sculpturing (D) Equatorial View



**Plate 103:** Zea mays (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Prominent Pore Orientation (C) Polar View (D) Scabrate, psilate Surface Sculpturing

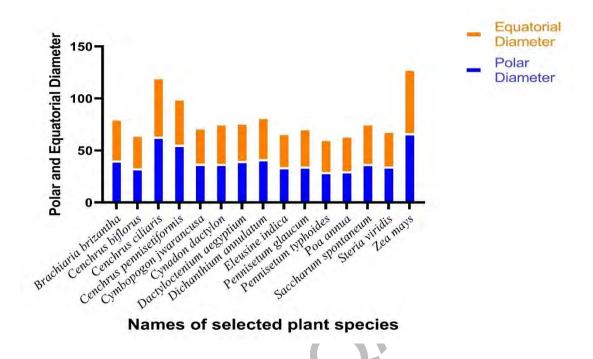


Figure 20: Mean values of Pollen diameter among Poaceous taxa

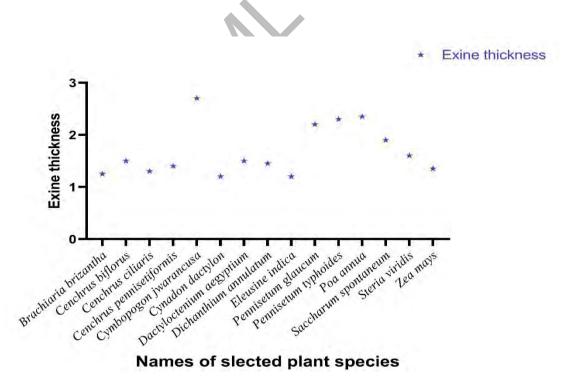


Figure 21: Mean exine thickness in Poaceous Species.

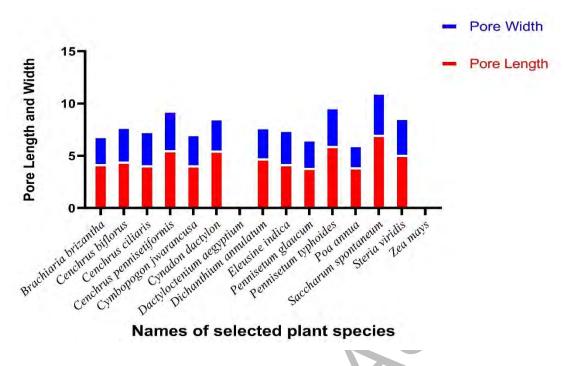


Figure 22: Data variations of pore length and width in Poaceous species

## **Taxonomic key based on Poaceous Pollen characters** 2a. Pollen shape sub-prolate...... Cenchrus ciliaris 3b.Exine ornamentation gemmate-verrucate ...... Cenchrus pennisetiformis 4a. Polar diameter lies in range above 41 m..... Dichanthium annulatum 4b.Polar diameter lies in range below 41m ...... 5 5a. Fertile pollen more than 500 in number ...... Steria viridis 6a. Exine sculpturing is echinate ......Pennisetum glaucum 7a. Fertility percentage is 80% ..... Eleusine indica 8a. Pore width is above 4 m ..... Saccharum spontaneum Exine has fine gemmate ornamentation ..... Cymbopogon 9a. jwarancusa 10a. Exine sculpture is granulate ...... Pennisetum typhoides 10b. Exine sculpture is scabrate ...... 11

11a.	Pollen	shape	prolate			•••••			Dac	tyloctenium
aegyptii	um									
11b. Po	llen shap	oe is sub	-prolate	and per	polate		•••••		12	
12a.Pol	len shap	e is prol	ate			•••••		. Cenc	hrus l	biflorus
12b.Po	llen shap	oe is sub	-prolate		•••••			•••••		. 13
13a. E brizanth		ulpturin	g is sc	abrate	perforat	e				Brachiaria
13b. Ex	ine sculp	pturing i	is scabrat	e verru	cate	••••		Cj	vnado	n dactylon

## **3.1.4 Poaceaous Species**

The pollen grains of the Poaceae generally present variations in shape, size and type, and exine ornamentation, as portrayed in this study. The pollen morphological characteristics of 15 species of grasses provide morphological and statistical data for identification of Poaceous species according to the shape, size, aperture(s) and sculpture of the pollen. Identifying grasses pollen to the species level with scanning microscopy is very helpful due to the greater variations of micromorphological features such as single pores and the high similarity of the sculpture patterns in most of the genera. The stenopalynous form having monoporate form of pollen offers an analytical feature for grasses, while trying to distinguish taxa within family this feature creates a limiting factor via morphology of pollen and sculpturing of exine (Chaturvedi et al. 1998). Whereas studying taxonomy of plants pollen morphology has been considered as the most significant tool. In previous studied pollen morphology, it was considered as very hard to categorize grasses only based on pollen features in the past. A combination of SEM and image analyses was successfully used to quantitatively classify the pollen size and density of exine sculptural elements and to measure the ornamentation complexity of 15 grass species.

Morphological characters of pollen can be valuable in solving complications linked with systematics study of grass and study on pollen provides the base for more structures to proper identify the plant species (Aftab and Parveen, 2006). Earlier morphological characters of Poaceae pollen have been studied by Chaturvedi et al., (1994, 1998), Kohler and Lange (1979) and Ma et al., (2001). In the current article quantitative and qualitative features of pollen grains were reported, as features of pollen like size and pattern of sexine are of great taxonomic importance (Woodehouse, 1935). Firbas (1937) in his work reported the size of pollen and utilize this feature for separating cultivated and wild grass species. [1] investigated morphological characters of pollen of *Cenhrus* genus and detected shapes of pollen grains in *C. pennisetiformis* to be spherical to per-oblate and in *C. setigerus* spheroidal to subulate. The position of pore was either ectoporus or endoporus. In this study 3 species such as *C. pennisetiformis, C. ciliaris* and *C. biflorus*, look like endoporus whereas *C. setigerus* is either ectoporous or endoporus. The results of current research are similar to the previous work on this genus. Variations were also

observed in the shapes of pollen as they were semi-circular or circular while in equatorial sight, the pollen of C. ciliaris are spheroidal in shape and C. biflorus pollen are prolate-spheroidal in appearance [2]. Parveen (2006) reported areolate type of exine ornamentation in Cynodon dactylon while the sculpturing of Cynodon dactylon in present study is scabrate. Ozeler et al., (2009) described pollen grains of Poaceae to be monoporate with scabrate exine sculpturing, size pollen and other characters, such as size of operculum, annulus are key features for identifying the taxa. Chaturvedi (1971) has observed monoporate grains in Saccharum which is similar to the findings of current study. In present study Dichanthium annulatum shows Gemmate-verrucate type of exine sculpturing was accordance with the findings of Ullah et al., (2018), while Zahoor et al. (2019) and Sardar et al. (2013) reported regulate and areolate scabrate type of sculpturing. While previous work of Ghosh and Karmaker (2017) described monoporate, operculate with micro-reticulated type pollen in D. annulatum show dissimilarity with our study. Pollen grains are generally monad in this family, shape of pollen in its polar view is circular, prolate to spheroidal or sometimes oblate to spheroidal, aperture comprises of pores, having operculum and annulus (Corrêa et al. 2005).

Moore et al. (1991) considered that the line of evolution in pollen class as from inaperturate pollen (without aperture) vs aperturate pollen and the latter is from monoaperturate to polyaperturate. Tectum present in the pollen of some members of Poaceae family is either aerolate or scabrate (Parveen, 2004). While same type of tectum has been observed in some members of Poaceae by Faegri and Iversen, (1964) and Andresen and Bertelsen, (1972).

Neha and Kalkar, (2010) earlier results elaborate 1-porate, annulate and brevicerebro ornate sculpture type grains in *P. glaucum* was not according to this research study. Significant difference was reported in *P. typhoides* exine sculpture, thickness, shape, and size of the grains in the earlier study (Ahmad et al. 2020). Inceoğlu and Karamustafa, (1977) observed pollen grains of some members of family Poaceae under light microscope and reported the presence of granulate exine ornamentation while study under scanning electron microscope revealed reticulate exine sculpturing.

The shape of pollen grains were spheroidal or rounded in polar view and in equatorial view shape of pollen were oblate, prolate or spheroidal (Khan et al., 2017). The result of current findings is similar as in equatorial view the shape of pollen is prolate, sub-prolate and polar view has circular pollen grains. Pollen grains are in the form of monad with most species having scabrate exine ornamentation similar findings are reported by Khan et al. (2017). In the pollen grains of P. annua monad, medium size, radial symmetry, heteropolar, circular, spheroidal, circular pore with annulus, exina tectate with micro-echinate ornamentation was examined previously by Radaeski et al. (2016). The grains of P. annua, we observed sizes slightly larger and examination of pollen sculpture are similar to those reported by Khan et al. (2017). Based on the value of polar to equatorial ratio Poa annua is considered to be the smallest size pollen while the study conducted by Meo (1999) documented the diameter at polar axis of P. annua is 24.13 µm and diameter at equatorial axis as 24.88 µm which is somehow similar to the findings of the present study. Monoporate type of pollen are found by Khan et al. (2017) which is same as reported in present study. In current study majority of plant species exhibit medium sized pollen grains except Zea mays which are similar to the findings of Morgado et al. (2015). Eleusine indica contained sub-prolate shape of pollen which is similar to the results of Corrêa et al. (2005) while small pollen and areolate micro-echinate sculpture was reported in E. indica by Morgado et al. (2015) was different from current findings. Whereas Begumand Mandal, (2016) examined granulate and oblate-spheroidal grains in E. indica somewhat differ from this research outcomes. Melhem et al. (1983), Salgado-Labouriau and Rinaldi (1990) and Salgado-Labouriau et al. (1990), reported in their studies pollen of family Poaceae with shape prolate to spheroidal and oblate to spheroidal.

Meo et al. (1989) investigated morphological characters of pollen of *Cenhrus* genus and detected shapes of pollen grains in *C. pennisetiformis* to be spherical to perbolate and in *C. setigerus* spheroidal to subulate. The position of pore was either ectoporus or endoporus. 3 species such as *C. pennisetiformis, C. ciliaris* and *C. biflorus*, look like endoporus whereas *C. setigerus* is either ectoporous or endoporus. The results of current research are similar to the previous work on this genus. Variations are also observed in the shapes of pollen as they were semi-circular or

circular while in equatorial sight, the pollen of *C. ciliaris* are spheroidal in shape and *C. biflorus* pollen are prolate-spheroidal in appearance (Shaheen et al., 2011).

Chaturvedi and Datta, (2001) studied pollen morphology of *Saccharum spontanum* and documented the presence of porate type pollen and insulae ornamentation of exine. However, thick psilate nature of exine was described by El-Amier (2015) in *S. spontanum* while our results revealed fine gemmate sculpturing. Based on microscopic techniques it has been cleared that most of the grasses pollen are monoporate, psilate with sunken pore orientation and this observation of exine sculpturing of *S. spontanum* is fine gemmate, but this result is dissimilar to the results of (Ullah et al. 2021). are similar to Ullah et al. (2021). The findings of present research are different as the pollen grains are monoporate and the exine ornamentation is fine gemmate. Previously Shaheen et al. (2011) examined angular grains in *S. viridis* and exine thickness 0.65  $\mu$ m while in this study gemmate scabrate sculpture elements as described. Yang et al. (2018) suggested that grains of *Z. mays* was spherical or ovoid and large sized, pore circular, sculpture psilate and slightly wrinkled while in present findings scabrated psilate ornamentation was found.

Fertility and sterility percentages in pollen grains gave indication about the relationship among the ancestors of species. Fertility of pollen grains helps in identifying the genetic variations among the flora (Khan, 1991). It is a useful tool to calculate the strength of species in a specific region. Data on sterile and fertile pollen and fertility levels are presented earlier by Bano et al. (2013) indicates that plant species are well established in given climatic conditions. Ullah et al. (2021) recently estimated pollen fertility and sterility for the development of anthers, is essential for process such as flowering, production of more pollen is linked to the pollen allergy. According to Khan et al. (2017) the fertility percentage of *Poa annua* pollen is highest (92.02%), while in present study *Poa annua* has fertility and sterility rate was determined for the pollen of study area which shows the stability of species in any area. The highest fertility range was determined for *Cymbopogon jwarancusa* in the present study.

The results of the studied of this family are compared with previously published work on these species which reveals that these species are quite similar to the previously reported species, with some differences. Both quantitative and qualitative characters observed through light and scanning electron microscopy revealed important information that may help in identifying the grass family members using advance microscopic techniques. Morphological study of pollen study helps in correct identification of the species.

Sr. No	Taxa	Shape	Aperture Condition	Mesocolpium	Exine ornamentation	Colpi orientation	
1.	Anagallis arvensis L	Oblate-spheroidal	Tricolporate	Scabrate	Scabrate	Sunken, angular	
2.	Asphodelus tenuifolius Cav.	Prolate-spheroidal	Tricolporate	Aerolate	Reticulate	Sunken, angular, margins distinct and ends tapering	
3.	Boerhivia procumbense	Prolate-spheroidal	Trizonocolporate	Reticulate	Scabrate- reticulate	Prominent and rounded at ends	
4.	Brachychiton acerifolius (A.Cunn. ex G.Don) F.Muell	Oblate-spheroidal	Tricolporate	Scabrate- reticulate	Aerolate	Sunken, slit like margins and end tapering	
5.	<i>Broussonetia papyrifera</i> (L.) L'Hér. ex Vent.	oblate	Tricolporate	Scabrate	Reticulate- Perforate	Prominent and rounded at ends	
6.	<i>Bryophyllum pinnatum</i> (Lam.) Oken	oblate-spheroidal	Tricolporate	Reticulate	Scabrate	Sunken, angular, margins distinct	
7.	Capparis decidua (Forssk.)edgeW.	Spherical	Tricolporate	Scabrate	Echinate	Sunken, further divided in three slit like portions, ends are tapering	
8.	Chorchorus depressus	oblate-spheroidal	5	Aerolate	Faveolate	Sunken, slit like, margins wavy and slightly pointed at ends	
9.	Cissampelos pareira L.	oblate-spheroidal	Tricolporate	Perforate	Scabrate	Sunken, slit like, margins distinct and pointed at ends	
10.	Citharexylum spinosum L.	Prolate-spheroidal	-	Scabrate	Reticulate	Prominent margins	
11.	<i>Cleome brachycarpa</i> (Forssk.) Vahl ex DC	oblate-spheroidal	Tricolporate	Reticulate	Aerolate	Sunken, long & slit like	
12.	Cleome viscosa L.	Oblate-spheroidal	Not visible	Scabrate	Reticulate- scabrate	Prominent and tapering ends	
13.	Commelina benghalensis L.	Sub-oblate	Tricolporate	Reticulate	Perforate	Sunken, angular, margins distinct	
14.	Convolvulus arvensis L.	oblate-spheroidal	Tricolporate	Aerolate- Perforate	Scabrate	Long & slit like, tapering ends	
15.	Cucumis melo sub agerestis L.	oblate-spheroidal	Tricolporate	Reticulate	Reticulate	Sunken and tapering ends	

 Table 13: Qualitative data of pollen of weeds members from Potohar region of Pakistan

16.	Datura innoxia Mill.	Prolate-spheroidal	Tricolporate	Scabrate	Scabrate-	Long and slit like margins
					reticulate	
17.	Euphorbia granulata L.	Oblate-spheroidal	Not Visible	Perforate	Perforate	Sunken, slit like, wavy margins
18.	Oxalis corniculata L.	Prolate-spheroidal	-Tricolporate	Scabrate-	Aerolate	Sunken, wavy and margins distinct
				reticulate		
19.	Solanum surattense	Sub-oblate	Tricolpate	Aerolate	Reticulate	Sunken and tapering ends
20.	Trianthema portulastrum	Spheroidal	Not Visible	Scabrate	Perforate	Prominent and tapering ends

NOT VISIBLE Scabrate Perforate

Plant name	Exine thickness Mean (Min-Max) S.E μm	Polar Diameter Mean (Min-Max) S.E μm	Equatorial Diameter Mean (Min-Max) S.E μm	P/E ratio	No of Pores	Colpi Length Mean (Min-Max) S.E µm	Colpi Width Mean (Min-Max) S.E µm	Pores Length Mean (Min-Max) S.E μm	Pores Width Mean (Min-Max) S.E μm
Anagallis arvensis L	1.56(1.35-1.95)	14.5(12.1-15.6)	15.6(12.3-18.0)	0.93	_	3.81(1.65-7.95)	2.55(1.50-4.50)	-	_
	±0.11	$\pm 0.63$	$\pm 0.96$			±3.81	±0.53		
Asphodelus tenuifolius	1.80(1.35-2.55)	18.03(16.8-18.6)	17.8(15.3-19.6)	1.01		6.12(1.65-10.20)	3.45(1.95-7.20)	_	_
Cav.	$\pm 0.23$	±0.34	$\pm .0.84$	1.01		±1.40	$\pm 0.97$		
Boerhivia	1.50(1.20-1.80)	28.8(25.5-33.15)	27.0(25.6-30.3)	1.07		10.05(4.05-17.55)	6.24(2.85-13.20)		
procumbense	$\pm 0.11$	$\pm 1.65$	$\pm 1.2$	1.07	-	±2.17	$\pm 1.92$	-	-
Brachychiton acerifolius (A.Cunn. ex G.Don) F.Muell	6.12(1.65-10.20) ±1.40	3.45(1.95-7.20) ±0.97	6.12(1.6-10.20) ±1.40	0.97	-	12.2(10.0-15.1) 2.44	5.79(3.00-8.55) ±2.31	-	-
<i>Broussonetia</i> <i>papyrifera</i> (L.) L'Hér. ex Vent.	2.01(1.80-2.10) ±0.10	11.7(11.4-11.9) ±0.15	17.3(17.2-17.5) ±0.09	0.67	-	12.30(9.30-15.10) ±1.80	7.20(5.70-9.20) ±1.03	-	-
Bryophyllum pinnatum (Lam.) Oken	$0.6(0.5 - 0.75) \pm 0.06$	13.3(12.2-15.25) ±0.5	13.5(12.2-15.5) ±0.54	0.98	-	$\begin{array}{c} 4.2 (3.2 - 5.00) \\ \pm \ 0.52 \end{array}$	5.4(4.9–6.3) ± 0.45	-	-
Capparis decidua (Forssk.)edgeW.	1.55(1.0-2.0) ±0.16	23.9(22.75-25.5) ±0.52	23.9(22.2-25.5) ±0.72	1.00	-	$5.5(4.6-6.7) \pm 0.62$	$2.9(2.3-3.5) \\ \pm 0.34$	-	-
Chorchorus depressus	$\begin{array}{c} 1.05(0.75\text{-}1.25) \\ \pm 0.09 \end{array}$	13.9(12.0-15.50) ±0.63	13.4(12.0-15.5) ±0.70	0.99	-	$5.2(4.2-6.1) \\ \pm 0.15$	$6.4(5.7-7.3) \pm 0.15$	-	-
Cissampelos pareira L.	2.5(2.25-2.75) ±0.11	21.0(15.5-24.75) ±1.63	22.6(18.0-25.2) ±1.3	0.92	-	7.8(6.8–9.1) ± 0.9	4.1(3.4–5.1) ± 0.4	-	-
Citharexylum spinosum L.	2.65(2.25-3.0) ±0.12	15.3(14.7-15.75) ±10.16	14.8(14.0-15.7) ±0.3	1.03	1	$\begin{array}{c} 4.8 (4.4 - 5.4) \\ \pm \ 0.29 \end{array}$	$2.7(2.4-3.1) \pm 0.2$	7.00(5.25-8.50) ±0.54	4.05(3.00-5.00) ±0.40
Cleome brachycarpa	1.9(1.0-2.75) ±0.34	14.0(13.0-14.7)	14.5(13.5-15.2)	0.96	-	6.7(5.8–8.1)	3.1(2.4-4.00)	-	-

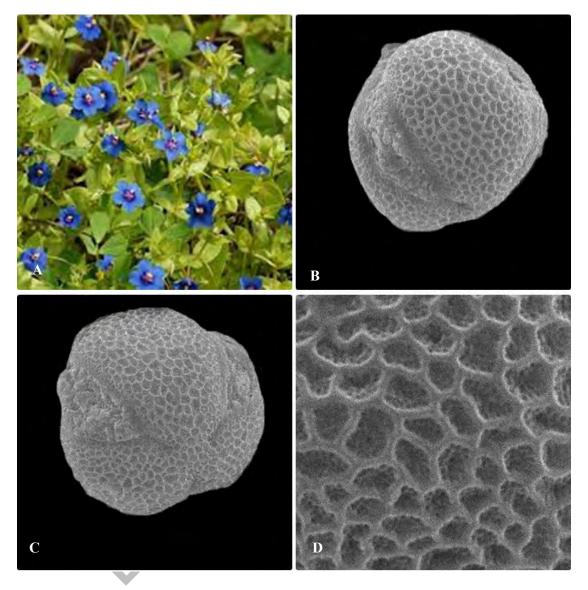
**Table 14:** Quantitative data of pollen of weeds members from Potohar region of Pakistan

(Forssk.) Vahl ex DC		±0.32	$\pm 0.3$			$\pm 0.6$	$\pm 0.4$		
Cleome viscosa L.	2.95(2.75-3.25)	36.9(33.7-43.00)	38.7(37.2-40.5)	0.95	_	7.8(5.8–9.5)	4.3(3.7–5.00)	_	_
	$\pm 0.09$	$\pm 1.74$	$\pm 0.65$	0.95	-	$\pm 1.07$	$\pm 0.3$	-	-
Commelina	2.80(2.25-3.25)	40.4(35.7-45.50)	47.2(45.2-48.5)	0.85		1.86 (1.35–2.40)	1.23 (.75–1.95)		
benghalensis L.	$\pm 0.16$	$\pm 1.68$	$\pm 0.65$	0.85	-	$\pm 0.17$	$\pm 0.21$	-	-
Convolvulus arvensis	0.78(0.6-1.1) ±0.09	27.3(22.7-32.7)	30.0(24.5-33.5)	0.91		1.65 (1.05-2.40)	1.68 (1.05–2.10)		
L.		$\pm 1.65$	$\pm 1.77$	0.91	-	±0.22	$\pm 0.20$	-	-
Cucumis melo sub	2.5(2.25-2.75) ±0.11	34.05(29.0-39.0)	35.1(32.2-39.2)	0.96		2.25 (.75-3.45)	2.64 (2.10-3.30)		
agerestis L.	$2.3(2.23-2.75) \pm 0.11$	$\pm 1.69$	$\pm 1.32$	0.90	-	$\pm 0.47$	$\pm 0.20$	-	-
Datura innoxia Mill.	2.15(1.5-3.0) ±0.3	16.2(13.5-18.0)	15.6(13.2-18.5)	1.03		2.70 (1.95-3.30)	3.03 (2.40-3.60)		
	$2.13(1.3-3.0)\pm0.3$	$\pm 0.85$	$\pm 1.0$	1.03		±0.22	$\pm 0.20$	-	-
Euphorbia granulata	0.95(0.75-1.25)	13.9(12.0-15.5)	13.4(12.0-15.5)	0.99		1.14 (.75–1.50)	1.77 (1.50–2.10)		
L.	$\pm 0.09$	$\pm 0.63$	$\pm 0.70$	0.99	±0.13	±0.13	±0.12	-	-
Oxalis corniculata L.	2.0(1.50-2.25) ±0.13	19.8(17.2-21.25)	18.2(16.2-20.5)	1.08	1				
	$2.0(1.30-2.23)\pm0.13$	$\pm 0.69$	±0.69	1.00	1	-	-		
Solanum surattense	se 1.85(1.7-2.1)±0.7	12.8(10.2-	11.10(10.3-	1.15		4.50(3.25-	6.55(5.25-	_	
	$1.03(1.7-2.1)\pm0.7$	15.5)±0.71	13.50)±0.6	1.15		5.25)±0.38	7.75)±0.46	-	-
Trianthema	2.8(1.7-4.5)±0.46	20.6(12.5-	19.6(18.2-	1.05		7.05 (7.75-	4.75 (5.25-		
portulastrum	2.8(1.7-4.3)±0.40	22.7)±0.57	20.7)±0.40	1.05		6.25)±0.69	4.25)±0.39	-	-
			5						

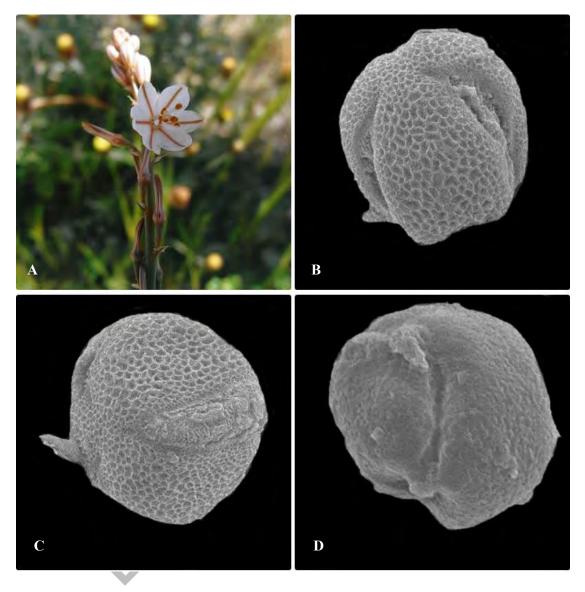
Sr. No	Taxa	No of Fertile Pollen	No of Sterile Pollen	Fertility %
1.	Anagallis arvensis L	70	15	82
2.	Asphodelus tenuifolius Cav.	90	40	69
3.	Boerhivia procumbense	77	10	88
4.	Brachychiton acerifolius (A.Cunn. ex G.Don) F.Muell	99	12	89
5.	Broussonetia papyrifera (L.) L'Hér. ex Vent.	88	22	80
6.	Bryophyllum pinnatum (Lam.) Oken	50	9	84
7.	Capparis decidua (Forssk.)edgeW.	80	2	97
8.	Chorchorus depressus	80	7	91
9.	Cissampelos pareira L.	75	10	88
10	Citharexylum spinosum L.	90	11	89
11	Cleome brachycarpa (Forssk.) Vahl ex DC	118	7	94
12	Cleome viscosa L.	102	10	91
13	Commelina benghalensis L.	90	5	95
14	Convolvulus arvensis L.	110	14	89
15	Cucumis melo sub agerestis L.	86	11	89
16	Datura innoxia Mill.	99	12	89
17	Euphorbia granulata L.	143	9	94
18	Oxalis corniculata L.	81	14	85
19	Solanum surattense	363	66	84.61
20	Trianthema portulastrum	636	123	83.79

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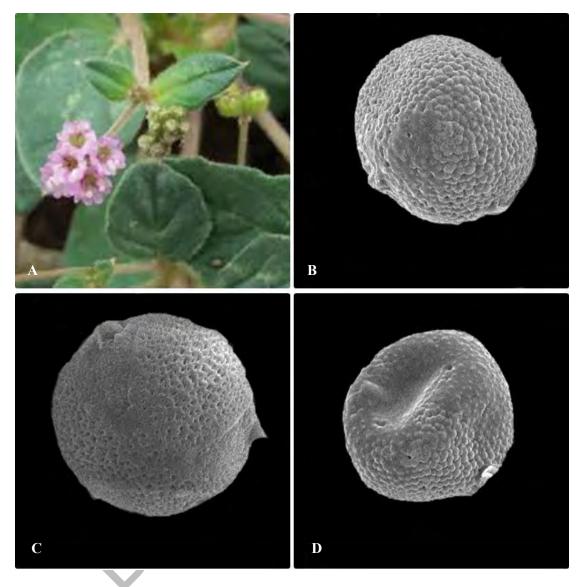
#### Table 15: Pollen Fertility percentages of examined taxa



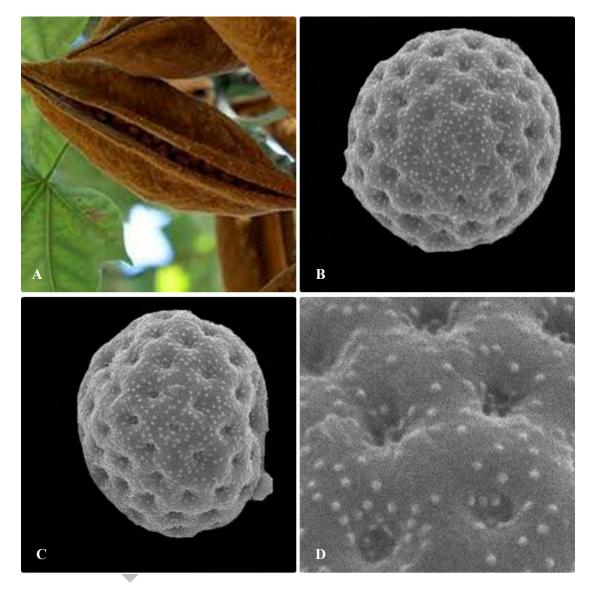
**Plate 104:** *Anagallis arvensis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken orientation (D) Surface Sculpturing



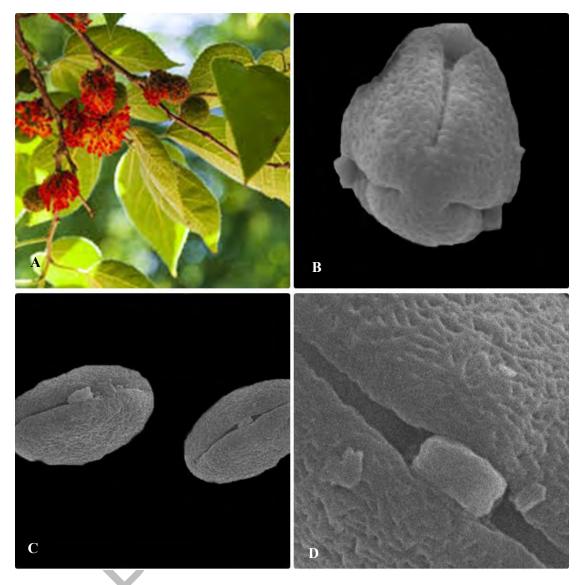
**Plate 105:** *Asphodelus tenuifolius* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Angular orientation (D) Reticulate Exine Sculpturing



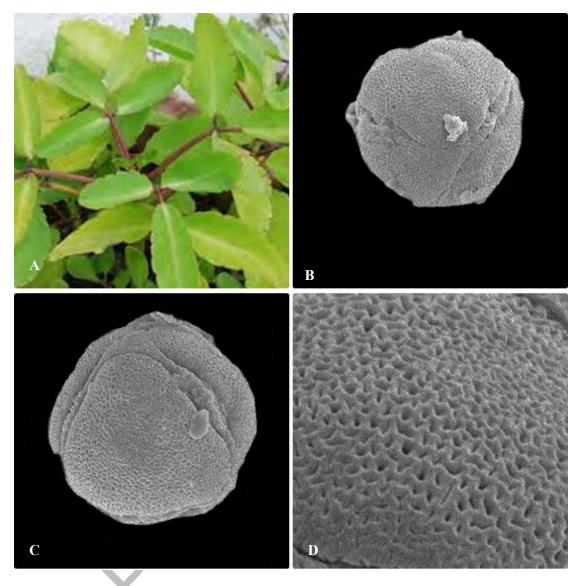
**Plate 106:** *Boerhivia procumbense* (A ) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Rounded orientation (D) Scabrate Exine Sculpturing



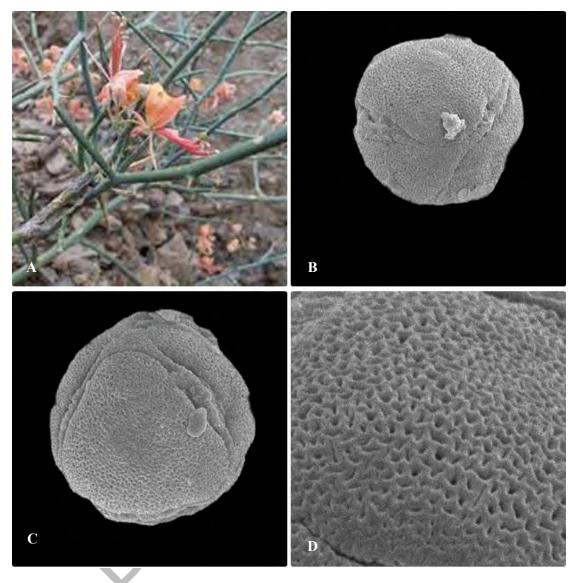
**Plate 107:** *Brachychiton acerifolius* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Perforate orientation (D) Surface Sculpturing



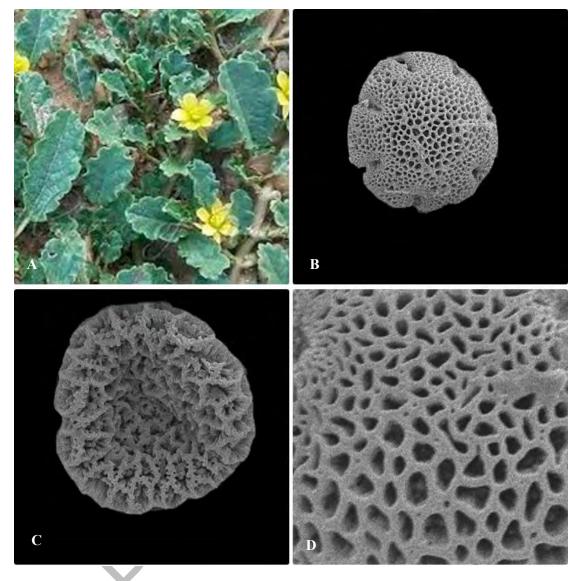
**Plate 108:** *Broussonetia papyrifera* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Surface Sculpturing



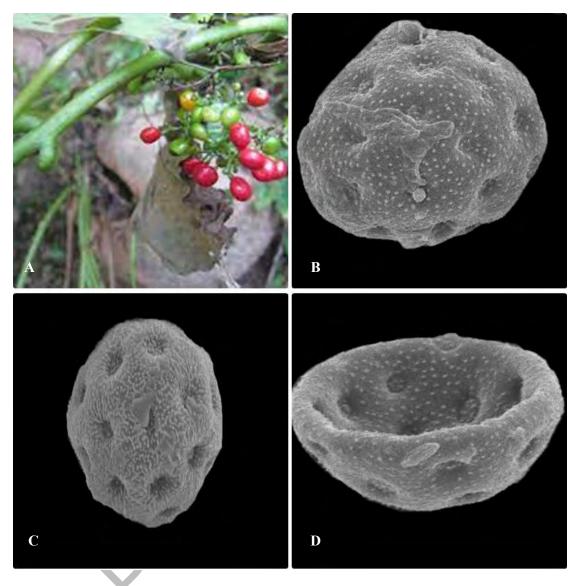
**Plate 109:** *Bryophyllum pinnatum* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken orientation (D) Surface Sculpturing



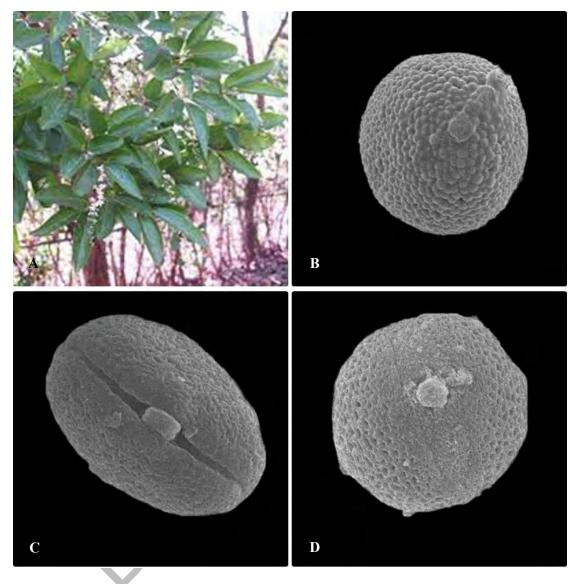
**Plate 110:** *Capparis decidua* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B)Polar View (C) Sunken orientation (D) Surface Sculpturing



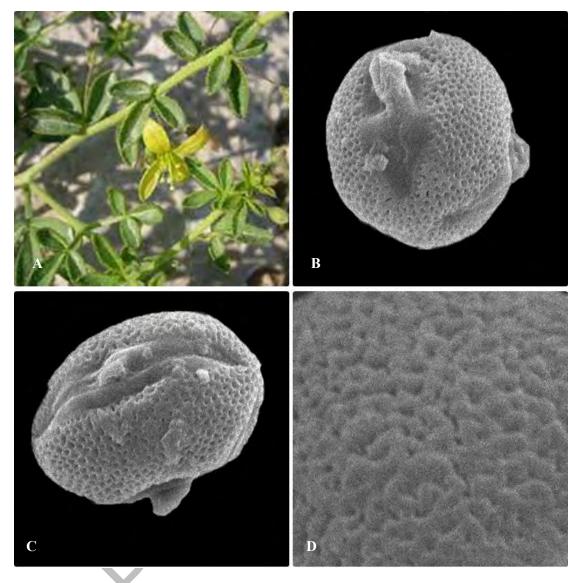
**Plate 111:** *Chorchorus depressus* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Wavy orientation (D) Surface Sculpturing



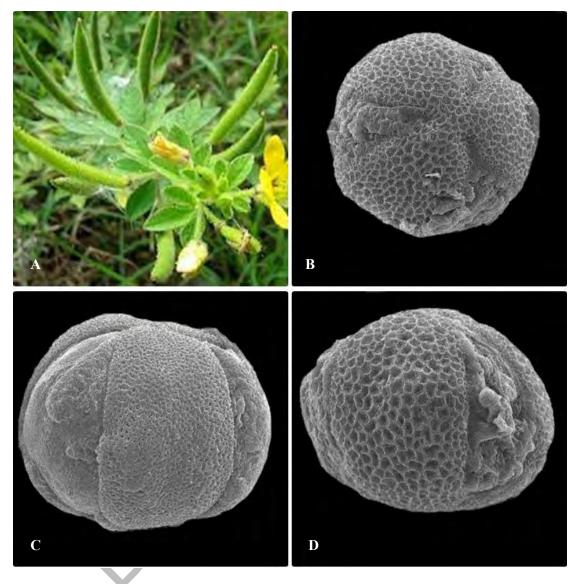
**Plate 112:** *Cissampelos pareira* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken orientation (D) Exine Sculpturing



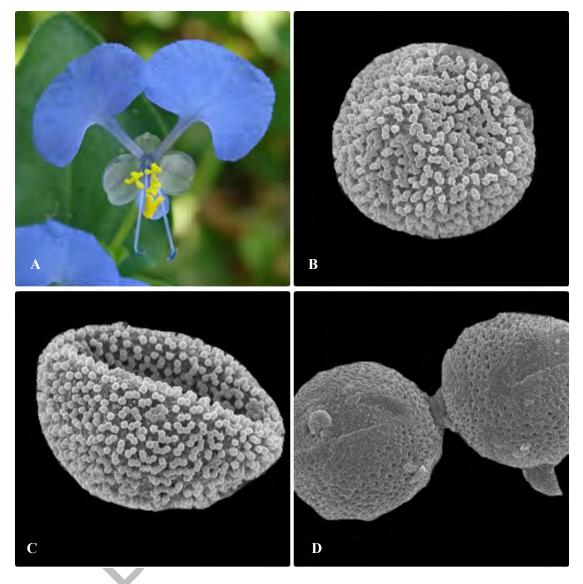
**Plate 113:** *Citharexylum spinosum* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Exine Sculpturing



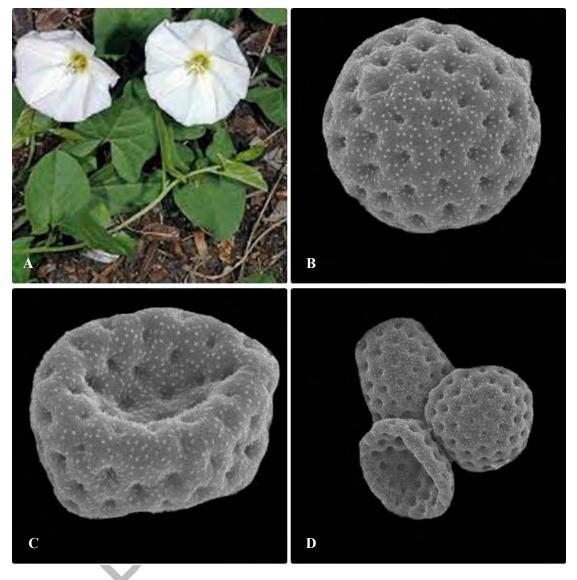
**Plate 114:** *Cleome brachycarpa* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken orientation (D) Surface Sculpturing



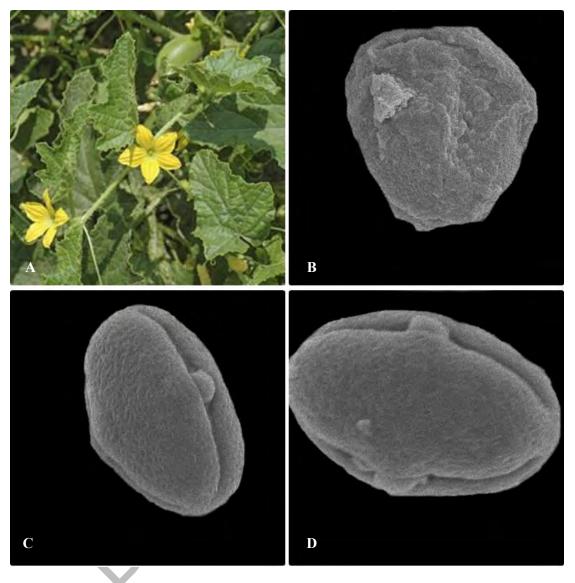
**Plate 115:** *Cleome viscosa* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Reticulate and Scabrate Surface (D) Exine Sculpturing



**Plate 116:** *Commelina benghalensis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken & Angular orientation (D) Exine Sculpturing



**Plate 117:** *Convolvulus arvensis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Scabrate Ornamentation(D) Exine Sculpturing



**Plate 118:** *Cucumis melo* sub *agerestis* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Equatorial View (D) Exine Sculpturing

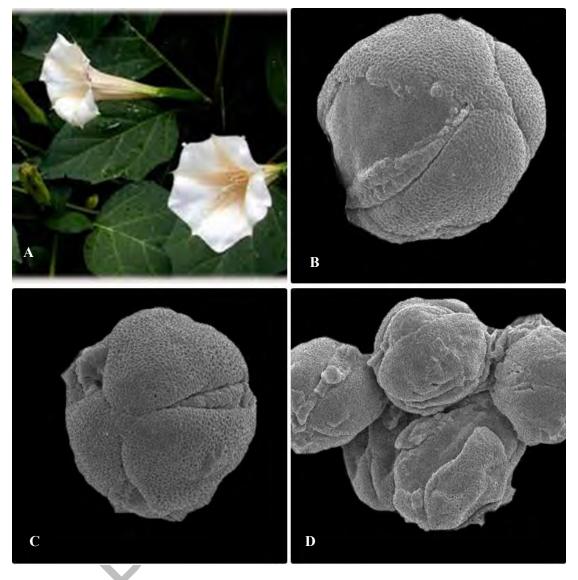
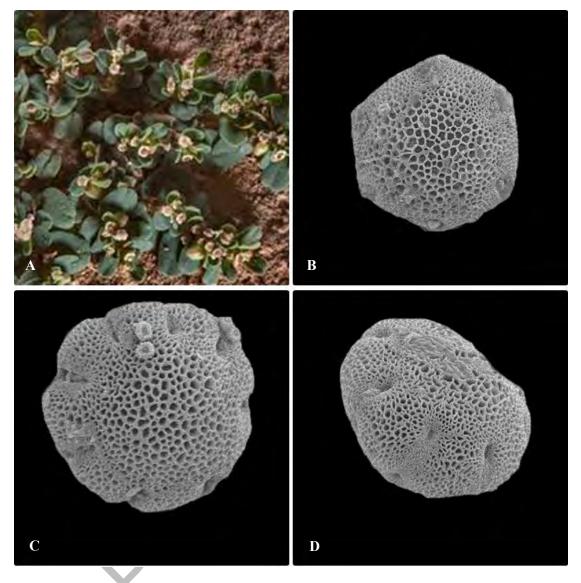


Plate 119: Datura innoxia (A) Floral Parts , Scanning Electron Micrographs of Pollen(B) Polar View (C) Scabrate & Reticulate Ornamentation (D) Echinate ExineSculpturing



**Plate 120:** *Euphorbia granulate* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken orientation (D) Exine Sculpturing

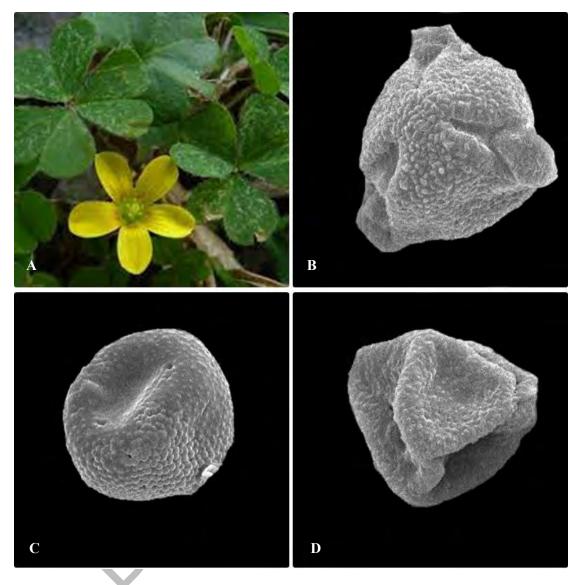
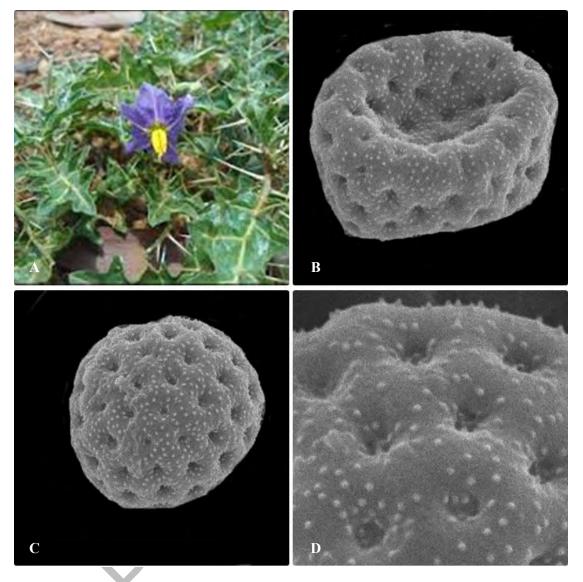
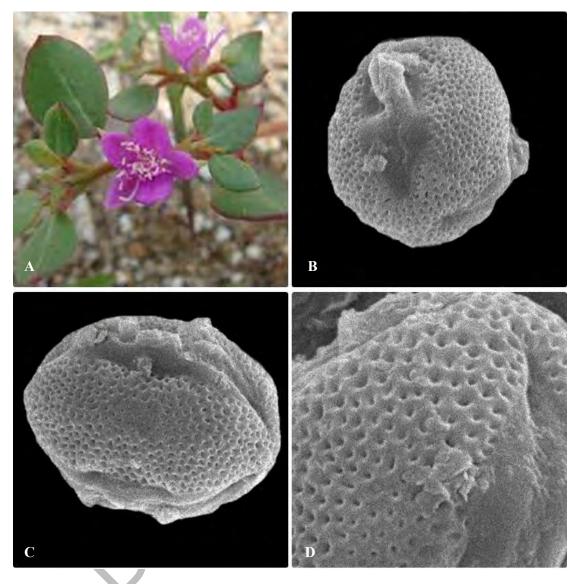


Plate 121: Oxalis corniculata (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Sunken orientation (D) Exine Sculpturing



**Plate 122:** *Solanum surattense* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Reticulate Ornamentation (D) Surface Sculpturing



**Plate 123:** *Trianthema portulastrum* (A) Floral Parts , Scanning Electron Micrographs of Pollen (B) Polar View (C) Perforate Exine Ornamentatin (D) Surface Sculpturing

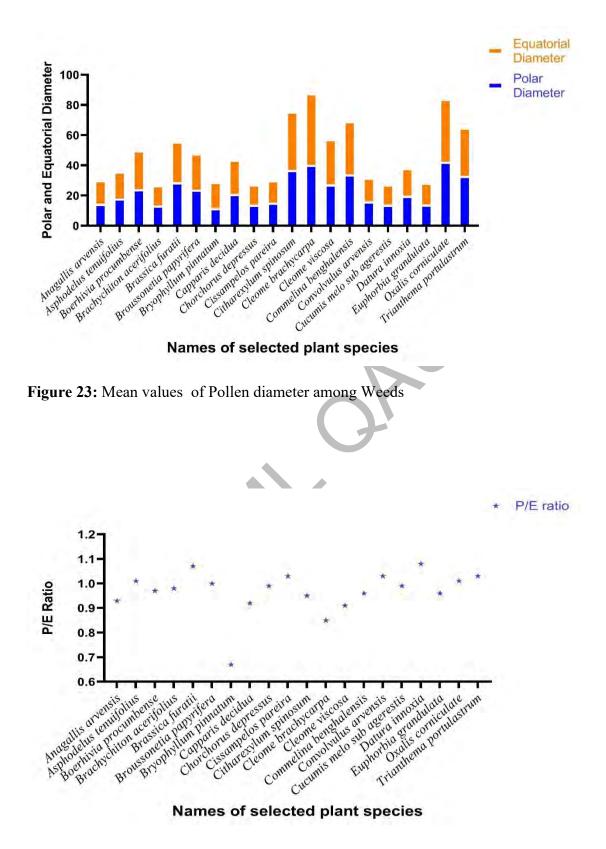


Figure 24: Variation in P/E ratio index of Weeds

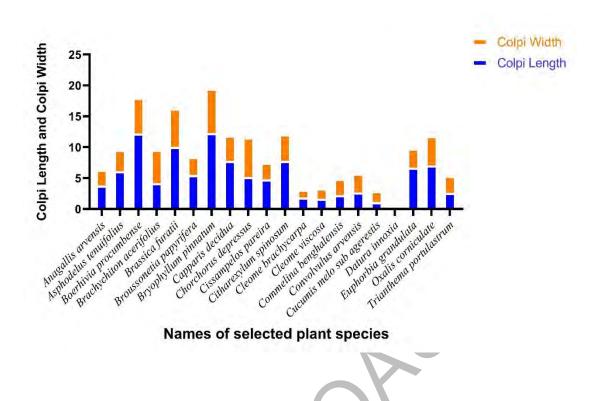


Figure 25: Mean data variations of colpi length and width in Weeds

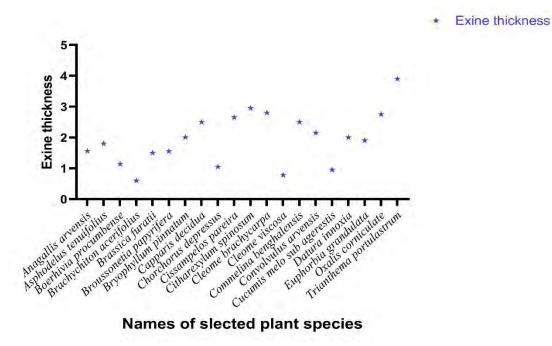


Figure 26: Mean exine thickness in Weeds.

#### 3.1.5 Discussion Of Weeds

The results explained in this part related to weeds demonstrate the role of palynological study for species identifications from the Potohar regions of Pakistan. For the documentation of a detailed description of morphological features of species light and scanning electron microscopic techniques have been used. Pollen grains of the weedy plants vary in shapes, apertures, mesocolpium, colpi orientation, and exine ornamentations. Pollen shape, exine sculpturing, spines, number of colpi, and exine thickness of weeds from district Bannu were described by Ahmad et al.,. A wide variation in different pollen parameters helps to differentiate closely related species and genera. Morphological characters of pollen provide additional knowledge in plant taxonomy and systematics(Ahmad et al. 2019; Amina et al. 2020; Perveen and Qaiser, 2012). Pollen investigated through LM showed great variability in equatorial diameter, polar axis diameter, width and length colpi, and thickness of exine(Aftab and Parveen, 2006). A very wide range of variations was observed in pollen morphology of selected species of weeds from the study area showing that the field of palynology plays a vital role in evolutionary studies of plant species. Variations in exine sculpturing are the diagnostic character in plant systematic study.

The current study showed that pollen morphological features, especially the shape of pollen grains, are the major distinguishing feature for the identification of weeds species. The present study varies from the previous studies conducted on weeds in terms of qualitative and quantitative microscopic characteristics. In the present research family, Amaranthaceae were considered the dominant family of weeds followed by Capparaceae while the rest of each family, i.e., Euphorbiaceae, Cucurbitaceae, Verbenaceae, Menispermaceae, Teliaceae, Chenopodiaceae, Poaceae, Asphodelaceae, Brasicaceae, and Nyctaginaceae contain 1 species each. Pollen grains are in the form of monad with most species having scabrate exine ornamentation similar findings are reported by Nazish et al., examined the apolar, spheroidal, scarbate. Micro Echinate pollen ornamentations and sunken pore ornamentations have a number of pores ranging from 11-23 Butt et al. (2021).

Various researchers working on the palynological study of weeds believed that exine ornamentation is the key feature for the differentiation of taxonomic characters (Aftab and Parveen, 2006). Quite differences were observed in the exine thickness, the diameter of the equatorial and polar axis, the size of the colpus, and the number of pores. Palynological studies are used as a tool for species identifications in plant taxonomy and systematics. The study had links directly with environmental sciences, molecular biology, plant ecology, pharmaceutical studies, genetics, and aerobiology. In our study, some pollen has the same aperture conditions i.e., tricolporate. Khan et al., worked on the pollen morphological features of 16 plants using scanning electron microscopy, whose result is somewhat similar to our results used light and scanning electron microscopy and described the pollen micro-morphology of 24 wetland weeds. Bolick et al., recorded the exine sculpturing patterns of the pollen of Asteraceae with special emphasis on evolution Ahmad et al. (2020).. Morphological characteristics of pollen can be valuable in solving complications linked with the systematic study of grass, and the study of the pollen provides the base for more structures to properly identify the plant species. Palynological study of Asteraceous taxa from the Potohar region helps to differentiate species taxonomically .Research work has been done on many aspects of the flora. The study area has diverse species, but no work is performed on pollen morphological study of weedy species(Khan, 1991).

Oblate-spheroidal observations were observed abundantly in this study. Most of the colpi orientations noted were sunken and angular. Also, many prominent, waxy, long, slit-like, and tapering ends colpi were seen in the pollen. Pollen examined in this study is reticulate in sculpturing, while some contain faveoelate. Cenhrus ciliaris contain a single pore in the center of pollen. Fertility and sterility percentages were determined for the stability of pollen in the study area. Fertility and sterility percentages in pollen grains indicated the relationship among species' ancestors. Pollen grains' fertility helps identify the flora's genetic variations. Meo et al., reported different shapes of pollen grains of the genus Cenchrus. He observed spherical to perbolate shape in C. pennisetiformis and spheroidal to subulate in C. setigerus. The pollen of this genus is mono-porate with psilate exine ornamentation. Position of pores is ectoporous but in some cases endoporus pores are also observed. 3 species i.e. C. ciliaris, C. biflorus and C. pennisetiformis are endoporus whereas C. setigerus is ectoporus. The size and shape of pollen grains of genus *Cenchrus* is used to distinguish among the species. It is a useful tool for calculating the strength of species in a specific region. Pollen examined in this study were oblate, suboblate, oblatespheroidal, and Prolate-spheroidal in shape with aperture conditions of tricolporate, mesocolpium scarbate, aerolate, reticulate, and perforated exine ornamentations. While the colpi orientation noted were long, slit-like, sunken, angular, margins distinct, waxy, rounded, and tapering ends.

Perveen et al., documented the morphological characteristics of pollen of the family Brassicaceae which are having reticulate ornamentation of exine compared to current study results *Brassica furatii* exhibit echinate type of exine surface. (Appel and Al-Shehbaz) reported that all pollen grains observed in their research exhibit tricolpate with the reticulate type of exine which is similar to the present study results as *Brassica furatii* has tricolpate pollen grains. Moore et al., reported tricolpate type of pollen with reticulate sculpturing of the exine. Much work has been done on weeds of the family Brassicaceae by (Moore et al., 2018) and concluded that the shape of the lumen is the most significant character in the delimitation of different species in the family Brassicaceae.

Weed flora has been documented previously by many researchers as reported 73 weed species that are medicinally important and utilized for curing different diseases of both human beings and animals. These weed species are helpful for the manufacturing of different plant yields and also useful for the indigenous sellers of crude drugs of plants. The pattern of distribution, richness, and abundance of weed species (e.g A. viridis, P. lanceolata, D. annulatum, V. thapsus, and C. dactylon) depend on the different environmental conditions such as CaCO<sub>3</sub>, pH, organic matter, soil texture, phosphorous, electrical conductivity and concentration of nitrogen. Rafay et al. (2014) reported that in the agroecosystem weeds play a significant role by giving the environmental heterogeneity and floral diversity increase in the cropping system. Williams and Kremen defined that different types of invertebrates are supported by non-crop plants because many insects for survival depend upon specific weeds. On the other hand, weeds play a serious role in crop production and threaten the cultivated lands because of the use of nutrients from the soil. Weeds badly affect the yield and growth of crop plants because of nutrient demand. Weed vegetation is eliminated by the use of different control measures for example chemical approaches, cultural procedures, and biological mechanisms. In cultural operations, weeds are thrown away from the rice field. Identification of different weed species and farming practices plan with revised management is suggested.

The results of the present study of this family are compared with previously published data showing that these species are quite similar to the previously reported species, with few variations. Butt et al., described the palynological study of weeds for pollen some features, i.e., pore number, the diameter of the equatorial and polar axis, length, width of colpi, and its outline, which are very helpful for the classification of species in plant taxonomy. According to our results, these characteristics are insufficient for species identifications; sometimes, many species hold similar features. The most vital characteristics are exine thickness, mesocolpium, and aperture conditions. The study also provides information about the delimitation of the species from the genus to the family level. Further research is needed to achieve more conclusive results using more cosmopolitan species.



## **SECTION 2**

# Seeds Morphology (LM & SEM)

### **3.2 Seed Morphology of Asteraceous and Fabaceous Plants from Potohar-Plateau Pakistan**

For Plants seeds are means of survival of the species as they carry parent germplasm and protected against heat, cold, drought and water. In current study the macro and micro morphological characters of seeds of Asteraceae and Fabaceae plants of Potohar-Plateau Pakistan, including seed shape, size, surface, seed inner color, position of hilum, compression, length, width and L/W ratio was observed using LM and SEM. Most taxonomists are in support of the idea that macro and micro structure of seeds are very useful for the classification of species and their correct identification. Dahlgren (1979-1980) stated the usefulness of micromorphology and ultrastructural data for evolution and classification of seed plants and play significant role in modern synthetic systems of angiosperms.

Morphological characters of seeds are used in for delimiting the taxa with quite conflicting morphological attributes. In present section morphology of Forty seeds of Asteraceae and Fabaceae Family were studied and data lies in Table 16,17,18 & 19. The Scanning Electron Microscopy (SEM) and Light Microscopy (LM) were applied. The current research study demonstrates the great variation in seed size, shape and ultra micromorphological features. The findings of research study showed that the seed morphological characteristics can be contribute as criteria to distinguish the taxa. Scanning electron microscopy is an innovative method used to study seed ornamentation, seed molding, and evaluation of micromorphological characters of seeds as compared to light microscopy findings (Môro et al., 2001). According to Luqman et al., 2018, seed size and shape along with micromorphological features can be used as diagnostic tools to identify the various plant species. Earlier literature has also proved that seed macro and micromorphological features can be used for correct identification purposes (Karakish et al., 2013)The collected seeds of Asteraceous and Fabaceous plants in present study were examined by using stereomicroscope. Kim et al. (2016) suggested that studies of seeds using stereomicroscope give an aid to facilitate morphological identification of these seeds.

Sr. No	Plant Taxa	Length (mm)	Width (mm)	L/W ratio	No of Ribs	Pappus Length	No of cypsela
1	Ageratum conyzoides	7.56	2.84	2.66	6-7	Absent	25
2	Artemisia absinthimum	2.16	0.82	2.63	12-14	3.36	17
3	Aster squamatus	3.98	0.92	4.32	Absent	1.76	13
4	Biden pilosa	3.52	1.08	3.25	13-15	24.6	14
5	Bidens bipinnata	3.36	0.92	3.65	7	16.96	10
6	Calendula arvensis	6.74	3.14	2.21	9	3.5	37
7	Calendula officinalis	2.04	1.04	1.96	21	4.12	33
8	Carthamus oxyacantha	2.82	1.16	2.43	23	17.04	36
9	Carthamus lanatus	3.72	0.94	3.95	8-9	5.3	16
10	Causinia prolifera	4.26	1.22	3.49	6-7	4	19
11	Cirsium arvense	3.28	1.22	2.68	9-12	6	21
12	Eclipta prostrate	6.04	3.04	1.98	13	11.87	23
13	Erigeron bonariensis	1.86	0.5	3.72	25	6.9	25
14	Erigeron canadensis	1.6	1.82	0.87	16	7.8	12
15	Ixeris polycephala	4.86	1.38	3.52	6	11.2	31
16	Lactuca sativa	3.48	1.28	2.71	15	12.3	121
17	Lactuca serriola	2.7	1.16	2.32	13	2.66	33
18	Launea naudicaulis	3.56	1.82	1.95	9-13	2.63	54
19	Launea procumbens	6.26	2.52	2.48	15-17	2.45	55
20	Parthenium hysterophorus	4.9	3.92	1.25	3-5	2.54	23

 Table 16: Quantitative Seed micro-morphological attributes of Asteraceous species

Sr. No	Plant Taxa	Cypsela Color	Cypsela Shape	Pappus	Carpopodium	Surface	Beak color	Beak Shape
				Color	Position	Pubescence		
1	Ageratum conyzoides	Light Brown	Linear	Absent	Basal	Scabrous	Absent	Absent
2	Artemisia absinthimum	Dark Brown	Linear fusiform	Light	Lateral	Glabrous	Brown	Slender
				Brown				
3	Aster squamatus	Dark Brown	Curved-ring	Dull White	Sub-Basal	Glabrous	Light Brown	Filiform
			shaped					
4	Biden pilosa	Straw White	Oblanceolate	Brown	Lateral	Scabrous	White	Filiform
5	Bidens bipinnata	Light to Dark Brown	Obovoid	Brown	Lateral	Scabrous	Brown	Filiform
6	Calendula arvensis	Greyish	Cylindrical	Brown	Basal	Scabrous	Greyish	Slender
7	Calendula officinalis	Brown	Pointed	White	Basal	Scabrous	Brown	Slender
8	Carthamus oxyacantha	Light Brown	Linear lanceolate	White	Basal	Scabrous	Brown	Slender
9	Carthamus lanatus	Dark Brown	Linear lanceolate	White	Lateral	Glabrous	White	Slender
10	Causinia prolifera	Black	Oblanceolate	Brown	Sub-Basal	Glabrous	White	Filiform
11	Cirsium arvense	Brown with black	Narrowly	Light	Lateral	Glabrous	Light Brown	Filiform
		spots	lanceolate	Brown				
12	Eclipta prostrate	Yellowish Brown	Obovoid	Dull White	Basal	Scabrous	Brown	Slender
13	Erigeron bonariensis	Black	Oblanceolate	Brown	Lateral	Scabrous	Brown	Slender
14	Erigeron Canadensis	Dark Brown	Linear	Brown	Basal	Glabrous	Brown	Slender
15	Ixeris polycephala	Black	Linear	Brown	Lateral	Glabrous	Light Brown	Filiform
16	Lactuca sativa	Light Brown	Linear	Brown	Sub-Basal	Glabrous	White	Filiform
17	Lactuca serriola	Green to Brownish	Curved	White	Lateral	Scabrous	Brown	Filiform
18	Launea naudicaulis	Red	Oblanceolate	White	Basal	Scabrous	Light Brown	Slender
19	Launea procumbens	Black	Oblanceolate	Light	Basal	Glabrous	White	Filiform
				Brown				
20	Parthenium hysterophorus	Brown	Obovate	White	Lateral	Scabrous	Brown	Slender

 Table 17: Qualitative Seed micro-morphological attributes of Asteraceous species ..

Sr. No	Plant Name	Surface	Epicuticular	<b>Carpopodium Shape</b>	Anticlinal wall	Anticlinal	Periclinal wall	Periclinal wall
		Pattern	Projection		Level	wall pattern	Level	Protuberance
1	Ageratum conyzoides	Striated	Platelets	Incompletely	Raised	Wavy	Depressed	Coarse Textured
				Developed				
2	Artemisia absinthimum	Irregularly	Bulges	Undeveloped Circular	Raised	Puzzled	Flat	Glabrous
		striated		Ring				
3	Aster squamatus	Irregularly	Absent	Undeveloped angular	Raised	Wavy	Depressed	Fine Textured
		Reticulate		Ring				
4	Biden pilosa	Irregularly	Granules	Completely Developed	Depressed	Entire	Raised	Glabrous
		Striato-		with Circular ring				
		Regulate		without interruptions				
5	Bidens bipinnata	Reticulate	Hair and	Completely developed	Raised	Grooved	Depressed	Medium Textured
			Granules	symmetrical ring				
				without interruptions				
6	Calendula arvensis	Verrucose	Dome shaped	Completely Developed	Depressed	Wavy	Raised	Glabrous
		papilate	Bulges	asymmetrical ring				
				without interruptions				
7	Calendula officinalis	Striated	Granules	Completely developed	Raised	Grooved with	Convex	Glabrous
				square ring without		acuminate		
				interruptions		apex		
8	Carthamus oxyacantha	Regulate	Micro Hair and	Completely Developed	Depressed	Slightly Wavy	Raised	Fine Textured
			Bulges	circular ring without				
				interruptions and 5				
				Lobed				
9	Carthamus lanatus	Reticulate	Hair	Undeveloped	Raised	Wavy	Raised	Coarse Textured
								with Bulges
10	Causinia prolifera	Verrucate-	Granules	Incompletely	Raised	Entire	Raised	Fine Textured
		Reticulate		Developed U-Shaped				

 Table 18: SEM Ultrastructural Seed coat micro-morphological features of Cypsela

				with interruption				
11	Cirsium arvense	Verrucate Striate	Granules	Undeveloped circular Ring	Raised	Grooved	Depressed	Glabrous
12	Eclipta prostrate	Striated	Bulges	Completely Developed Circular Ring without interruption and slightly lobed	Raised	Undulate	Raised	Glabrous with Bulges
13	Erigeron bonariensis	Papilate	Micro Hairs	Incompletely developed asymmetrical Ring	Depressed	Wavy	Slightly Raised and Mostly Depressed	Glabrous
14	Erigeron canadensis	Reticulate	Granules	Undeveloped and regular	Depressed	Puzzled	Convex	Medium Textured
15	Ixeris polycephala	Regulate	Bulges	Developed Circular Ring	Raised	Entire	Depressed	Coarse Textured
16	Lactuca sativa	Scrobiculate	Absent	Undeveloped angular ring	Depressed	Slightly Wavy	Raised	Glabrous
17	Lactuca serriola	Striated	Micro Hairs with Bulges	Incompletely angular ring	Raised	Undulate with acute apex	Depressed	Fine Textured
18	Launea naudicaulis	Reticulate	Granules	Irregular ring	Raised	Undulate	Raised	Coarse Textured with Bulges
19	Launea procumbens	Verrucose	Absent	Completely developed with circular ring without interruptions	Raised	Wavy	Flat	Medium Textured
20	Parthenium hysterophorus	Striated	Bulges	Incompletely developed	Raised	Entire	Raised	Glabrous

#### 3.3.1 Micro-morphological characters of cypsela

#### 1. Morphology of Ageratum conyzoides Cypsela

The surface pattern of Cypsela observed was Striated, Epicuticular projections observed on Cypsela surface was Platelets. Carpopodium Shape was Incompletely Developed .Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls Pattern were Wavy while Periclinal wall level were Depressed and Periclinal Protuberance were Coarse Textured. Cypsela color Light Brown, Cypsela Shape is Linear. Carpopodium position were Basal. Surface Pubescence is Scabrous. Pappus absent. The average length is 7.56mm and the average width is 2.84mm. L/W ratio is 2.66 mm. Number of ribs 6-7. Beak absent. Average number of Cypsela per capitulum 25 (Plate 124).

### 2. Morphology of Artemisia absinthimum Cypsela

The surface pattern of Cypsela observed was irregularly striated, Epicuticular projections observed on Cypsela surface was Bulges. Carpopodium Shape was Undeveloped Circular Ring.Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls pattern were Puzzled while Periclinal wall level were flat and Periclinal Protuberance were Glabrous. Cypsela color Dark Brown, Cypsela Shape is Linear fusiform. Carpopodium position were Lateral. Surface Pubescence is Glabrous. Pappus Color Light Brown and length 3.36mm. The average length is 2.16mm and the average width is 0.82mm. L/W ratio is 2.63 mm. Number of ribs 12-14. Beak color is Brown and shape is slender. Average number of Cypsela per capitulum 17 (Plate 124).

### 3. Morphology of Aster squamatus Cypsela

The surface pattern of Cypsela observed was Irregularly reticulate, Epicuticular projections observed on Cypsela surface was Absent. Carpopodium Shape was underdeveloped angular ring.Anticlinal walls on Cypsela surface were Raised and Anticlinal walls pattern were Wavy while Periclinal wall level were depressed and Periclinal Protuberance were fine textured. Cypsela color Dark Brown, Cypsela Shape is Curved-ring shaped. Carpopodium position were Sub-Basal. Surface Pubescence is Glabrous. Pappus color dull white and length is 1.76mm. The average length is 3.98mm and the average width is 0.92mm. L/W ratio is 4.32 mm. Number of ribs

absent. Beak color is Light Brown and shape is Filiform.. Average number of Cypsela per capitulum 13 (Plate 125)

## 4. Morphology of Biden pilosa Cypsela

The surface pattern of Cypsela observed was Irregularly Striato-Regulate, Epicuticular projections observed on Cypsela surface was Granules. Carpopodium Shape was Completely Developed with Circular ring without interruptions.Anticlinal walls level on Cypsela surface were Depressed and Anticlinal walls Pattern were Entire while Periclinal wall level were Raised and Periclinal wall Protuberance were Glabrous. Cypsela color Straw White, Cypsela Shape is Oblanceolate. Carpopodium position were Lateral. Surface Pubescence is Scabrous. Pappus color is Brown and length 24.6mm. The average length is 3.52mm and the average width is 1.08mm. L/W ratio is 3.25mm. Number of ribs 13-15. Beak absent. Average number of Cypsela per capitulum 14 (Plate 125).

## 5. Morphology of Bidens bipinnata Cypsela

The surface pattern of Cypsela observed was Reticulate, Epicuticular projections observed on Cypsela surface was Hair and Granules. Carpopodium Shape was completely developed symmetrical ring without interruptions. Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls pattern were grooved while Periclinal wall level were depressed and Periclinal Protuberance were Medium textured. Cypsela color Light to Dark Brown, Cypsela Shape is obovoid. Carpopodium position were lateral. Surface Pubescence is Scabrous. Pappus color Browna and length 16.96. The average length is 3.36mm and the average width is 0.92mm. L/W ratio is 3.65 mm. Number of ribs 7. Beak color Brown and shape filiform. Average number of Cypsela per capitulum 10 (Plate 126).

### 6. Morphology of Calendula arvensis Cypsela

The surface pattern of Cypsela observed was Verrucose papilate, Epicuticular projections observed on Cypsela surface was Dome shaped bulges. Carpopodium Shape was completely developed asymmetrical ring without interruptions. Anticlinal walls level on Cypsela surface were Depressed and Anticlinal walls pattern were Wavy while Periclinal wall level were Raised and Periclinal Protuberance were Glabrous. Cypsela color Greyish, Cypsela Shape is cylindrical. Carpopodium position

were Basal. Surface Pubescence is Scabrous. Pappus color Brown and length 3.5mm. The average length is 6.74mm and the average width is 3.14mm. L/W ratio is 2.21 mm. Number of ribs 9. Beak color Greyish and shape is slender. Average number of Cypsela per capitulum 37 (Plate 126).

## 7. Morphology of Calendula officinalis Cypsela

The surface pattern of Cypsela observed was Striated, Epicuticular projections observed on Cypsela surface was Granules. Carpopodium Shape was completely developed square ring without interruptions. Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls pattern were grooved with acuminate apex while Periclinal wall level were convex and Periclinal Protuberance were glabrous. Cypsela color Brown, Cypsela Shape is Pointed. Carpopodium position were Basal. Surface Pubescence is Scabrous. Pappus color is white and length 4.12 absent. The average length is 2.04mm and the average width is 1.04mm. L/W ratio is 1.96 mm. Number of ribs 21. Beak color Brown and shape were slender. Average number of Cypsela per capitulum 33 (Plate 127).

## 8. Morphology of Carthamus oxyacantha Cypsela

The surface pattern of Cypsela observed was Regulate, Epicuticular projections observed on Cypsela surface was Micro Hair and Bulges. Carpopodium Shape was completely developed circular ring without interruptions and 5 lobed. Anticlinal walls level on Cypsela surface were Depressed and Anticlinal walls pattern were slightly Wavy while Periclinal wall level were Raised and Periclinal Protuberance were fine textured. Cypsela color Light Brown, Cypsela Shape is Linear lanceolate. Carpopodium position were Basal. Surface Pubescence is Scabrous. Pappus color white . The average length is 2.82mm and the average width is 1.16mm. L/W ratio is 2.43 mm. Number of ribs 23. Beak color Brown and shape slender. Average number of Cypsela per capitulum 36 (Plate 127).

### 9. Morphology of Carthamus lanatus Cypsela

The surface pattern of Cypsela observed was Reticulate, Epicuticular projections observed on Cypsela surface was Hair. Carpopodium Shape was UnDeveloped .Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls pattern were Wavy while Periclinal wall level were raised and Periclinal Protuberance were coarse textured with bulges. Cypsela color Dark Brown, Cypsela Shape is Linear Lanceolate. Carpopodium position were Lateral. Surface Pubescence is Glabrous. Pappus color white and length 5.3 absent. The average length is 3.72mm and the average width is 0.94mm. L/W ratio is 3.95 mm. Number of ribs 8-9. Beak color white and shape slender. Average number of Cypsela per capitulum 16 (Plate 128).

## 10. Morphology of Causinia prolifera Cypsela

The surface pattern of Cypsela observed was Striated, Epicuticular projections observed on Cypsela surface was Platelets. Carpopodium Shape was Incompletely Developed .Anticlinal walls on Cypsela surface were Raised and Anticlinal walls were Wavy while Periclinal wall level were depressed and Periclinal Protuberance were coarse textured. Cypsela color Black, Cypsela Shape is Oblanceolate. Carpopodium position were sub-Basal. Surface Pubescence is Glabrous. Pappus color Brown and length is 4mm. The average length is 4.26mm and the average width is 1.22mm. L/W ratio is 3.49 mm. Number of ribs 6-7. Beak color white and shape Filiform. Average number of Cypsela per capitulum 19 (Plate 128).

## 11. Morphology of Cirsium arvense Cypsela

The surface pattern of Cypsela observed was Verrucate Striate, Epicuticular projections observed on Cypsela surface was Granules. Carpopodium Shape was UnDeveloped Circular Ring .Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls Pattern were Grooved while Periclinal wall level were depressed and Periclinal Protuberance were coarse Glabrous. Cypsela color Brown with black spots, Cypsela Shape is narrowly lanceolate. Carpopodium position were Lateral. Surface Pubescence is Glabrous. Pappus color Light brown and length 6mm. The average length is 3.28mm and the average width is 1.22mm. L/W ratio is 2.68 mm. Number of ribs 9-12. Beak light brown and shape Filiform. Average number of Cypsela per capitulum 21 (Plate 129).

### 12. Morphology of Eclipta prostrate Cypsela

The surface pattern of Cypsela observed was Striated, Epicuticular projections observed on Cypsela surface was Bulges. Carpopodium Shape was completely developed circular ring without interruption and slightly lobed. Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls pattern were undulate while Periclinal wall level were Raised and Periclinal Protuberance were Glabrous with Bulges. Cypsela color Yellowish Brown, Cypsela Shape is Obovoid. Carpopodium position were Basal. Surface Pubescence is Scabrous. Pappus color Dull White and length 11.87mm. The average length is 6.04mm and the average width is 3.04mm. L/W ratio is 1.98 mm. Number of ribs 13. Beak Color Brown and shape slender. Average number of Cypsela per capitulum 23 (Plate 129).

### 13. Morphology of Erigeron bonariensis Cypsela

The surface pattern of Cypsela observed was Papilate, Epicuticular projections observed on Cypsela surface was Micro Hairs. Carpopodium Shape was Incompletely Developed asymmetrical ring .Anticlinal walls level on Cypsela surface were Depressed and Anticlinal walls Pattern were Wavy while Periclinal wall level were Slightly raised and mostly depressed and Periclinal Protuberance were Glabrous. Cypsela color Black, Cypsela Shape is Oblanceolate. Carpopodium position were Lateral. Surface Pubescence is Scabrous. Pappus color Brown and length 6.9mm. The average length is 1.86mm and the average width is 0.5mm. L/W ratio is 3.72 mm. Number of ribs 25. Beak color Brown and shape slender. Average number of Cypsela per capitulum 25 (Plate 130).

### 14. Morphology of Erigeron canadensis Cypsela

The surface pattern of Cypsela observed was Reticulate, Epicuticular projections observed on Cypsela surface was Granules. Carpopodium Shape was Undeveloped and regular. Anticlinal walls level on Cypsela surface were Depressed and Anticlinal walls Pattern were Puzzled while Periclinal wall level were convex and Periclinal Protuberance were medium textured. Cypsela color Light Brown, Cypsela Shape is Linear. Carpopodium position were Basal. Surface Pubescence is Scabrous. Pappus Length 7.8mm absent. The average length is 1.6mm and the average width is 1.82mm. L/W ratio is 0.87 mm. Number of ribs 16. Beak absent. Average number of Cypsela per capitulum 12 (Plate 130).

### 15. Morphology of Ixeris polycephala Cypsela

The surface pattern of Cypsela observed was Regulate, Epicuticular projections observed on Cypsela surface was Bulges. Carpopodium Shape was Developed circular ring.Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls Entire while Periclinal wall level were depressed and Periclinal Protuberance were coarse textured. Cypsela color Black, Cypsela Shape is Linear. Carpopodium position were Lateral Surface Pubescence is Glabrous. Pappus color brown and length 11.2mm. The average length is 4.86mm and the average width is 1.38mm. L/W ratio is 3.52mm. Number of ribs 6. Beak color Light Brown and shape filiform. Average number of Cypsela per capitulum 31 (Plate 131).

#### 16. Morphology of Lactuca sativa Cypsela

The surface pattern of Cypsela observed was Striated, Epicuticular projections observed on Cypsela surface was Platelets. Carpopodium Shape was Incompletely Developed .Anticlinal walls on Cypsela surface were Raised and Anticlinal walls were Wavy while Periclinal wall level were depressed and Periclinal Protuberance were coarse textured. Cypsela color Light Brown, Cypsela Shape is Linear. Carpopodium position were Basal. Surface Pubescence is Scabrous. Pappus absent. The average length is 7.56mm and the average width is 2.84mm. L/W ratio is 2.66 mm. Number of ribs 12. Beak color White and Shape filiform. Average number of Cypsela per capitulum 25 (Plate 131).

### 17. Morphology of Lactuca serriola Cypsela

The surface pattern of Cypsela observed was Striated, Epicuticular projections observed on Cypsela surface was Micro Hairs with Bulges. Carpopodium Shape was Incompletely angular ring. Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls pattern were undulate with acute apex while Periclinal wall level were depressed and Periclinal Protuberance were fine textured. Cypsela color Green to Brownish, Cypsela Shape is Curved. Carpopodium position were Lateral. Surface Pubescence is Scabrous. Pappus color is white and length 2.66mm absent. The average length is 2.7mm and the average width is 1.16mm. L/W ratio is 2.32 mm. Number of ribs 13. Beak color Brown and shape Filiform. Average number of Cypsela per capitulum 33 (Plate 132).

### 18. Morphology of Launea naudicaulis Cypsela

The surface pattern of Cypsela observed was Reticulate, Epicuticular projections observed on Cypsela surface was Granules. Carpopodium Shape was Irregular ring.Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls

pattern were Undulate while Periclinal wall level were Raised and Periclinal Protuberance were coarse textured with bulges. Cypsela color Light Brown, Cypsela Shape is Oblanceolate . Carpopodium position were Basal. Surface Pubescence is Scabrous. Pappus color white and length 2.63 . The average length is 3.56mm and the average width is 1.82mm. L/W ratio is 1.95mm. Number of ribs 9-13. Beak color Light Brown and Shape Slender. Average number of Cypsela per capitulum 54 (Plate 132).

#### 19. Morphology of Launea procumbens Cypsela

The surface pattern of Cypsela observed was Verrucose, Epicuticular projections observed on Cypsela surface was Absent. Carpopodium Shape was completely Developed with circular ring without interruptions .Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls pattern were Wavy while Periclinal wall level were flat and Periclinal Protuberance were Medium textured. Cypsela color Black, Cypsela Shape is Oblanceolate. Carpopodium position were Basal. Surface Pubescence is Glabrous. Pappus color Light Brown and length 2.5. The average length is 6.26mm and the average width is 2.52mm. L/W ratio is 2.48 mm. Number of ribs 15-17. Beak color White and shape Filiform. Average number of Cypsela per capitulum 55 (Plate 133)

### 20. Morphology of Parthenium hysterophorus Cypsela

The surface pattern of Cypsela observed was Striated, Epicuticular projections observed on Cypsela surface was Bulges. Carpopodium Shape was Incompletely Developed .Anticlinal walls level on Cypsela surface were Raised and Anticlinal walls pattern were Entire while Periclinal wall level were Raised and Periclinal Protuberance were Glabrous. Cypsela color Brown, Cypsela Shape is Obovate. Carpopodium position were Lateral . Surface Pubescence is Scabrous. Pappus color White and length 2.56. The average length is 4.9mm and the average width is 3.92mm. L/W ratio is 1.25 mm. Number of ribs 3-5. Beak color Brown and shape Slender. Average number of Cypsela per capitulum 23 (Plate 133).

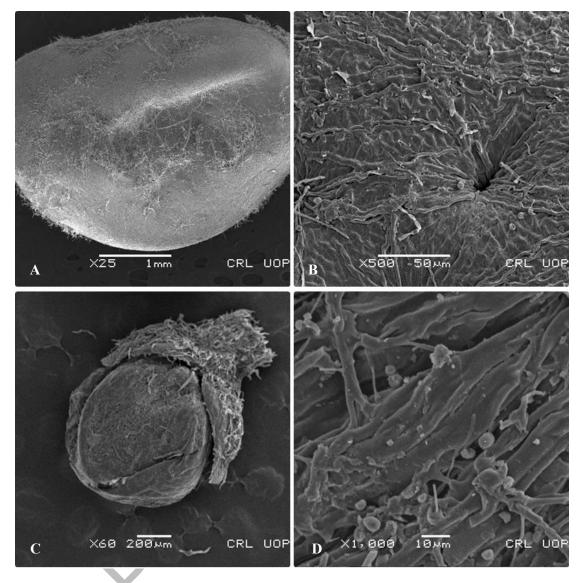


Plate 124: Cypsela micrographs of *Ageratum conyzoides* (A) General View (B) Surface Sculpturing , *Artemesia absanthimum* (C) General View (D) Surface Sculpturing

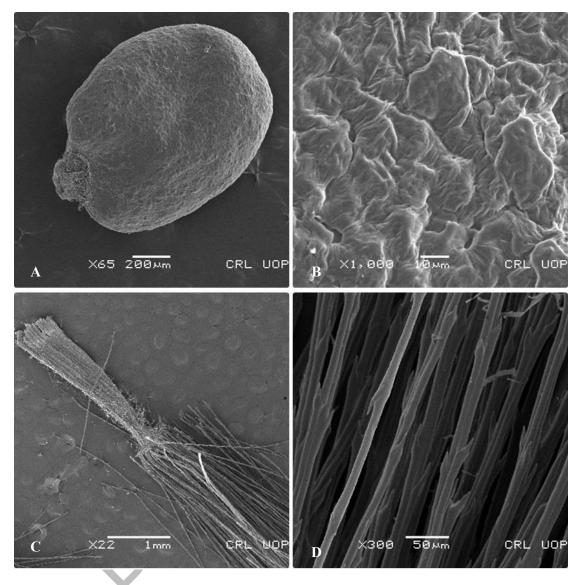
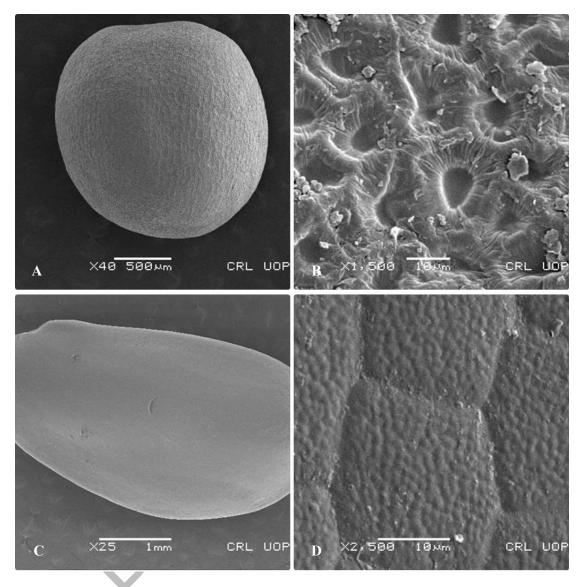
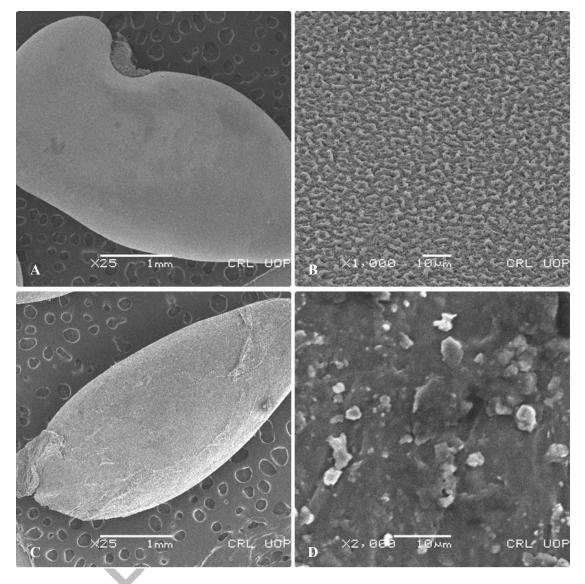


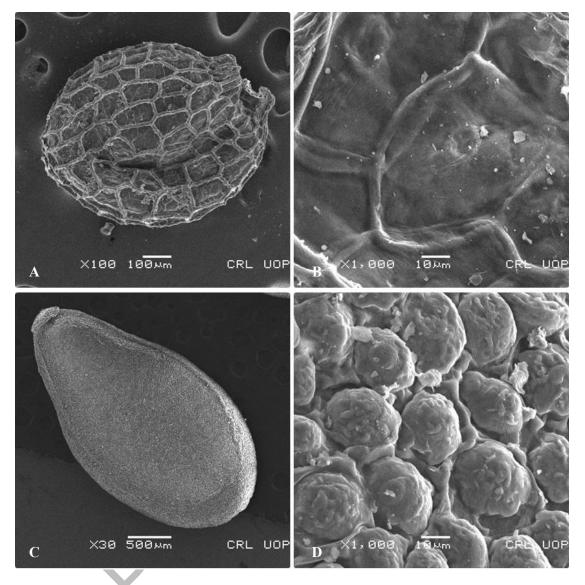
Plate 125: Cypsela micrographs of *Aster squamatus* (A)General View (B) Surface Sculpturing , *Biden pilosa* (C)General View (D) Surface Sculpturing



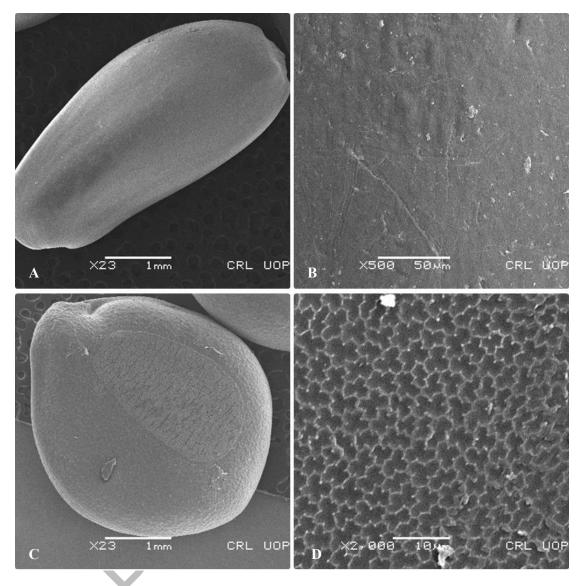
**Plate 126:** Cypsela micrographs of *Bidens bipinnata*, (A)General View (B) Surface Sculpturing , *Calendula arvensis*(C)General View (D) Surface Sculpturing



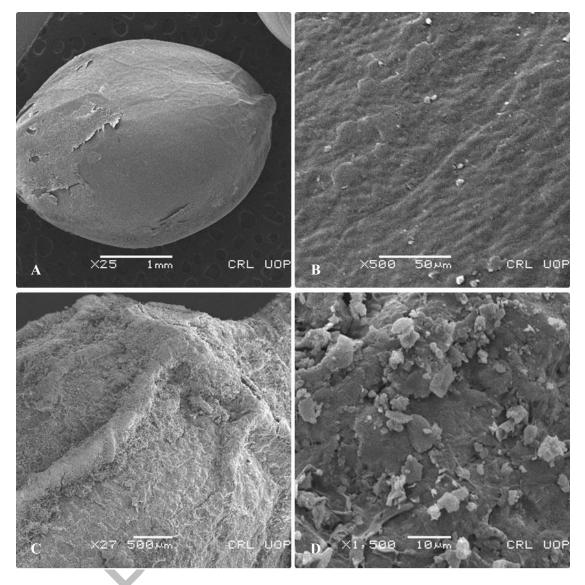
**Plate 127:** Cypsela micrographs *Calendula officinalis* (A)General View (B) Surface Sculpturing , *Carthamus oxyacantha* (C)General View (D) Surface



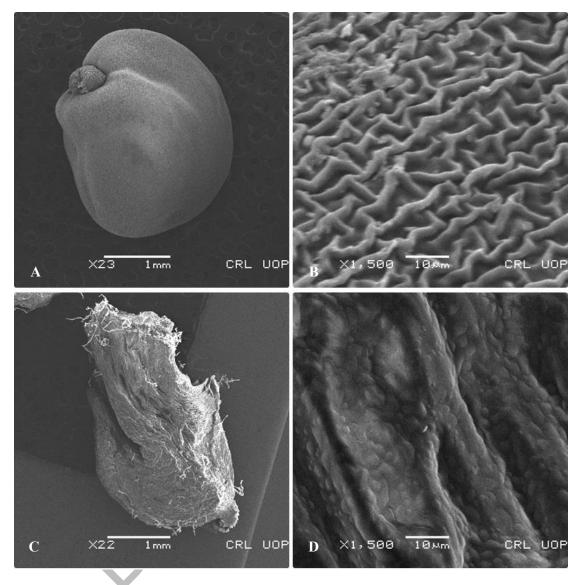
**Plate 128:** Cypsela micrographs of *Carthamus lanatus*(A)General View (B) Surface Sculpturing , *Causinia prolifera* (C)General View (D) Surface Sculpturing



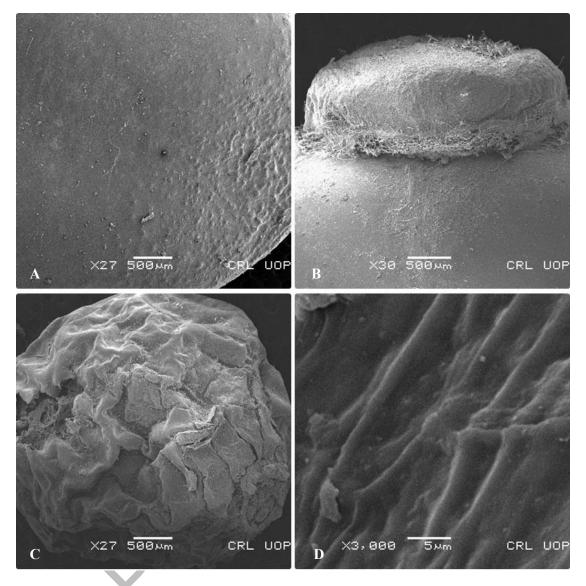
**Plate 129:** Cypsela micrographs of *Cirsium arvense* (A)General View (B) Surface Sculpturing , *Eclipta prostrate* (C)General View (D) Surface Sculpturing



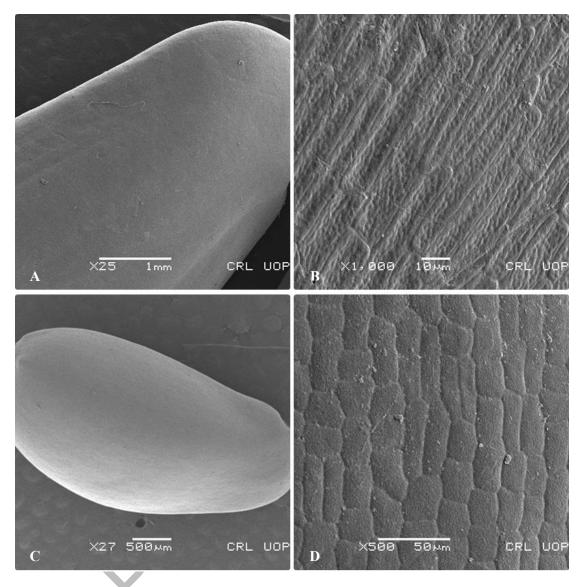
**Plate 130:** Cypsela micrographs of *Erigeron bonariensis* (A) General View (B) Surface Sculpturing , *Erigeron Canadensis* (C) General View (D) Surface Sculpturing



**Plate 131:** Cypsela micrographs of *Ixeris polycephala* (A) General View (B) Surface Sculpturing , *Lactuca sativa* (C) General View (D) Surface Sculpturing



**Plate 132:** Cypsela micrographs of *Lactuca serriola*(A) General View (B) Surface Sculpturing , *Launea naudicaulis* (C) General View (D) Surface Sculpturing



**Plate 133:** Cypsela micrographs *Launea procumbens* (A) General View (B) Surface Sculpturing , *Parthenium hysterophorus* (C) General View (D) Surface Sculpturing

# Taxonomic key based on Seed characters of Asteraceae

1a. Pappus absent	ratum conyzoides
1b. Pollen present	2
2a. Slender beak shape	3
2b. Filiform beak shape	11
3a. basal carpopodium laune	ea procumbens
3b. sub-basal or lateral carpopodium	4
4a. Ribs absent	. Aster squamatus
4b. Ribs present	5
5a. Seed length is below 3 mm	.Lactuca serriola
5b. Seed length is above 3 mm	
6a. Seed width is below 1 mm	Bidens bipinnata
6b. Seed width is above 1 mm	7
7a. Surface pubescence is scabrous	Biden pilosa
7b. Surface pubescence is glabrous	
8a. Epicuticular projections absent	Lactuca sativa
8b. Epicuticular projections present	9
9a. Surface pattern is rugulate	Ixeris polycephala
9b. Surface ornamentation is other than rugulate	10
10a. Cypsela color is black	Causinia prolifera
10b. Cypsela color is brown	Cirsium arvense
11a. Pappus length is above 15 mm	Carthamus oxyacantha
11b. Pappus length is below 15 mm	12
12a. L/W ratio is below 1 mm	Erigeron Canadensis
12b.L/W ratio is above 1 mm	

13a. Cypsela color is red	Launea naudicaulis
13b. Cypsela color is grey, brown or white	14
14a.Number of ribs are 5 or less than 5	Parthenium hysterophorus
14b. Number of ribs are more than 5	
15a. Anticlinal wall patterns are grooved	Calendula officinalis
15b. Anticlinal walls are wavy or puzzled	
16a. Periclinal walls are convex	Erigeron canadensis
16b. Periclinal walls are raised or flat	
17a. Periclinal wall has coarse texture	Carthamus lanatus
17b. Periclinal walls are glabrous	
18a. Seed length below 2mm	
18b. Seed length above 2 mm	
19a. Pappus length is above 10 mm	Eclipta prostrate
19b. Pappus length is below 10 mm	
20a. Seed surface glabrous	Artemesia annua
20b. Seed surface scabrous	Calendula arvensis

#### 3.2.1 Asteraceous Seeds

In this recent Research, macro-morphological and micro-morphological characters of cysela were observed systematically through light microscopy as well as scanning electron microscopy. Cypsela appeared to be variable in terms of exomorphology, as infra generic and infra-specific diversity has been observed for the studied characters of every taxon. Morphological characters of the cypsela such as epicuticular projections and their types, shape of cypsela, surface patterns, anticlinal wall and periclinal wall level were found to be extremely inconsistent within family Asteraceae.

### Variations in Cysela Quantitative Features

Size and shape of cypsela along with micro-morphological characters are significant indicative feature in species classification. The features can be utilized at diverse taxonomic positions to demonstrate resemblance among species (Maciejewska-rutkowska and Bednorz,2004) Quantitative characters such as seed length, seed width, pappus length, number of ribs, number of cypsela/ capitulum has been investigated. The results of this research clearly indicated the disparities in cypsela size among the examined species.

The mean length of the studied mature cypselae can be helpful to differentiate cypselae of *Ageratum conyzoides* and *Erigeron canadensis*. Cypsela length ranges from 1.6-7.56 mm. Among the examined species, Ageratum conyzoides is noted with maximum length that is 7.56mm minimum length was observed in *Erigeron Canadensis* that is 1.6mm. Biden pilosa 3.52, *Bidens bipinnata* 3.56,*Calendula arvensis* 6.74.

Width of cypsela was noted in the range of 0.5-3.92. *Parthenium hysterophorus* Has maximum width of cypsela 3.92 whereas *Erigeron bonariensis* has minimum width of 0.5mm . Range Of Length width ratio in studied species was recorded from 0.87 to 4.32. *Aster squamatus* has maximum value that is 4.32 while minimum value was observed for Erigeron Canadensis that is 0.87. *Causinia prolifera* 3.49, *Cirsium arvense* 2.68, *Eclipta prostrate* 1.98 Length and width was recorded in different species.

In the studied species pappus length was recorded from 1 to 24.6mm. Among the examined species *Circium arvense* has maximum value that is 24.6mm followed by *cirsium arvense* 24.6mm, *Cirsium sorocephalum* 17.04mm, and *Himalaiella hetromella* 16.96 mm, wheras *Ageratum conyzoides* has minimum value that is 1mm followed by *centaurea iberica* 1.76mm, and youngia japonica 3mm.

The studied cypselae are either ribbed or ribless, (Hussein and Eldemerdash, 2016) reported that some asteraceous species have reveal disparities in number of ribs made the use of this trait controversial. But, in the identification keys the use of number of ribs as a characteristics for differentiation of species from Asteraceae has been adopted. The number of Cypselae per capitulum had a great diversity. Some taxa such as *sonchus asper* had 123, *Himalaiella hetromella* had 117, and *Sonchus arvensis* has 112 cypselae per capitulum.

### Variations in Cypsela Qualitative Characters Via LM

Light microscope was used to examine macro-morphological features of Cypsela. The investigated species have great variation in term of color. Majority of the cypselae has Brown Color. The color of cypsela seems clearly distinctive to cypselae of *Carthamus lanatus* having straw white color. Reference.

Stated that the color of cypsela is not reliable distinctive character because the color changes with the degree of maturity of Cypselae. The Cypsela shape appeared unique in *Calendula arvensis* and *Calendula officinalis* being curved. In addition, the cypsela shape seems to be clearly diagnostic for some of the examined taxa. Most of the species show variations in cypselae shape. Linear to linear lanceolate, oblanceolate to obovate, curved to ring and obovoid cypselae shapes were examined. *Aster squamatus, Bidens bipinnata* has linear shape, *Erigeron bonariensis, Erigeron candadensis* has linear lanceolate shape while *Bidens bipinnata* has linear fusiform shape. *Centaurea iberica, Parthenium hystiphorus, Silybum marianum* has obovoid shape, Obovoid shape was observed in *Xanthium stromarium, youngia japonica* obovoid to cylindrical cypsela was present in cichorium intybus, While narrowly lanceolate cypsela was present in *Launea procumbens*. Dominant shape type was oblanceolate observed in the *Sonchus, Lactuca* and rest of the examined taxa.

Also, color of pappus have been found supportive for taxonomic delimitation of the studied taxa. Pappus is of various colors such as white, dull white, dirty white, silky white, brown. In the investigated species pappus is either fragile or not fragile. Carpodium position have also found supportive for taxonomic limitation of the studied taxa. Carpopodium position is basal, sub-basal, and lateral in the studied taxa. Cypsela surface Pubescence is scabrous in twelve species while remaining species have glabrous surface pubescence.

The presence of beaked cypselaeis diagnostic for the four examined species of *Lactuca* having obviously one long beaked cypselae, also for *Ixeris polycephala*, with two short beaked cypselae and *Bidens bipinnata*, *Bidens psilosa* with four short beaks. Beak was observed in most of the species in tribe Cichorieae *Lactuca sativa*, *Lctuca dissecta*, *Lactuca serriola* and white, filiform beak whereas *Bidens bipinnata*, *Bidens psilosa*, *Ixeris polycephala*, have brown slender beak. Lactuca serriola has white slender beak and rest of the examined species are observed without beak.

### Variations in Qualitative features of Cypsela Via SEM

Use of scaning electron microscopy can be very helpful in plant taxonomy as it reveals surface morphology of various vegetative and reproductive organs (Barthlott, 1984). In Present research SEM was used to study micro-morphological features such as surface pattern, epicuticular projections, carpopodium shape, anticlinal and periclinal walls pattern and level.

In studied Plants, surface pattern observed was of 13 types irregularly reticulate, Irregularly striate, Striato-knotted, Irregularly striato- regulate, Verrucatestriate, Reticulate, Scrobiculate, Rugose papillate, Regulate, Striated, Irregularly scrobiculate, Verrucose pappillate, Verrucate reticulate. *Carthamus lanatus*, *Carthamus oxycantha*, *Causinia prolifera* and *Cirsium arvens* has reticulate surface Pattern. While *Lactuca dissecta*, *Lactuca sativa*, *Lactuca serriola*, *Sonchus oleraceus*, and *Carthamus lanatus* has verrucate papillate surface pattern. Certain variations were observed in the species of same genus as sonchus oleraceous has verrucose papillate surface pattern and *Sonchus* arvensisand Sonchus asper has Striated surface Pattern. Surface pattern of the cypsela of *Bidens bipinnata* is irregularly striated whereas *Bidens psilosa* has striato-knotted surface pattern, making surface pattern valuable as one more diagnostic characterto differentiate the genera . *Carthamus lanatus* has verrucose pappillate surface pattern while rest of the species from the same tribe has different surface patterns making it easy to distinguish from each other. In addition the studied five taxa of tribe Heliantheae *Eclipta prostrate*, *Lactuca sativa*, *Parthenium hystiphorus*, *Calendula officinalis*, and *Carthamus lanatus* could be easily delimited at the generic level through their cypsela surface micro- sculpture. *Parthenium hystiphorus* and *Eclipta prostrata* has regulate surface pattern while helianthus annus has irregularly striated surface pattern and *Zinnia elegans* has verrucate reticulate surface pattern.

Epicuticular projections reveal variations in terms of Micro-morphological features Reference add. In studied species, five type of epicuticular projections were observed as granules, Platelets, bulges, hair, dome shaped bulges whereas absent in some taxa. *Lactuca dissecta*, *Lactuca sativa*, *Lactuca serriola*, *Sonchus arvensis*, *Sonchus oleraceous, Calendula arvensis*, *Launea naudicaulis, Launea procumbens*, has granules. *Calendula arvensis* and *Calendula officinales* has bulges while *Parthenium hystiphorus* has dome shaped bulges. *Bidens bipinnata, Bidens psilosa, Launea procumbens, Ageratum conyzoides* and *sonchus asper* has platelets Epicuticular projections are absent in rest of ythe taxa.

Observations of cypselae under scanning electron microscopy has revealed the presence of the obcission scar termed as carpopodium in all the studied taxa either at a basal or sub-basal or lateral position. The carpopodium in Aster squamatus, Bidens bipinnata, Bidens psilosa, Centaurea ibrica, Cirsium arvense, Cirsium sorocephalum, Eclipta prostrate, Erigeron bonariensis, Helianthus annus, Himalaiella hetromalla,Lactuca dissecta, Silybum marianum, Taraxacum campylodes, Youngia japonica is completely developed without any interruption. Undeveloped in Cichorium intybus and Launea procombens and incompletely developed in rest of examined taxa. This observation matches with that previously reported by References add. The shape of the Carpodium may seem to be a reliable character for generic separation as observed in the three studied species of Lactuca. Carpopodium features have always been found useful for the taxonomic delimitation of the various taxa of the family asteraceae. Anticlinal and periclinal wall except Calendula officinalis, Carthamus lanatus, Carthamus oxycantha, Cirsium arvense, Parthenium hystiphorus,

Sonchus oleraceous, Launea naudicaulis, Launea procumbens in which anticlinal wall is depressed. Anticlinal wall pattern were observed as entire, slightly wavy to wavy, grooved and puzzled. The examined species have flat, depressed, raised, and convex level of periclinal walls. The protuberance of periclinal wall is mainly glabrous, five species have fine texture while four species are medium textured. But coarse texture protuberance is observed in three species *Parthenium hystiphorus*, *Lactuca sativa*, *Launea procumbens*.

Sr. No	Plant Taxa	Seed Color	Seed Shape	Seed outline	Seed apex	Seed Base	Hilum Position	Sculpturing	Anticlinal Wall Pattern	Epidermal cell shape	Hilum
1	Acacia nilotica	Gray	Flat	Irregular	Slightly Round	Round	NA	Papillate	Wavy	Irregular	Not Visible
2	Accacia modesta	Dark Gray	Oval- Flat	Oblong	Round	Pointed	NA	Striate	Wavy	Radiate	Not Visible
3	Arachis hypagae	Grayish Black	Round- Oval	Elliptic	Round	Oval- Round	NA	Reticulate	Undulate	Flat	Not Visible
4	Astragalus psilocentros	Gray	Ovate	Irregular	Round	Round	Terminal	Reticulate	Not-visible	Flat	Visible
5	Astragalus scoripiurus	Gray	Oblong	Oblong	Pointed	Round	NA	Reticulate	Smooth	Round	Not Visible
6	Cassia fistula	Light Gray	Ovate	Ovate	Pointed	Slightly Round	NA	Striate	Straight	Wavy	Not Visible
7	Cassia italica	Grayish Black	Elliptic	Oval	Beak Like	Narrow	Central	Granulate	Wavy	Wavy	Visible
8	Cassia occidentalis	Gray	Slightly Round	Irregular	Flat	Round	Central	Smooth	Undulate	Curved	Visible
9	Crotolaria burhia	Gray	Bean Shaped	Ovate	Round	Narrow	Terminal	Granulate	Undulate	Flat	Visible
10	Dilbergia sisso	Gray	Oval	Oblong	Round	Pointed	NA	Slightly-Striate	Wavy	Round	Not Visible
11	Lathyrus aphaca	Grayish Black	Elliptic	Ovate	Pointed	Round	NA	Granulate	Smooth	Round	Not Visible
12	Lespedeza juncea	Grayish Black	Ovate	Irregular	Beak Like	Oval- Round	Central	Striate	Straight	Radiate	Visible
13	Leucaena	Gray	Round	Oval	Flat	Slightly	Terminal	Papillate	Straight	Irregular	Visible

 Table 19: Micro- morphological attributes of seeds of family Fabaceae

4	leucocephalla					-Round					
	Melilotus indicus	Gray	Flat	Elliptic	Pointed	Pointed	Central	Striate	Undulate	Round	Visible
15	Pigomia pinnata	Light Gray	Ovate	Oblong	Pointed	Round	Central	Reticulate	Wavy	Flat	Visible
16	Prosopis cineraria	Grayish Black	Elliptic	Elliptic	Round	Pointed	NA	Papillate	Wavy	Round	Not Visible
17	Prosopis juliflora	Grayish Black	Slightly Rpound	Oval	Flat	Narrow	NA	Granulate	Wavy	Radiate	Not Visible
18	Pueraria tuberosa	Gray	Ovate	Oblong	Beak Like	Narrow	Central	Striate	Undulate	Wavy	Visible
19	Rhynchosia capitata	Gray	Elliptic	Elliptic	Round	Round	Terminal	Reticulate	Smooth	Irregular	Visible
20	Sophora mollis	Light Gray	Flat	Oval	Pointed	Round	Central	Papillate	Wavy	Radiate	Visible

#### 3.3.2 Seeds Micromorphology of Fabaceous Species

#### 1. Acacia nilotica

The seed color observed was Gray, shape flat, seed outline regular, seed apex was slightly round and seed base is round. Epicuticular projections observed on seed surface was Papillate. Anticlinal walls pattern Wavy. Epidermal cell shape Irregular and Hilum not visible (Plate 134).

### 2. Accacia modesta

The seed color observed was Dark Gray, shape Oval-Flat,seed outline Oblong,seed apex was round and seed base is Pointed. Epicuticular projections observed on seed surface was Striate. Anticlinal walls pattern Wavy. Epidermal cell shape Radiate and Hilum not visible (Plate 134).

### 3. Arachis hypagae

The seed color observed was Grayish Black, shape Round-Oval, seed outline Elliptic, seed apex was Round and seed base is Oval-Round. Epicuticular projections observed on seed surface was Reticulate. Anticlinal walls pattern Undulate. Epidermal cell shape Flat and Hilum not visible (Plate 135).

## 4. Astragalus psilocentros

The seed color observed was Gray, shape Ovate, seed outline Irregular, seed apex was round and seed base is round. Epicuticular projections observed on seed surface was Reticulate. Anticlinal walls pattern not visible. Epidermal cell shape Round and Hilum not visible (Plate 135).

## 5. Astragalus scoripiurus

The seed color observed was Gray, shape oblong, seed outline oblong, seed apex was slightly pointed and seed base is round. Epicuticular projections observed on seed surface was Reticulate. Anticlinal walls pattern smooth. Epidermal cell shape Round and Hilum not visible (Plate 136).

### 6. Cassia fistula

The seed color observed was Light Gray, shape ovate ,seed outline ovate,seed apex was slightly pointed and seed base is slightly round. Epicuticular projections observed on seed surface was striate. Anticlinal walls pattern straight. Epidermal cell shape wavy and Hilum not visible (Plate 136).

#### 7. Cassia italica

The seed color observed was Grayish black, shape Elliptic, seed outline oval, seed apex was sbeak like and seed base is narrow. Hilum position central. Epicuticular projections observed on seed surface was Granulate. Anticlinal walls pattern Wavy. Epidermal cell shape wavy and Hilum visible (Plate 137).

### 8. Cassia occidentalis

The seed color observed was Gray, shape slightly round, seed outline irregular, seed apex was slightly round and seed base is round. Hilum position central. Epicuticular projections observed on seed surface was smooth. Anticlinal walls pattern undulate. Epidermal cell shape curved and Hilum visible (Plate 137).

#### 9. Crotolaria burhia

The seed color observed was Gray, bean shaped, seed outline ovate, seed apex was round and seed base is narrow. Hilum position terminal. Epicuticular projections observed on seed surface was Granulate. Anticlinal walls pattern undulate. Epidermal cell shape flat and Hilum visible (Plate 138).

#### 10. Dilbergia sisso

The seed color observed was Gray, shape oval, seed outline oblong, seed apex was round and seed base is pointed. Epicuticular projections observed on seed surface was Granulate. Anticlinal walls pattern smooth. Epidermal cell shape smooth and Hilum not visible (Plate 138).

## 11. Lathyrus aphaca

The seed color observed was Grayish black, shape elliptic ,seed outline ovate,seed apex was pointed and seed base is round. Epicuticular projections observed on seed surface was Granulate. Anticlinal walls pattern smooth. Epidermal cell shape round and Hilum not visible (Plate 139).

### 12. Lespedeza juncea

The seed color observed was Grayish black, shape ovate, seed outline irregular, seed apex was beak like and seed base is oval round. Hilum Position is central. Epicuticular projections observed on seed surface was striate. Anticlinal walls pattern straight. Epidermal cell shape radiate and Hilum visible (Plate 139).

### 13. Leucaena leucocephalla

The seed color observed was Gray, shape round, seed outline oval, seed apex was slightly round and seed base is round. Epicuticular projections observed on seed surface was Papillate. .Anticlinal walls pattern Wavy. Epidermal cell shape Irregular and Hilum not visible (Plate 140)

#### 14. Melilotus indicus

The seed color observed was Gray, shape flat, seed outline regular, seed apex was slightly round and seed base is round. Epicuticular projections observed on seed surface was Papillate. Anticlinal walls pattern Wavy. Epidermal cell shape Irregular and Hilum not visible (Plate 140).

### 15. Pigomia pinnata

The seed color observed was Gray, shape flat, seed outline regular, seed apex was slightly round and seed base is round. Epicuticular projections observed on seed surface was Papillate. Anticlinal walls pattern Wavy. Epidermal cell shape Irregular and Hilum not visible (Plate 141)

## 16. Prosopis cineraria

The seed color observed was Gray, shape flat, seed outline regular, seed apex was flat and seed base is slightly round. Hilum position terminal. Epicuticular projections observed on seed surface was Papillate. .Anticlinal walls pattern straight. Epidermal cell shape Irregular and not visible (Plate 141)

### 17. Prosopis juliflora

The seed color observed was Grayish black, shape slightly round, seed outline oblong, seed apex was beak like and seed base is narrow. Hilum position central. Epicuticular projections observed on seed surface was striate. Anticlinal walls pattern Wavy. Epidermal cell shape Irregular and Hilum not visible (Plate 142).

### 18. Pueraria tuberosa

The seed color observed was Gray, shape ovate, seed outline oblong, seed apex was beak like and seed base is narrow. Hilum position central. Epicuticular projections observed on seed surface was striate. Anticlinal walls pattern undulate. Epidermal cell shape wavy and Hilum visible (Plate 142).

### 19. Rhynchosia capitata

The seed color observed was Gray, shape elliptic, seed outline elliptic, seed apex was round and seed base is round. Hilum position is Terminal. Epicuticular projections observed on seed surface was Reticulate. Anticlinal walls pattern smooth. Epidermal cell shape Irregular and Hilum visible (Plate 143).

#### 20. Sophora mollis

The seed color observed was Light Gray, shape flat, seed outline oval, seed apex was pointed and seed base is round. Hilum position central. Epicuticular projections observed on seed surface was Papillate. Anticlinal walls pattern Wavy. Epidermal cell shape radiate and Hilum visible (Plate 143).

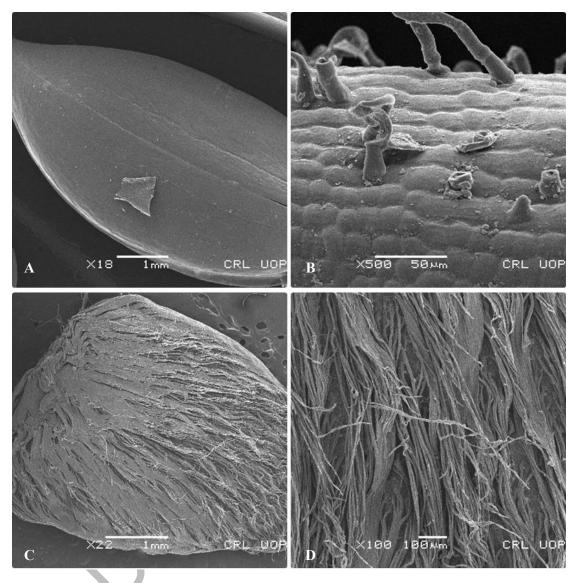


Plate 134: Seed micrographs of *Acacia nilotica* (A) General View (B) Surface Sculpturing, *Accacia modesta* (C) General View (D) Surface Sculpturing

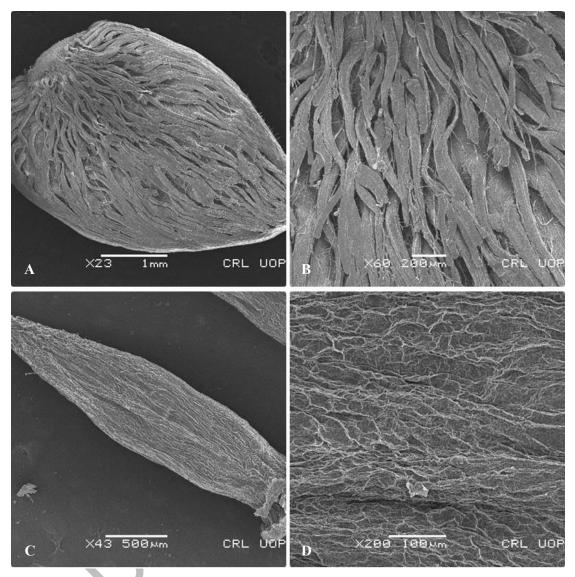


Plate 135: Seed micrographs *Arachis hypagae* (A) General View (B) Surface Sculpturing , *Astragalus psilocentros* (C)General View (D) Surface Sculpturing

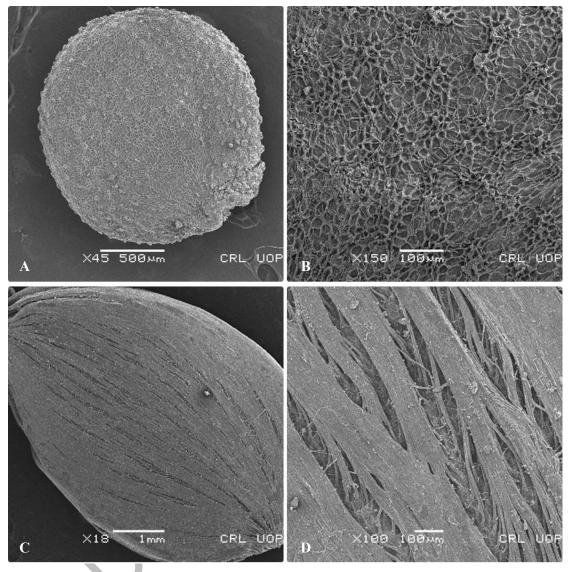


Plate 136: Seed micrographs of *Astragalus scoripiurus* (A) General View (B) Surface Sculpturing , *Cassia fistula* (C)General View (D) Surface Sculpturing

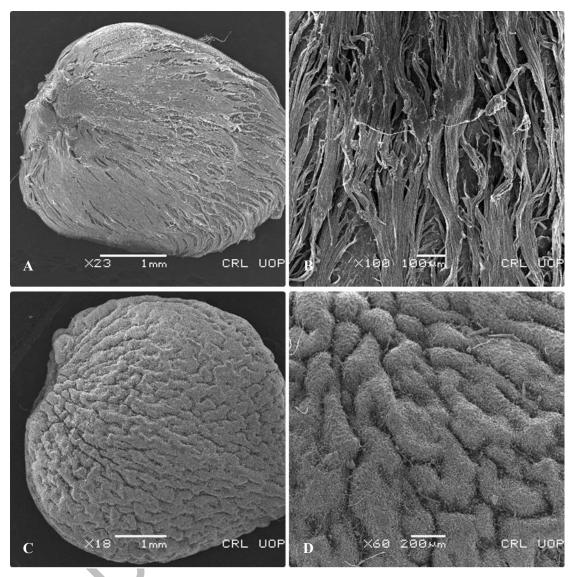


Plate 137: Seed micrographs, *Cassia italic* (A) General View (B)Surface Sculpturing, *Cassia occidentalis* (C) General View (D) Surface Sculpturing

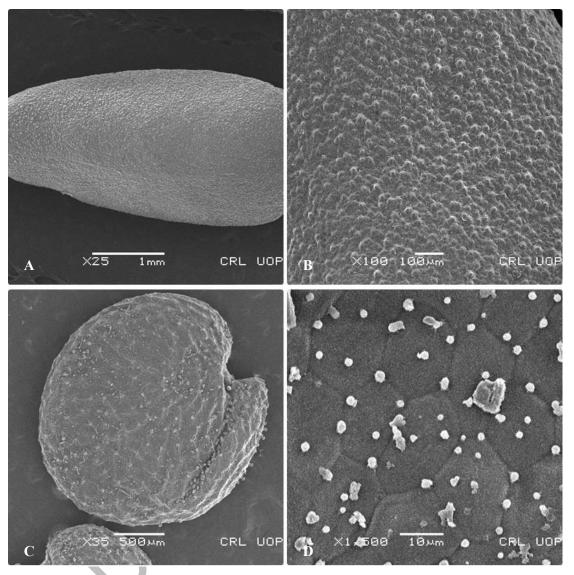


Plate 138: Seed micrographs of *Crotolaria burhia* (A) General View (B) Surface Sculpturing , *Dilbergia sisso* (C) General View (D) Surface Sculpturing

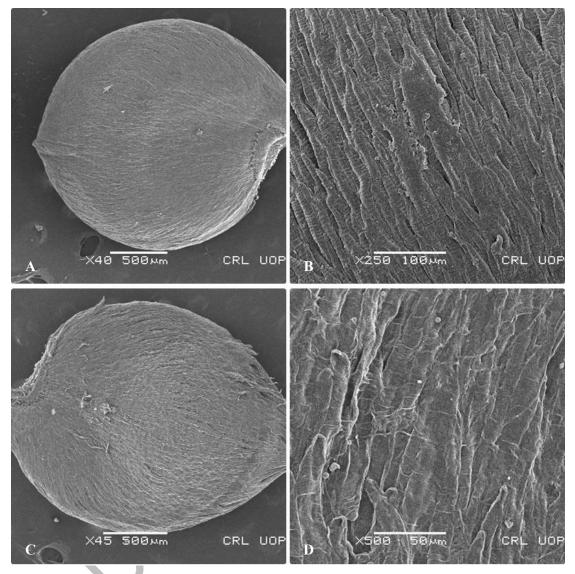


Plate 139: Seed micrographs of *Lathyrus aphaca*, (A) General View (B) Surface Sculpturing , *Lespedeza juncea* (C) General View (D) Surface Sculpturing

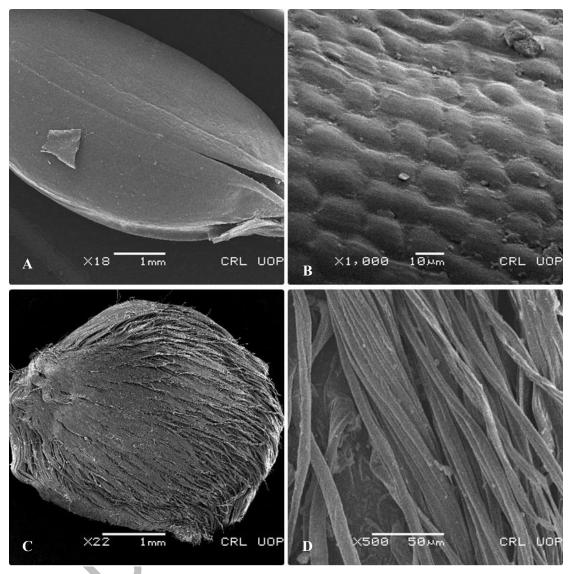


Plate 140: Seed micrographs of *Leucaena leucocephalla* (A) General View (B) Surface Sculpturing , *Melilotus indicus* (C)General View (D) Surface Sculpturing

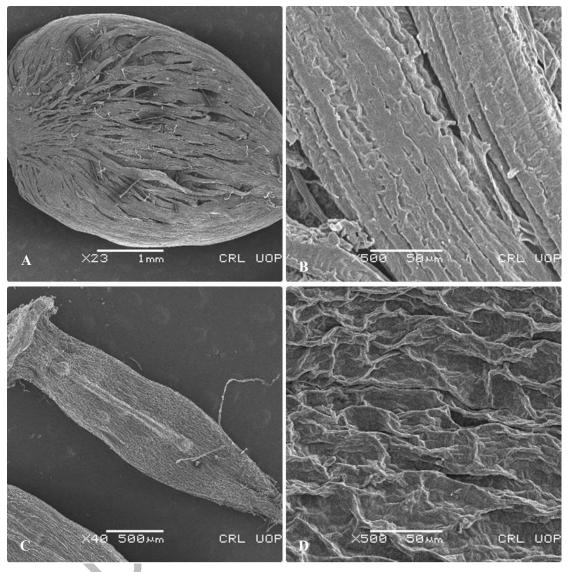
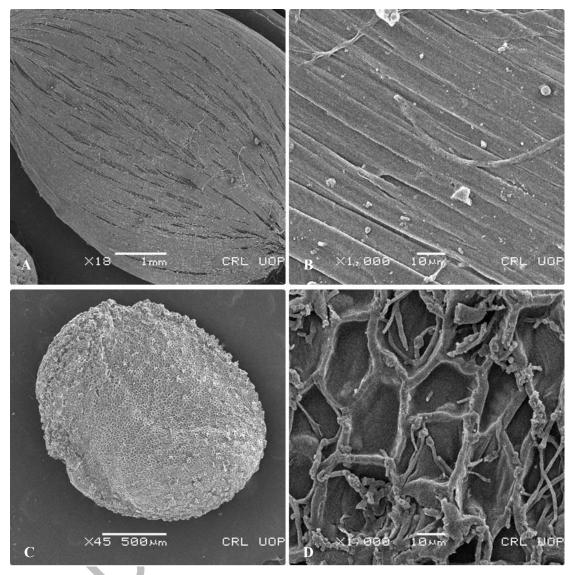


Plate 141: Seed micrographs *Pigomia pinnata* (A)General View (B) Surface Sculpturing , *Prosopis cineraria* (C)General View (D) Surface Sculpturing



**Plate 142:** Seed micrographs of *Prosopis juliflora* (A) General View (B) Surface Sculpturing , *Pueraria tuberose* (C) General View (D) Surface Sculpturing

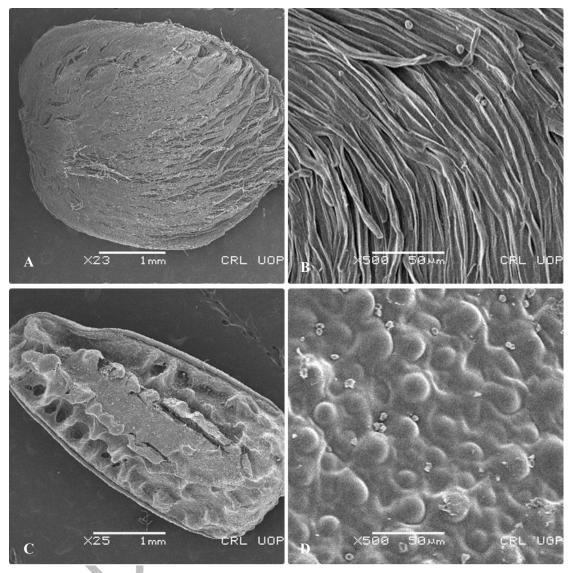


Plate 143: Seed micrographs of *Rhynchosia capitata* (A) General View (B) Surface Sculpturing , *Sophora mollis*(C) General View (D) Surface Sculpturing

**Taxonomic key of seed morphological characters of Fabaceaous Species** 2a. Seed apex is flat ..... Prosopis juliflora 3a. Seed outline is irregular ..... Acacia nilotica 3b. Seed outline is regular ...... 4 4a. Seed shape is ovate ..... Cassia fistula 5a. Epidermal cell shape is radiated ..... Accacia modesta 5b. Epidermal cells are flat or round ..... 6a. Cell of epidermis are flat ..... Arachis hypagae 6b. Cell of epidermis are round ..... 7a. Seed shape is oblong ...... ..... Astragalus scoripiurus 8a. Anti clinal wall pattern is smooth ...... Lathyrus aphaca 9a. Seed sculpturing is slightly striated ...... Dilbergia sisso 9b. Seed sculpturing is pappilate ..... Prosopis cineraria 10a. Seed sculpturing is smooth ..... Cassia occidentalis 10b. Seed is sculptured with pattern ...... 11 11a. Seed is bean shaped ..... Crotolaria burhia 11b. Seed shape is other than bean shaped ...... 12 12 a. Anticlinal wall pattern is in visible ..... Astragalus psilocentros

## 3.2.2 Fabaceaous Seeds

This study describes the seed morphological characters of 20 Fabaceaous Plants. Seeds display variation in features like shape, texture, color, sculpturing, testa wall, epidermal cell shape and presence or absence of papillae.

Seeds morphology provide useful characters for investigating systematic affinities in various plant families (Bus et.al; 2001) Gontcharova et.al;2009, Zhang et.al 2005). The seed morphology as well as sculpturing and texture attributes were observed to vary greatly among different taxa and can play important role in systematic plant studies. (Al-Ghoray and Mohamed, 2007, Chowdhary and Buth, 1970). The outer surface features of seeds studied by scanning electron Microscopy(SEM), have been extensively applied in resolving taxonomic ambiguities within plant groups and evolutionary relationships(Koul et.al 2000. Sagarra and Mateu, 2001). The ultra structural pattern of the seed coat examined under the SEM is known as trustworthy tool to evaluate phonetic affinities and delimitation of Plant taxa (Barthlott, 1981; Koul et.al, 2000; Yoshizaki. 2003).

## 1 Seed Shape and Size

Seed shape varies from oval to roundin majority of studied taxa with few exceptions including flat shaped seed in *Argyrolobium roseum, Astragalus, astragalus psilentros* and bean shaped in Acacia nilotica, Accacia modesta, Arachis hypagae .Seed outline appears ovate or oblong in most of the species with addition of elliptic, round and irregular seed outline in few species. Two types of seed texture are recorded i.e smooth (9 species) Hairy (11 species) . Seed display less variation in color ranging from gray (6 species) , grayish black (7 species) or dark gray(7 species) . Seed size varies from 1-15 mm.

Previous studies on seed micromorphology and surface structure specify that seed characteristics are valuable in the systematics of Fabaceous species (Gune and Ali 2011; Lersten, 1979).Taia, (2004b) studied the seed morphological characters in genus Lythrus and noticed oval shaped seeds thus supporting the present findings. IICIM AND Behcet,(2016), described sub-reniform seed shape in three Astragalus species in Turkey. This studied also reported elliptic and oblong seed outline in genus Astragalus. Similarly members of genus Trigonella were observed to be globoseobovoid in outline in earlier literature (Muuml et.,2012). These findings corroborate with recent results reporting seeds of Trigonella foenumgraceum and Trigonella monantha with oblong outline.

Even though the shape and size of seeds are reasonably consistent among the species examined, yet some features have been recognized as distinctive like seed size in Cassia italic and Cassia occidentalis are small (1-4mm) in comparsion to other species. Vural et.al;(2008) and Saghi et.al; (2015) observed oblate-sub-oblate and ovoid-reniform seed shape in Crotolaria burhia, Dilbergia sisso, Lespedeza juncea thus supporting the recent results. Abid et.al; (2018) studied the seed micromorphology in genus Melilotus and reported elliptic seed shape. These findings corroborate with findings presented in this study. Similarly, Lazarevic et.al;(2017) studied the seed morphology in field pea genotypes and noticed spherical or round seed shape which supports the result of present study. Comparative seed morphological features of Trifolium taxa was studied by Zoric et.al; (2010) describing round-ovoid seeds which is in line with the results presented in the current study. The members of genus vicia are reported with round shaped seed which is confirmed by reviewing earlier study of Hosseinzadeh et.al;(2008) and Lersten,(1979) reporting spherical shaped seeds inVicia species.

# 2 Seed Sculpturing

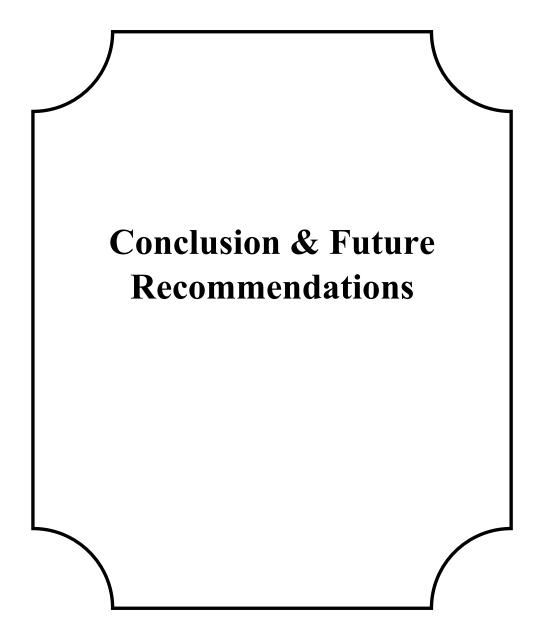
Seed sculpturing pattern display great variation in studied taxa. On the basis of seed coat surface features, the studied plant species are separated into five groups. (i) Species with papillate sculpturing pattern (ii) Species having reticulate seed sculpturing (iii) Species with striate seed sculpturing (iv) Species with granulate surface pattern (v) Species having smooth seed coat surface.

Acacia nilotica, Leucaena leucocephalla, Prosopis cineraria, Sophora mollis are observed with Papillate surface sculpturing.. Papillae are described as protuberances appearing from the testa cells. In Leucaena leucocephalla Papillae come into view as prominent, simple, and often showing a very complex pattern toward the apex. Intra specific inconsistency of this characteristics is observed in Posopis as both conditions exist in genus. In Leucaena leucocephalla, simple and small simple papillae are noticed close to the notch. While recorded mainly on the sides of the seed members of genus except Pigomia pinnata. Earlier studied carried out to study the variation in seed coat morphology by using SEM techniques, documented practical morphological and structural features of seed comprising the pattern of testa cell arrangement (Clark and Jamstedt, 1978) Cell shape (Canne,1979) radial wall outline as well as Papilleae distribution (Carolin 1980). The current study supports the use of seed structural features as descrete characters used in delimitation of plant taxa. Heyn and Hermstdt , (1977) revealed differences in surface characters of Sophora seed using SEM techniques and concluded seed morphology can prove significant in systematics.

In tribe Phoseoleae ,Sharma et.al; (1977) also used SEM in investigating seed structural pattern in distinguishable the plant species within the genus Phoseoleus. Similarly Gutterman and Heydecker, (1973) reported that the seed surface characters play an important role in delimitation of genus Ononis Species. A distinct seed coat pattern was reported in tribe Genisteae descriing its importance in delimitation of Plant species and genera within this tribe (Polhill,1976) . In tribe Vicia Lerstern (1979) observed a reduced rim of hilum and conspicuous papillose pattern of ornamentation reporting these features characteristics for this tribe. The Striate testa pattern in genus Lythrus and Cicer, the reticulate pattern in Vicia hirsute are new characters for the tribe.

The papillose testa pattern reported in literature in Vicieae (Lersten, 1979; Chernoff et.al, 1992; Lersten and Gunn,1982), has also been noticed this study in Astragalus scoripiurus, Astragalus psilocentros, Cassia fistula,. The study carried out by Abou-EI-Enain et.al; (2007) on the seed surface features in genus Lythrus described the anticlinal wall pattern in detail. About nine multi-cellular ornamentation patterns and three secondary ornamentation patterns had been recorded with stellate like structures present on the sides of the anticlinal walls. These are similar with the fndings of the recent study with the little difference in sculpturing pattern. A similar study on the seed morphology of various species of tribe vicie was carried out by khalik and Al-Ghiory (2013) and noticed that the secondary sculpturing pattern in seed coat wall consists of papillose pattern in majority of Vicia species with exception of Vicia hirsute bearing lophate ornamentation pattern. These findings are in agreement with the study under discussion reporting papillose seed ornamentation in Genus Vicia. Regarding the Phenomenon of Polymorphism in the vicia about 50% of the taxa exhibit fairly dissimilar seed texture thus confirming the high rate of Polymorphism in tribe than observed in previous studies. Even inside the general grouping, a distinct discrepancy has been recorded reagarding papillae distribution and significant for systematic delimitation. For example in genus Acacia and Lhythrus large papilleae are observed wheras in genus Sophara generally small papilleae noticed. The characteristics testa features observed Pueraria tuberosa make it distinct from rest of the studied vetch taxa. These findings are in harmony with the earlier study of smykal et.al (2014). The members of Vicia are distinct from rest of genera in Vicieae by bearing small papilleae. The presence of a characteristics striate testa pattern in Sophora mollis can prove helpful since taxonomic ambiguities exist in this genus and its addition and elimination from Vicieae is debatable till now.

Javadi and Yamaguchi,(2004) documented three different types of sculpturing patterns (tuberculate, granulate-pappilate & reticulate) in thirteen species of genus Astragalus. This study also describes that Astragalus displayed significant seed morphological features distinctive from rest of genera in Vicieae. Similar findings were reported on monotypic genus Astragalus by Kupicha,(1977). The common sculpturing pattern is illustrated by presence of elongated cells in majority of Plant species but disparity observed in epidermal cells shape as well as density provides additional data important for identification and classification of the taxa. In Vicieae radiate-wavy cells are observed generally on seed surface while some species such as Arachis hypagae, Astragalus psilocentros, Astragalus scoripiurus, are noticed with curved, irregular, and round shaped cells respectively. The degree of differentiation exhibited by species in testa cells possibly reveals their characteristics configuration pattern (smykal et.al,2014). The variation observed in seed morphological features in this study may prove helpful at species, genus and tribe level.



## Conclusion

This is the first comprehensive documentation with respect to taxonomic characteristics (LM & SEM) of floral diversity of 103 species belonging to 15 families from Potohar-Plateau Pakistan. It is concluded from this study that palynological, Seed micromorphological features are very helpful in the identification and delimitation of flora of Potohar Plateau.

# Palynology

The Pollen attributes of the 103 species were helpful in the comparison among the species, the statistical analysis revealed significant information about the resemblance among the species. Asteraceae was the most dominating family representing 29 species revealed different types of exine stratification such as smooth sparsely granulate, scabrate-spinolose, micro-spinolose, micro-spinolose perforate, meta-reticulate, nano-spinulate, granulate-spinulose perforate, and granulate-perforate echinate. Exine thickness was highest in *Biden psilosa* 2.17um and lowest in *Eclipta prostrate* 0.65um. Taxonomic keys were constructed based on findings that highlight the importance of the micromorpholgical ultrastructural diversity of Pollen.

# Seed Morphology

The Present study has revealed that utilization of SEM proved to be a significant attribute in the study of seed morphology of family Asteraceae and Fabaceae as SEM exposed the surface sculpture pattern and epicuticular projection types of Seeds. Previously, surface sculpture pattern of Seeds was given less or no importance but present studied showed that major variations among the studied Asteraceous and Fabaceaus species was observed for surface pattern and epicuticular projection types of Seeds. Hence the present study offers valuable diagnostic characters for morphological characterization of Seeds. Moreover, use of advance microscopic techniques is recommended to study internal features of Seeds, as it can be helpful to establish phylogenetic relationship among the taxa of Asteraceae and Fabaceae.

# **Future Recommendations**

• The establishment of district based regional floras is immediately required in Potohar Plateau for Eco-taxonomist, botanist and herbalist to protect the potohar Plateau biodiversity of plants for sustainable utilization.

- Necessary to conserve the plant resources of Potohar plateau and to document the associated indigenous knowledge for the welfare of mankind.
- The relationship of different species and genera of the selected family may be explained by phylogenetic studies in future in Potohar Plateau.
- Awareness program at gross root level should be introduced in the potohar plateau to educate the local people. There is a need to provide technical assistance and training for proper and sustainable collection of Plants.
- Efforts are required to identify the endemic, endangered and threatened flora of potohar plateau in order to highlight the ways of their conservation.
- It is suggested that endangered plants of Potohar Plateau may be cultivated and promoted through the In Situ and Ex situ practices.

Phytodiversity using Microscopic Characterization in Potohar Plateau-Pakistan



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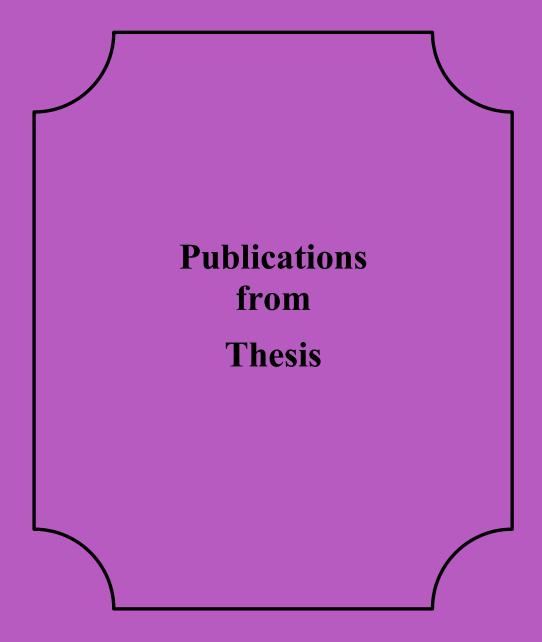
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## Article Palynological Study of Weed Flora from Potohar Plateau

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Abstract: The pollen morphology of weeds was investigated by scanning electron microscopy (SEM). A morpho-palynological investigation of 18 species of weeds that belongs to 16 angiosperms families was performed using SEM to document distinguishable microscopic features. The main objective of the present study was to provide basic knowledge about morpho-palynological features of weed species that helps delimit the weed flora of the Potohar Plateau. The results show diversity among the qualitative and quantitative characteristics of pollen shape, equatorial and polar axis diameter, the exine's thickness, and the exine's surface ornamentation. The pollen grains were spherical, prolate-spheroidal, oblate-spheroidal, and sub-oblate. The exine ornamentation in most species was reticulate, scarbate, aerolate, faveolate, reticulate-perforate, and reticulate-scabrate. All the species described possessed tricolpate pollen. The variations found in the thickness of the exine and other characters were helpful at the genus and species-specific levels. In accordance with these variations, a taxonomic key was prepared using these characteristics to identify and differentiate weed plant species. SEM images of pollen grains can help delimit the taxa to the species level. This study provides baseline information to distinguish the species of weeds.

Keywords: exine sculpturing; palynomorph; pollen grains; scanning electron microscopy; Pakistan

#### 1. Introduction

Weeds are considered undesirable plants competing with crops' growth and development. The growth of weeds in agricultural fields can limit the production of staple crops and causes severe damage to their production [1]. According to Khan et al. [2] weeds grow in cultivated and wild habitats and damage the growing crop plants [3]. Weedy plants threaten human health, welfare, biodiversity, ecosystem services, and food security. Here, weeds are defined as any plants that have detrimental socioeconomic and environmental effects, endanger human health, and jeopardize global food security, biodiversity, and ecosystem services [4]. Weeds play an important role in achieving an ecological balance in a cropping system by supporting different life forms. Throughout the world, major vegetation of weeds belongs to families such as Asteraceae, Poaceae, Amaranthaceae, and Fabaceae. The growth of weeds depends mostly on climatic conditions [5]. In some previously reported studies, the most commonly grown weeds of cotton crops are Cynodon dactylon, Amaranthus viridis, Phyla nodiflora, and Phragmites australis [6]. Common weeds of wheat crops are *Phragmites* sp., *Cynodon* sp., *Ranunculus* spp., and *Polypogon* sp. [6]. Similarly, Achyranthes aspera is a common weed in sugarcane crops and Plantago lanceolata is an important weed in chickpea fields [7]. Weeds belonging to the family Fabaceae can



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#### RESEARCH ARTICLE



# <u>CHRITECHNIQUE</u> VVIL.

# Micro-morphological diversity of pollen among Asteraceous taxa from Potohar Plateau-Pakistan

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#### Abstract

Microscopic pollen characters from 19 species of family Asteraceae collected from different areas of the Potohar Plateau of Pakistan have been investigated using light and scanning electron microscopy. This study uses microscopic techniques to provide detailed information on pollen and its exine structure based on qualitative and quantitative characters. The present study has observed three types of pollen grains based on their apertures, that is, trizonocolporate, tetrazonocolporate, and tricolporate. Exine sculpturing is echinate in all studied taxa. Some species also exhibit Lacunae which differ in their shape and sizes. The size of the spines and the number of spines between the colpi play a vital role in differentiating the species of the same genus. Pollen fertility ranges from 94.3 to 74.19, showing that these species are well established in the study area. For the correct identification of species, a light and scanning electron microscopy was used that provides sufficient information for a taxonomist to distinguish species taxonomically.

#### **Research Highlights**

- Light and Scanning electron microscope is comparatively used to investigate the pollen characters of some species of the family Asteraceae.
- A scanning electron microscopy is used to study the micro-morphological characters of pollen grains.
- Qualitative and quantitative characters of pollen were studied.

#### KEYWORDS

Asteraceae, echinate, Pakistan, pollen morphology, Potohar Plateau

#### 1 | INTRODUCTION

Potohar Plateau is situated northeast of Pakistan with approximately 25,000 Square kilometers; topographically, the area includes hilly and plain areas. The area is situated between attitude 32.534°N and longitude 71.3073°E. The lowest altitude from sea level is Jhelum plain 250 m (825 ft), and the crop production depends upon monsoon rains (Rashid and Rasul, 2011). Potohar Plateau is a densely populated area with residents from different cultures, frequently introducing exotic plants. The construction of houses, roads, and industries limits the range of natural vegetation through land use, leading to soil composition changes. To increase greenery in the study area, foreign plants are being introduced intentionally for afforestation. The native flora of the Potohar region consists of dry deciduous scrubby vegetation and few tree species.

Family Asteraceae consist of 1530 genus and approximately 2300 species distributed within 3 subfamilies and 17 tribes (Anar et al., 2022; Hayat et al., 2009; Khan et al., 2021). It is the largest family of angiosperms and is cosmopolitan in distribution. For example, in Pakistan, this family comprises 650 species distributed in 15 tribes

## **Publications Submitted**

- Pollen diversity of Poaceous Taxa using microscopy in Potohar Plateau Pakistan. (Under-Review in NASB Journal Elsevier)
- Pollen morphological characters of tribe Cichorieae (Asteraceae) and its systematic implications (Submitted in JAPS)
- Pollen Morphology and Taxonomic Relations among the Genera of Amaranthaceae from Potohar-Plateau.(Under-Review in Plants Journal)