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THE SEED ATLAS OF PAKISTAN-VI. CARYOPHYLLACEAE

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Abstract

Seed micro morphological characters of 59 species, distributed in 21 genera of the family Caryophyllaceae were studied using light and scanning electron microscopy. There is a large variety of seed characters such as seeds reniform, sub reniform and rarely elliptic pyriform, cuneate, transversely cuneate, ovate, oblong, orbicular or sub orbicular, size ranges from 0.4-3mm x 0.3-3mm. Seed morphological data is found to be slightly significant for the tribal delimitation, while it strongly supports the generic and specific delimitations in the family Caryophyllaceae.

Introduction

The family Caryophyllaceae is a large dicotyledonous family comprises 3 subfamilies viz., Paronychioideae, Alsinoideae and Caryophylloideae and 11 tribes (Bittrich,1993) with 85 genera and 2630 cosmopolitan species (Mabberley, 2008).The family Caryophyllaceae is represented in Pakistan by 26 genera and 110 species (Ghazanfar & Nasir, 1986) excluding the representatives of the tribe Paronychieae of the subfamily Paronychioideae which were previously treated under the family Illeceberaceae (Ghafoor, 1973). The genera *Spergulla, Spergularia* and *Polycarpea* are placed in the tribe Polycarpeae, while the genera *Cometes, Herniaria* and *Pteranthus* are included in the tribe Paronychieae of the subfamily Paronychioideae, similarly the genera *Arenaria, Cerastium, Leprodiclis, Minuartia, Myosoton, Holosteum, Stellaria* and *Sagina*, are included in the tribe Alsineae of the subfamily Alsinoideae. Moreover, the genera *Dianthus, Gypsophila, Petrorhagia, Saponaria* and *Vaccaria* fall in the tribe Caryophylleae, while the two genera *Cucubalus* and *Silene* are placed in the tribe Sileneae of the subfamily Caryophylloideae (Bittrich, 1993). Seed morphological studies have been found useful and provide additional information for solving various taxonomic and evolutionary problems (Bergreen, 1981; Akbari & Azizian, 2006; Abid & Ali, 2010; Rajbhandary & Shrestha, 2010; Ackin & Binzet, 2011). Previously Crow (1979) studied 15 species of the genus *Sagina* and concluded that the different seed shapes can be utilized for sectional and specific delimitation in the genus *Sagina*. Bergreen (1981) studied the seed morphology of Northwest European plants belonging to the family Caryophyllaceae and utilized seed characters in the classification of taxa at various levels. However, Wofford (1981) examined the seed morphology of the genera *Arenaria* and *Minuartia* and concluded that seed morphology was not helpful at the generic level but found useful for specific delimitation. Yildiz (2002) while studying the seed morphology of different genera of Caryophyllaceae viz., *Agrostema, Cerastium, Dianthus, Gypsophila, Lychnis, Minuartia, Moenchia, Petrorhagia, Silene, Stellaria* and *Velezia* demonstrated the utility of seed characters for the generic and specific delimitation. The seeds of the genus *Silene* were studied and this data was utilized for specific delimitation (Yildiz, 2006; Perveen, 2009; Fawzi, 2010). Although there are number of reports available on seed morphology of different taxa of the family Caryophyllaceae from various parts of the world, but no detailed reports are available on seed morphology for the entire family from Pakistan. The purpose of the present report is two fold first is to provide the seed morphological data as an additional information for the taxonomic delimitation of the family Caryophyllaceae and secondly for designing a Seed Atlas of Pakistan.

Materials and Methods

Mature and healthy seeds of 59 species belonging to the family Caryophyllaceae were collected from herbarium specimens. Mostly 20 seeds per plant and 10 plants per species were studied. The list of voucher specimens is deposited in KUH. Seed morphological characters were examined under light microscope (Nikon Type 102) and scanning electron microscope (JSM-6380A).For scanning electron microscopy dry seeds were directly mounted on metallic stubs using double adhesive tape and coated with gold for a period of 6 minutes in a sputtering chamber and observed under SEM. The terminology used is in accordance of Lawrence (1970), Bergreen (1981) and Stearn (1983) with slight modifications. The following characters were studied: size, colour, shape, surface (testa), surface plate size, plate margin and position of hilum.

General seed characters of the family Caryophyllaceae: Seeds 0.4-3 x 0.3-3mm, angular or non angular, reniform- sub-reniform, elliptic pyriform, cuneate, transversely cuneate, ovate, oblong, globose, sub globose, light brown, brown, dark brown, chocolate brown, grey, greyish black, orange brown, maroom brown, rust brown or black, shiny or un shiny, surface smooth, rugose, ruminate, granulate, ridged, reticulate, puncticulate, sparsely tuberculate, papillate , or covered with plates, plate margin entire, serrate, serrulate, crenate, undulate or dentate , 45-254 x 12-104 µm, with smooth, granulate or rugose surface. Hilum marginal, central, sub-central, basal, sub-basal, on ridge or indistinct (Table 1).

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Table 1. Seed morphological characters of the family Caroyphyllaceae.

Name of taxa	Size (mm)	Shape	Colour	Surface	Plate size(µm)	Hilum
Arenaria leptoclados	0.9-1 (±0.034) x 0.9-1 (±0.034)	Reniform	Brown	Covered with plates, margin crenate	123-176 x 27-36	Sub-central
A. neelgherrensis	0.5-0.7(±0.04) x 0.4-0.5 (±0.024)	Reniform	Brown	Depressed centrally, covered with plates, margin serrate	90.95.4 x 16-32	Sub-central
A. orbiculata	0.6-0.7(±0.04) x 0.4-0.5 (±0.04)	Reniform	Brown	Covered with linear-star shape plates, margin undulate, plate surface granulate & tuberculate at margin	47-85.3 x 19-20	Sub-central
A. serpyllifolia	0.4-0.5(±0.04) x 0.4-0.5 (±0.04)	Reniform	Chocolate brown-brown	Covered with linear-round plates, margin crenate plate surface appressedly granulate	125 x 25	Sub-central
Cerastium cerastioides	0.7-1 (±0.05) x 0.4-1 (±0.11)	Elliptic pyriform	Pale yellow-brown	Covered with plates, margin papillate	90-91 x 48-56	Sub-central
C. dichotomum	0.8-1 (±0.04) x 0.8-1 (±0.03)	Elliptic pyriform	Brown	Covered with plates, depressed centrally, margin smooth	73-79.7 x 61-65	Sub-central
C. fontanum subsp. *triviale*	0.5-1 (±0.27) x 0.4-0.7 (±0.07)	Angular & cuneate	Brown	Covered with plates, margin papillate	70-81.8 x 41-42	Sub-basal
C. glomeratum	0.5-1 (±0.27) x 0.4-0.7(±0.07)	Angular & cuneate	Brown	Covered with plates, margin papillate	78.3-81.4 x 22.7-27.6	Sub-basal
C. pusillum	0.8-1 (±0.04) x 0.6-0.8(±0.03)	Angular & cuneate	Brown	Covered with plates, margin papillate	90-92 x 40-55	Sub-basal
C. thomsoni	0.7-1 (±0.05) x 0.4-0.5 (±0.04)	Angular & cuneate	Brown	Covered with plates, margin papillate	75-162 x 14-21	Sub-basal
C. tomentosum	1-1.5(±0.08) x 0.9-1(±0.034)	Elliptic pyriform	Brown	Covered with plates, depressed centrally, margin smooth	93-120 x 15.4 x 30.2	Sub-basal
Cometes surattensis	2.5-3 (±23) x 1.5-2 (±0.08)	Elliptic, beaked basally, punctate at one side	rust brown, black at punctation unshiny	rugose, granulate	-	basal
Cucubalus baccifer	1.5-2 (±0.08) x 0.8-1(±0.03)	Reniform	Black & shiny	Smooth	-	Sub-central
Dianthus anatolicus	2-3 (±0.18) x 1.2-2(±0.15)	Ovate and ridged dorsally	Dark brown & shiny	Covered with plates, margin serrulate, plate surface appressedly granulate	92-97 x 30-37	On ridge
Gypsophila alsinoides	1-1.5 (±0.08) x 0.8-1(±0.03)	Elliptic pyriform	Light brown-black, shiny	Covered with plates, margin dentate, plate surface granulate	95-130 x 19.7-20	Sub-central
G. bellidifolia	0.8-1.5(±0.12) x 0.5-1 (±0.27)	Reniform-transversely cuneate	Dark brown-black & shiny	Covered with plates, margin serrate, plate surface smooth	145-205 x 31-45.6	Central- sub-central
G. cerastioides	0.9-1 (±0.034) x 0.9- 1 (±0.034)	Transversely cuneate	Dark brown & shiny	Covered with plates, margin smooth, plate surface granulate	76-87 x 18-32.5	Central- sub-central
G. floribunda	0.9-1 (±0.034) x 0.6-0.7 (±0.02)	Angular & elliptic pyriform	Dark brown & shiny	Covered with plates, margin serrate, plate surface granulate	92-160 x 30-35	Sub-central
G. makranica	1-1.5 (±0.08) x 0.8-1 (±0.03)	Broadly reniform	Light brown & shiny	Covered with plates, margin serrulate, plate surface granulate	93.8-105 x 33-34.5	Central- sub-central
Herniaria cinerea	0.8-1 (±0.03) x 0.5-0.7 (±0.02)	Triangular	light brown-dark brown, unshiny	ridged and colliculate	-	lateral

Table 1. (Cont'd.).

Name of taxa	Size (mm)	Shape	Colour	Surface	Plate size(µm)	Hilum
H. cachemiriana	0.8-1 (±0.03) x 0.5-1 (±0.27)	obovate	maroom brown -dark brown, shiny	smooth	-	lateral
H. hirsuta	0.8-1 (±0.03) x 0.5-0.7 (±0.02)	obovate	dark brown, shiny	faintly reticulate	-	lateral
Holosteum umbellatum	0.9-1 (±0.034) x 1-1.5 (±0.08)	Oblong, dorsally grooved & ventrally ridged	Light brown & shiny	Covered with plates, margin papillate	106-200 x 37.9-64	On ridge
Leprodiclis holosteoides	1.5-2 (±0.08) x 1.5-2 (±0.08)	Angular & elliptic pyriform	Dark brown	Covered with plates, margin dentate	210-227 x 48-66	Sub-central
L. stellarioides	1-1.5 (±0.08) x 1-1.5 (±0.08)	Angular & reniform	Dark brown	Covered with plates, margin dentate	161-243 x 56-80.8	Sub-central
L. tenera	1-1.5 (±0.08) x 0.9-1 (±0.34)	Broadly reniform	Grey	Covered with plates, margin dentate	191-220 x 59-100	Sub-central
Minuartia biflora	0.9-1 (±0.34) x 0.4-0.5 (±0.04)	Sub-reniform	Brown	Covered with irregular plates, surface granulate	Not prominent	Sub-central
M. hybrida	0.8-1 (±0.03) x 0.5-0.8 (±0.05)	Reniform	Brown	Covered with plates, depressed centrally, margin smooth	Not prominent	Central
M. kashmirica	0.9-1 (±0.34) x 0.5-0.8 (±0.05)	Reniform	Brown	Covered with plates, margin serrulate	57-65 x 20-30	Central
M. meyeri	0.9-1 (±0.34) x 0.9-1 (±0.34)	Reniform	Brown	Covered with plates, margin serrate, surface smooth	57-65 x 25-33	Sub-central
Myosoton aquaticum	0.8-1 (±0.03) x 0.8-1 (±0.03)	Reniform	Chocolate brown-black	Covered with plates, margin serrate, surface smooth	145-180 x 54-60	Sub-central
Petrorhagia alpina	0.8-1 (±0.03) x 0.4 0.5 (±0.04)	Ovate, compressed & ridged on ventral side	Dark brown & shiny	Covered with plates, margin papillate, surface faintly granulate	45-65 x 12-13	On ridge
Polycarpea corymbosa	0.4-0.5(±0.04) x 0.4-0.5 (±0.04)	Sub-reniform	Brown & shiny	Rugose	-	Sub-basal
P. spicata	0.5-1 (±0.27) x 0.5-1 (±0.27)	Elliptic & grooved ventrally	Brown-dark brown & shiny	Rugose	-	Sub-basal
Pteranthus dichotomus	2-2.5 (±0.08) x 0.8-1 (±0.03)	Obovate, beaked basally	Cream with dark brown spot at base	Rugose	-	basal
Sagina saginoides	0.4-0.5(±0.04) x 0.2-0.3 (±0.02)	Triangular	Light brown	Covered with plates, margin dentate	46-48 x 15-16	Indistinct
Saponaria griffithiana	1-1.5 (±0.08) x 1-1.5 (±0.08)	Reniform	Dark brown & shiny	Covered with plates, margin smooth	136-165 x 50-63	Central-sub central
S. subrosularis	1-2 (±0.05) x 1-1.5 (±0.03)	Broadly reniform	Dark brown & shiny	Covered with plates, margin serrate	253-254 x 51.5-55.7	Marginal
Silene arenosa	0.5-0.8(±0.05) x 0.4-0.6 (±0.03)	Angular & reniform	Grey-brown	Covered with plates, margin crenate, plate surface rugose	115-131 x 23-28	Central
S. brahuica	1.5-1.7(±0.04) x 0.8-1 (±0.03)	Angular & reniform	Grey	Covered with plates, margin serrulate, plate surface appressedly granulate	208-215 x 60.9-75.8	Central

Table 1. (Cont'd.).

Name of taxa	Size (mm)	Shape	Colour	Surface	Plate size(µm)	Hilum
S. citrina	0.8-1 (±0.03) x 0.8-1 (±0.03)	Angular & reniform	Greyish black	Surface centrally depressed,covered with plates, margin serrate	136-151 x 29.2-44	Central
S. coeli-rosea	0.8-1 (±0.03) x 0.5-0.7 (±0.02)	Angular & reniform	Brown	Covered with plates, margin serrulate surface appressedly granulate	160-170 x 41-59	Central
S. conoidea	1-1.5 (±0.08) x 0.8-1 (±0.03)	Angular & reniform	Grey	Covered with plates, margin serrulate, surface smooth	180-204 x 60.9-68	Central
S. falconeriana	1-1.5 (±0.08) x 0.5-0.7 (±0.02)	Angular & reniform	Brown	Surface centrally depressed,Covered with plates,margin serrate, surface rugose	122-131 x 50-70	Central
S. kunawarensis	1-1.5 (±0.08) x 0.9-1 (±0.34)	Angular & reniform	Brown	Covered with plates, margin serrate, surface granulate	122-128 x 34-35	Central-sub central
S. longisepala	1.3-1.5(±0.04) x 0.7-0.8 (±0.02)	Angular & reniform	Grey	Covered with plates, margin denate, surface granulate	176-216 x 74-79	Central
S. moorcroftiana	0.8-1 (±0.03) x 0.8-1 (±0.03)	Angular & reniform	Greyish black	Covered with plates, margin papillate, surface granulate		Sub-central
S. nana	0.8-1 (±0.03) x 0.6-0.7 (±0.02)	Angular, reniform-transversely cuneate	Black	Covered with plates, margin serrate, surface granulate	102-105 x 56.3-58	Sub-central
S. pseudo-verticellata	1-1.5 (±0.08) x 0.8-1 (±0.03)	Angular & reniform	Black	Covered with plates, margin crenate, surface rugose	206-250 x 34.3-37.2	Central
S. tenuis	0.8-1 (±0.03) x 0.8-1 (±0.03)	Angular & reniform grooved at back	Orange brown	Covered with plates, margin serrate surface appressedly granulate	174-176 x 40-49	Sub-central
S. vulgaris	0.8-1 (±0.03) x 0.5-0.8 (±0.05)	Angular & sub reniform	Brown	Covered with plates, margin undulate surface granulate	70-71 x 35.4-43.2	Sub-central
Spergula fallax	1-1.5 (±0.08) x 1-1.5 (±0.08)	Elliptic pyriform & winged	Black & shiny	Puncticulate & sparsely tuberculate	-	Basal
Spergularia diandra	0.4-0.5(±0.04) x 0.3-0.5 (±0.05)	Cuneate	Light brown	Appressedly papillate & granulate	-	Sub-central
S. marina	0.5-0.7(±0.02) x 0.4-0.5 (±0.04)	Elliptic pyriform	Light brown	Papillate & granulate	-	Sub-central
S. media	1-1.5 (±0.08) x 0.8-1 (±0.03)	Elliptic pyriform & winged	Light brown	Ruminate & granulate	-	Indistinct
Stellaria kotschyana subsp. *glabra*	1.5-2 (±0.08) x 2.5-3 (±0.23)	Broadly reniform	Brown	Covered with plates, depressed centrally margin smooth-undulate, surface smooth	120-179 x 52-81.5	Central
S. media	1-1.5 (±0.08) x 0.8-1 (±0.03)	Broadly reniform	Dark brown	Covered with plates, margin papillate, surface granulate	187-195 x 75-104	Central
S. montioides	0.4-0.6(±0.02) x 0.5-1 (±0.27)	Broadly reniform	Brown	Covered with plates, margin papillate-dentate, surface smooth	223-240 x 31-36	Central
Vaccaria hispanica	1.5-2 (±0.08) x 1.4-1.5 (±0.04)	Angular & globose-sub globose	Dark brown & shiny	Covered with linear-round plates, margin crenate, surface rugose	87-140 x 42-90.3	Indistinct

Key to the genera

1 + Seeds smooth ……. *Cucubalus*
- Seeds not smooth …….. 2
2 + Seeds beaked basally …… 3
- Seeds not beaked ……... 4
3 + Seeds elliptic, surface granulate …….. *Cometes*
- Seeds obovate, not granulate ……. *Pteranthus*
4 + Seeds reniform-transversely cuneate, elliptic pyriform, cuneate or triangular …… 5
- Seeds ovate, globose-sub-globose or oblong ……..……..... 14
5 + Seed surface covered with plates ……..……..…….. 6
- Seed surface not covered with plates ……..……..…….. 10
6 + Seeds triangular ……..…… *Sagina*
- Seeds not angular ……. 7
7 + Seeds shiny ……... 8
- Seeds not shiny ……..……..……... 9
8 + Surface plates granulate ……..……..…. *Gypsophila*
- Surface plates smooth ……..…. *Saponaria*
9 + Seeds cuneate-elliptic pyriform ……..... *Cerastium*
- Seeds reniform-sub reniform or transversely cuneate ……..……..…
……... *Silene, Arenaria, Minuartia, Stellaria, Leprodiclis, Myosoton*
10 + Seeds elliptic-sub reniform ……... *Polycarpea*
- Seeds triangular, obovate elliptic pyriform or cuneate ……..……..…. 11
11 + Seeds with lateral hilum ……..……..……. *Herniaria*
- Seeds with basal hilum ……..…… 12
12 + Seeds black and shiny ……..……..…….. *Spergulla*
- Seeds light brown and not shiny ……..……..…... *Spergularia*
13 + Seeds globose-sub globose or oblong ……..……..... 14
- Seeds ovate ……..……..……..…… 15
14 + Seeds globose-sub globose, angular ……..... *Vaccaria*
- Seeds oblong, non-angular ……..……..... *Holosteum*
15 + Seeds 0.8-1 x 0.4-0.5 mm, surface plates with papillate margin ..……..... *Petrorhagia*
- Seeds 2-3 x 1-2 mm, surface plates serrulate margin ……..……..……... *Dianthus*

Arenaria L.

Seeds 0.4-1 x0.4-1mm, angular, reniform, centrally depressed or not, brown-chocolate brown, unshiny surface coverd with plates, 47-176x16-36µm, plate surface granulate-tuberculate, margin crenate, serrate or undulate, hilum sub-central (Table 1; Figs. 1 A-I).

It is represented by 4 species viz., *A. leptoclados* (Reichb.) Guss., *A. neelgherrensis* Wight & Arn., *A.orbiculata* Royle ex Edgew., *A. serpyllifolia* L.

Key to the species

1 + Seeds depressed centrally ……... *A. neelgherrensis*
- Seeds not depressed centrally ……..……..……... 2
2 + Surface plates tuberculate at margins …… *A. orbiculata*
- Surface plates not tuberculate at margins ……..…….. 3
3 + Seeds 0.4-0.5 x 0.4-0.5 mm ..…… *A. serpyllifolia*
- Seeds 0.9-1 x 0.9-1 mm ..…… *A. leptoclades*

Cerastium L.

Seeds 0.5-1.5x0.4-1 mm, angular or non-angular, cuneate or elliptic pyriform, brown or yellowish brown, unshiny surface covered with plates, depressed centrally or not, 70-120x14-65 µm, margin smooth or papillate, hilum sub–basal or sub-central (Table 1; Figs. 1J-R, 2A-G).

It is represented by 7 species viz., *Ceratium cerastioides* (L.) Britton., *C. dichotomum* L., *C. fontanum* Baumg., *C. glomeratum* Thuill., *C.pusillum* ser. *C. thomsoni* Hook. f., *C. tomentosum* L.

Key to the species

1 + Seeds angular,cuneate ..…… *C. fontanum ssp. trivale, C. glomeratum, C. pusillum, C. thomsonii*
- Seeds non-angular, elliptic pyriform ……..……... 2
2 + Seeds brown, surface plates with smooth margin ……..……... 3
- Seeds yellowish brown, surface plates with papillate margin ..…… *C. cerastioides*
3 + Hilum sub-central ……..…….. *C. dichotomum*
- Hilum sub-basal ……..…… *C. tomentosum*

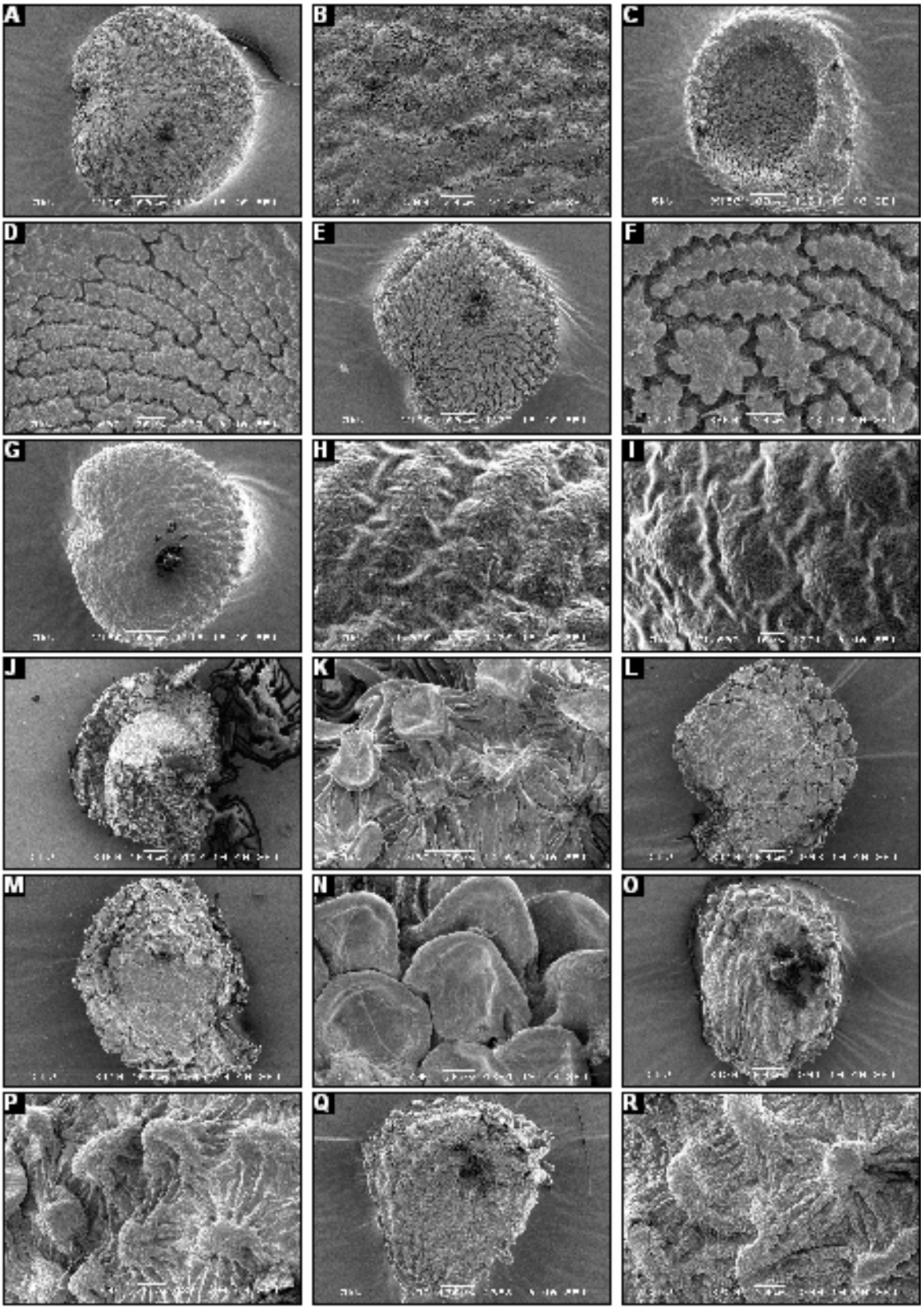

Fig. 1. Scanning electron micrographs. *Arenaria leptoclados*: A, seed; B, surface. *A.neelgherrensis*: C, seed; D, surface. *A.orbiculata*: E, seed; F, surface. *A. serpyllifolia*: G, seed; H,I, surface. *Cerastium cerastioides*: J, seed; K, surface. *C.dichotomum*: L, M, seeds; N, surface. *C.fontanum*: O, seed; P, surface. *C. fontanum* subsp.*trivale*: Q, seed; R, surface. (Scale bars: A, C, E, G, J, L, M, O, Q=100µm; K=50µm; B, D, F, N, P, R=20µm; H,I=10µm).

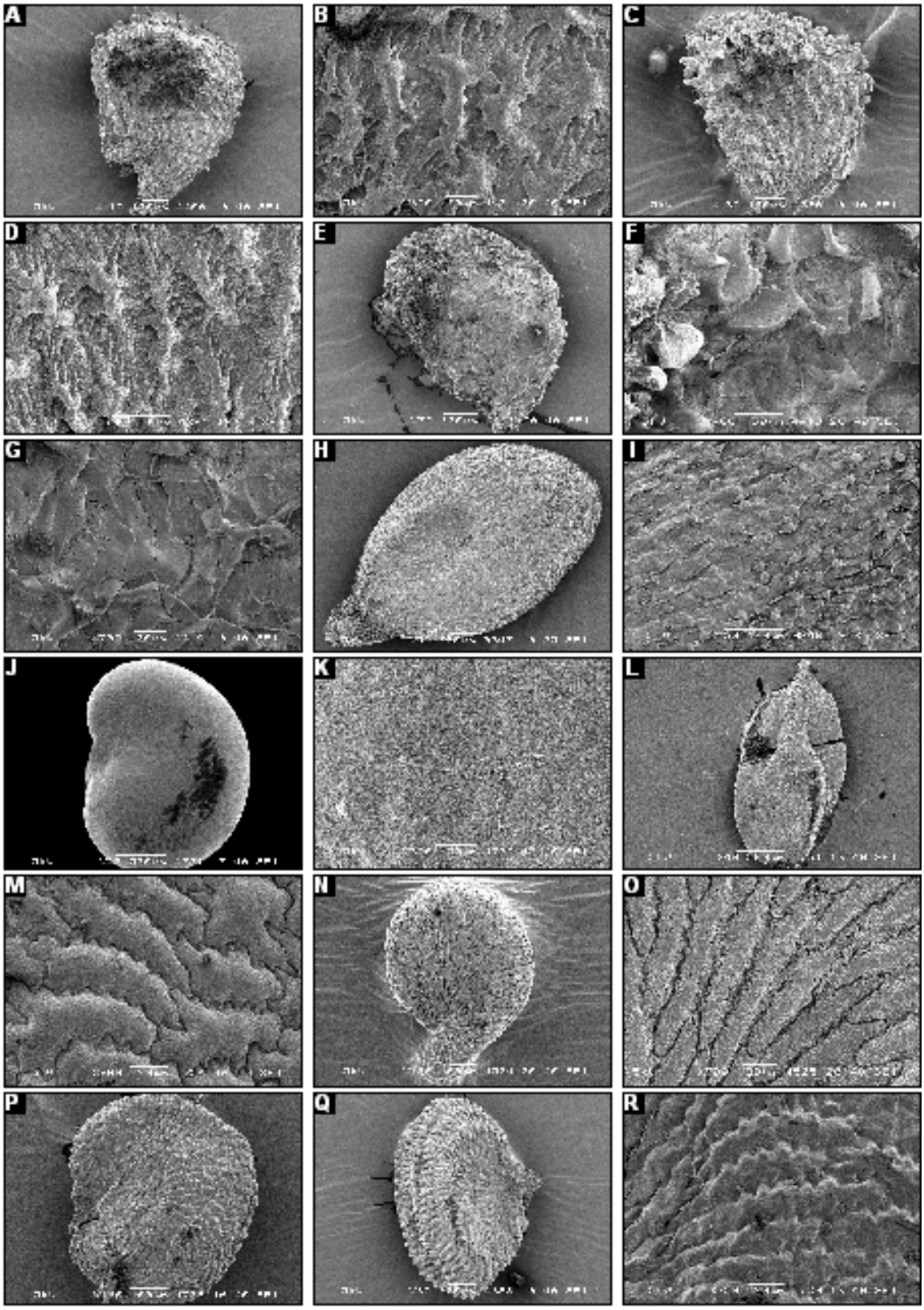

Fig. 2. Scanning electron micrographs: *C.glomeratum*: A, seed; B, surface. *C. pusillum*: C, seed; D, surface. *C.thomsonii*: E, seed; F,G, surface. *Cometes surattensis:* H, seed: I, surface. *Cucubalus baccifer*: J, seed; K, surface. *Dianthus anatolicus*: L, seed; M, surface. *Gypsophila alsinoides*: N, seed; O, surface. *G.bellidifolia*: P, Q, seeds; R, surface. (Scale bars: J, L=500; E, H, Q=200µm; A, C, I, N, P=100µm; D, F, K=50µm; B, G, M, O, R=20µm).

Cometes L.

Seeds 2.5-3 x 1.5-2 mm, elliptic, beaked basally, punctuate at one side, rust brown, black at punctuation, unshiny, surface rugose and granulate, hilum basal (Table 1; Fig. 2H-I). It is represented by a single species viz., *C. surattensis* L.

Cucubalus L.

Seeds 1.5-2x0.8-1mm, reniform , black and shiny, smooth, hilum sub-central (Table 1; Fig. 2J-K). It is represented by a single species viz., *Cucubalus baccifer* L.

Dianthus L.

Seeds 2-3x1.2-2mm, ovate, ridged at dorsal side, dark brown and shiny, surface covered with plates, margin serrulate, 92-97 x 30-37 µm, surface appressedly granulate, hilum on ridge (Table 1; Fig. 2L-M). It is represented by a single species viz., *D. antolicus* Boiss.

Gypsophila L.

Seeds 0.8-1.5 x 0.5-1mm angular, elliptic pyriform, reniform or transversely cuneate, light brown, dark brown or black and shiny, surface covered with plates, plate margin smooth, serrate, serrulate or dentate, 76-205x18-45.6 µm surface smooth or granulate hilum central or sub-central (Table 1; Figs. 2N-R, 3A-G).

It is represented by 5 species viz., *Gypsophila alsinoidies* Bunge, *G .bellidifolia* Boiss., *G. cerastioides*, D. Don, *G. floribunda* (Kar. & Kir) Turcz, *G. makranica* (Rech.f.) S.A. Ghazanfar.

Key to the species

1 + Seeds elliptic pyriform ……..……..…….... 2
- Seeds reniform-transversely cuneate ……...……. 3
2 + Surface plates with dentate margin ……..……..... *G. alsinioides*
- Surface plates with serrate margin ……...……...….. *G. floribunda*
3 + Seeds light brown, surface plates with serrulate margin ....……. *G. makranica*
- Seeds dark brown-black, surface plates with smooth or serrate margin …...…….……... 4
4 + Plate surface granulate with smooth margin ……..……. *G. cerastioides*
- Plate surface non-granulate with serrate margin ……..……..... *G. bellidifolia*

Herniaria L.

Seeds 0.8-1 x 0.5-1 mm, obovate, triangular, light brown, dark brown, maroon brown, shiny or unshiny, surface smooth, ridged, colliculate, reticulate, hilum lateral (Table 1; Fig. 3H-M). It comprises 3 species viz., *H. cachemiriana* J. Gay, *H. cinerea* DC and *H. hirsuta* L.

Key to the species

1 + Seeds obovate ….……. 2
- Seeds triangular ..…… *H. cinerea*
2 + Seeds smooth ….……..… *H. cachemiriana*
- Seeds faintly reticulate ……... *H. hirsuta*

Holosteum L**.**

Seeds 0.9-1x1-1.5 mm, oblong, dorsally grooved and ventrally ridged, light brown, and shiny, surface covered with plates,106-200 x 37.9-64 µm, surface smooth, margin papillate, hilum on ridge (Table 1; Fig. 3N-Q).

It is represented by a single species viz., *Holosteum umbellatum* L.

Leprodiclis Fenz L.

Seeds 1-2 x 0.9-2 mm, angular, reniform or elliptic pyriform, grey-dark brown, unshiny, surface covered with plates, 161-243 x 48-100µm, plate margin dentate, surface smooth, hilum sub-central (Table 1; Figs. 3R, 4A-E).

It is represented by 3 species viz., *Leprodiclis holosteoides* (C.A. Meyer), *L. stellarioides* F. & M., *L. tenera* Boiss.

Key to the species

1 + Seeds grey ……..…….. *L. tenera*
- Seeds dark brown ……..……..……..……..…… 2
2 + Seeds elliptic pyriform ……..……..……. *L. holosteoides*
- Seeds reniform ...……..……... *L. stellarioides*

Minuartia L.

Seeds 0.8-1 x 0.4-1mm, reniform-subreniform, brown, unshiny, seeds centrally depressed or not, surface covered with plates, 57-65 x 20-30 µm , surface granulate or non-granulate, margin smooth, serrulate or serrate, hilum central-sub central (Table 1; Fig. 4F-K).

It comprises of 4 species viz., *Minuartia biflora* (L.) Schinz & Thell., *M. hybrida* (Vill.) Schischkin, *M. kashmirica* (Edgew.) Mattf., *M. meyeri* (Boiss.) Bornm.

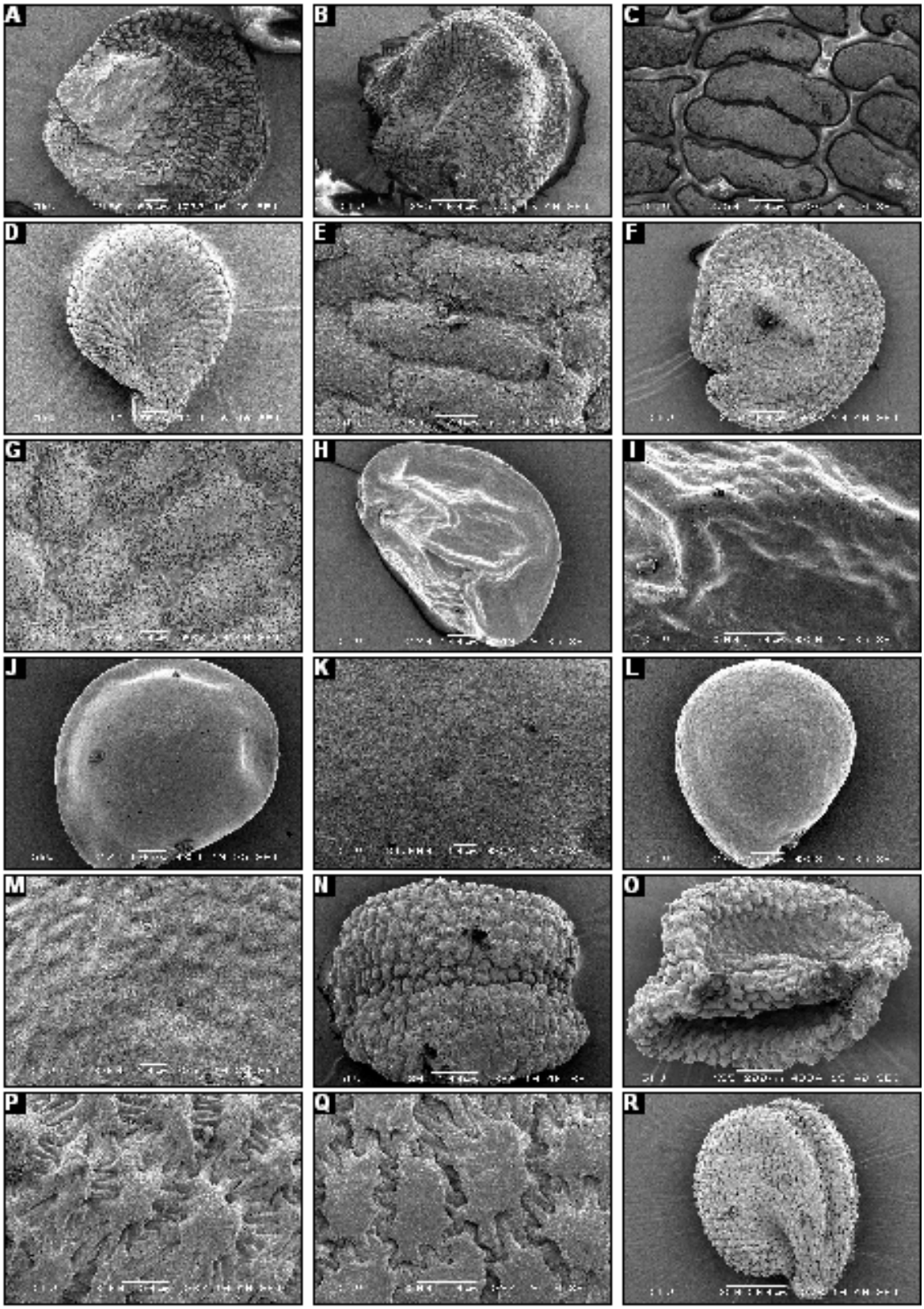

Fig. 3. Scanning electron micrographs: *G. cerastioides*: A, B, seeds; C, surface. *G. floribunda*: D, seed; E, surface. *G.makaranica*: F, seed; G, surface. *Herniaria cinerea*: H, seed; I, surface. *H.cachemiriana*: J, seed; K, surface. *H.hirsuta*: L, seed; M, surface. *Holosteum umbellatum* var. *umbellatum*: N, O, seeds; P, Q, surface. *Leprodiclis holosteoides*: R, seed. (Scalebars: R=500µm; B, F, N, O=200µm; A, D, H, J, L=100µm; I, P, Q=50µm; C, E, G, M=20µm; K=10µm).

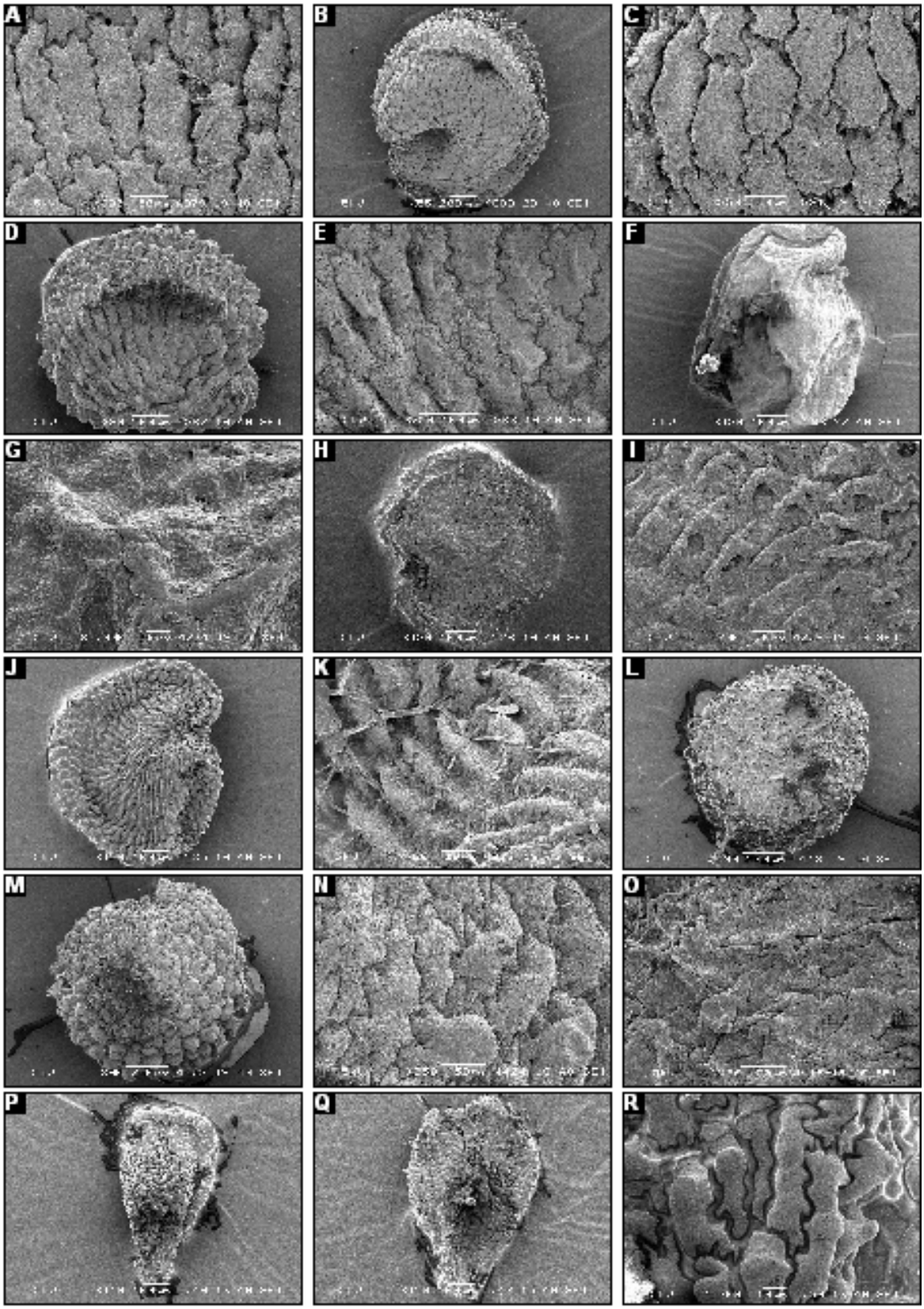

Fig. 4. Scanning electron micrographs: *Leprodiclis holosteoides*: A, surface. *L.stellarioides*: B, seed; C, surface. *L. tenera*: D, seed; E, surface. *Minuartia biflora*: F, seed; G, surface. *M. hybrida*: H, seed; I, surface. *M. kashmirica*: J, seed; K, surface. *Myosoton aquaticum*: L, M, seeds; N, O, surface. *Petrorhagia alpina*: P, Q, seeds; R, surface. (Scale bar: B, D, L, M=200µm; E, F, H, J, P, Q=100µm; A, C, N, O=50µm; I=20µm; G, K, R=10µm).

Key to the species

1 + Hilum central ……..…….…….....…….. 2
- Hilum sub-central ……..…….…….. 3
2 + Seeds centrally depressed, surface plates with smooth margin ……..……..…..…… *M. hybrida*
- Seeds not centrally depressed, surface plates with serrulate margin ……..……. *M. kashmirica*
3 + Surface plates granulate, arranged in regular pattern ……. *M. biflora*
- Surface plates smooth, arranged in regular pattern ……...……. *M. meyeri*

Myosoton Moench

Seed 0.8-1x 0.8-1 mm, reniform, chocolate brown-black, unshiny, surface plates 145-180 x 54-60 µm, surface smooth, margin serrate, hilum sub-central (Table 1; Fig. 4L-M).

It is represented by a single species viz., *Myosoton aquaticum*(L.) Moench

Petrorhagia (Ser. ex DC.) L.

Seeds 0.8-1x0.4-0.5mm, ovate, compressed, ventrally ridged, dark brown and shiny, surface plates 45-65 x 12-13 µm, suface faintly granulate,margin papillate, hilum on ridge (Table 1; Fig. 4P-R)).

It is represented by a single species viz., *Petrorhagia alpina* (Habl.) Ball & Heywood

Polycarpea Lam.

Seeds 0.4-1 x 0.4-1mm, elliptic-sub reniform, brown-dark brown, shiny, surface rugose, hilum sub-basal (Table 1; Fig. 5A-F)).

It comprise of 2 species viz., *Polycarpea corymbosa* (L.) Lam., and *P. spicata* Wight & Arn.

Key to the species

1 + Seeds elliptic ……..……..... *P. spicata*
- Seeds sub reniform ……..……..…..... *P. corymbosa*

Pteranthus Forssk

Seeds 2-2.5 x 0.8-1 mm, obovate and beaked basally, cream with dark brown spot at base, rugose, hilum basal (Table 1; Fig. 5-G-H). It comprises of single species *P.dichotomus* Forssk.

Sagina L.

Seeds 0.4- 0.5 x0.2- 0.3mm, triangular, light brown, surface covered with plates, 46-48 x 15-16 µm, margin dentate, hilum indistinct (Table 1; Fig. 5I-J).

It is represented by a single species viz., *Sagina saginoides* (L.) Karst.

Saponaria L.

Seeds 1-2 x 1-1.5 mm, reniform, dark brown and shiny, surface plates 136-254 x 50-63 µm, margin smooth or serrate, hilum marginal or central-sub central (Table 1; Fig. 5K-O).

It comprises of 2 species viz., *Saponaria griffithiana* Boiss., and *S. subrosularis* Rech. f.

Key to the species

1 + Surface plates with smooth margin, hilum central-sub central ……..….. *S. griffithiana*
- Surface plates with serrate margin, hilum marginal ……..…..... *S. subrosularis*

Silene L.

Seeds 0.5-1.7x0.4-1mm, angular, reniform or transversely cuneate, grey, brown, greysih black , orange brown or black, unshiny, surface covered with plates, 70-250 x 23-79 µm, margin serrate, serrulate, crenate, dentate, papillate or undulate, plate surface granulate or rugose, hilum central-sub central (Table 1; Figs. 5P-R, 6A-R, 7A-G).

It comprises of 13 species viz., *Silene arenosa* C. Koch, *S. brahuica* Boiss., *S. citrina* Boiss., *S. coeli-rosea* (L.) Godron, *S. conoidea* L., *S. falconeriana* Benth., *S. kunawarensis* Benth., *S. longisepala* E. Nasir, *S. moorcroftiana* Wall. ex Benth., *S. nana* Kar & Kir., *S. pseudo-verticellata* E. Nasir, *S. tenuis* Willd., and *S. vulgaris* (Moench) Garcke.

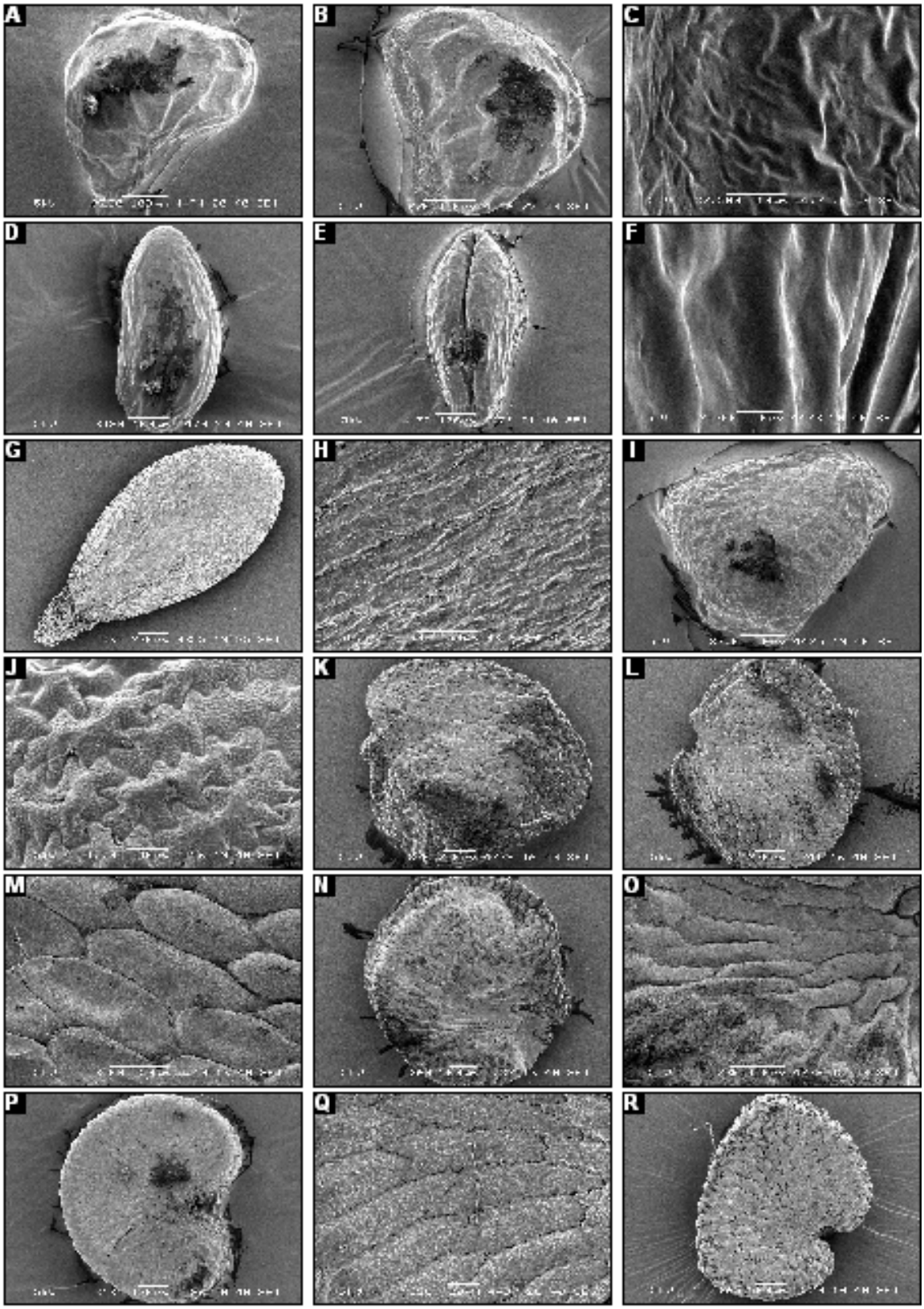

Fig. 5. Scanning electron micrographs: *Polycarpea corymbosa*: A, B, seeds; C, surface. *P. spicata*: D, E, seeds; F, surface. *Pteranthus dichotomus*: G, seed; H, surface. *Sagina saginoides*: I, seed; J, surface. *Saponaria griffithiana*: K, L, seeds; M, surface. *S. subrosularis*: N, seed; O, surface. *Silene arenosa*: P, seed; Q, surface. *S.brahuica*: R, seed. (Scale bars: G, K, L, N, R=200µm; A, B, D, E, O, P=100µm; H, I, M=50µm; Q=20µm; C, F, J=10 µm).

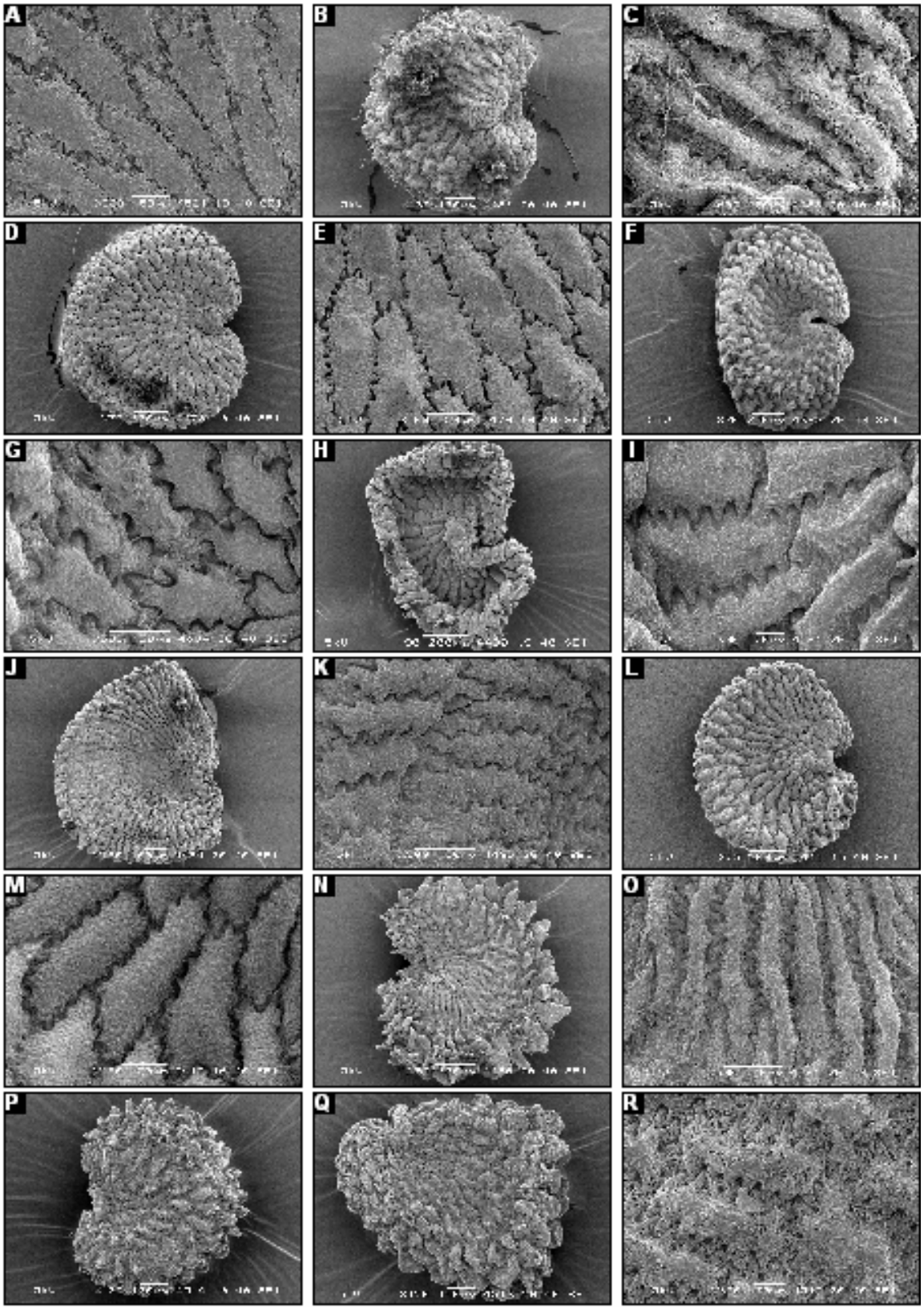

Fig. 6. Scanning electron micrographs: *S.brahuica*: A, surface. *S.citrina:* B, seed; C, surface. *S.coeli-rosea*: D, seed; E, surface. *S.conoidea*: F, seed; G, surface. *S. falconeriana*: H, seed; I, surface. *S. kunawarensis*: J, seed; K, surface. *S. longisepala*: L, seed; M, surface. *S. moorcroftiana*: N, seed; O, surface. *S. nana*: P, Q, seeds; R, surface. (Scale bars: D, F, H, L, N=200µm; B, J, P, Q=100µm; A, E, G, K, M, O=50µm; C, I, R=20µm).

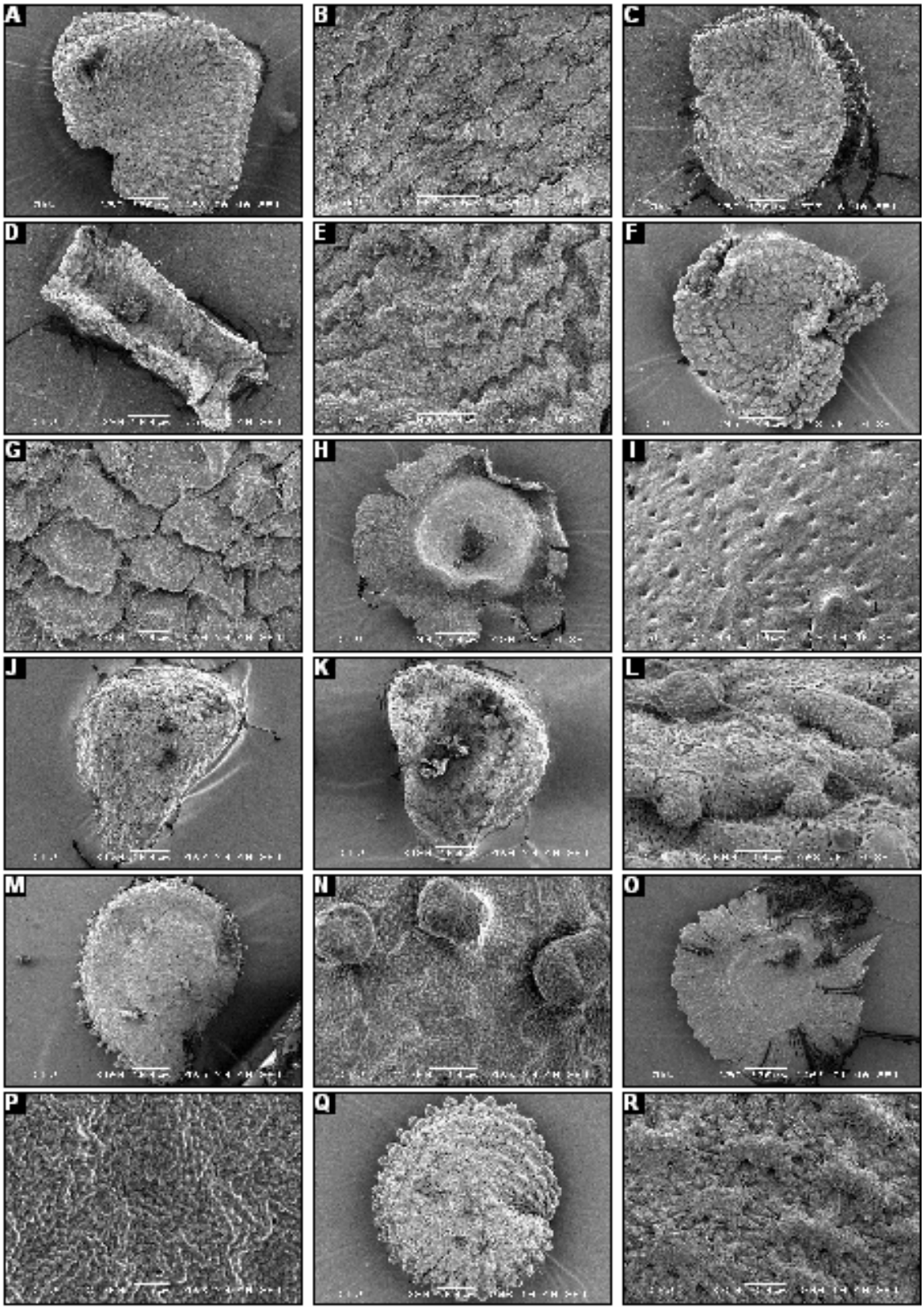

Fig. 7. Scanning electron micrographs: *S. pseudoverticellata*: A, seed; B, surface. *S. tenuis*: C, D, seeds; E, surface. *S. vulgaris*: F, seed; G, surface. *Spergula fallax*: H, seed; I, surface. *Spergularia diandra*: J, K, seeds; L, surface. *S. marina*: M, seed; N, surface. *S. media*: O, seed; P, surface. *Stellaria kotschyana* subsp. *glabra*: Q, seed; R, surface. (Scale bars: A, C, D, F, H, O, Q=200µm; J, K, M=100µm; B, E, R=50µm; G=20 µm; I, L, N=10µm; P=5µm).

Key to the species

1 + Seeds greyish black or black ……..... 2
- Seeds grey, brown or orange brown ……..…. 5
2 + Hilum central ……...……...…… 3
- Hilum sub-central ……...……..... 4
3 + Seeds black, plate surface rugose, margin crenate …….. …… *S. pseudo-verticellata*
- Seeds greyish black, plate surface not rugose, margin serrate ..…..…… *S. citrina*
4 + Seeds greyish black, plate margin papillate ……...…..…… *S. moorcroftiana*
- Seeds black, plate margin serrate ……...…….……..... *S. nana*
5 + Seeds orange brown, grooved at back ……...……...……..... *S. tenuis*
- Seeds grey-brown, not grooved at back ……...……...……..... 6
6 + Surface plates granulate ……...……...…..……...…….. 7
- Surface plates non-granulate ……...……...……...…….. 11
7 + Plates with serrate, serrulate or undulate margin ……..... 8
- Plates with dentate margin ……...……..... *S. longisepala*
8 + Seeds grey ……...……..…… *S. brahuica*
- Seeds brown ……...……...……. 9
9 + Plates with serrulate margin ……..……..……..……... *S. coeli-rosea*
- Plates with serrate or undulate margin ……...…….....… 10
10 + Plates with serrate margin ……..……... *S. kunawarensis*
- Plates with undulate margin ……...……...…..…… *S. vulgaris*
11 + Plates rugose, margin crenate-serrate ……...……...……..…… 12
- Plates smooth, margin serrulate ……..……..……...…… *S. conoidea*
12 + Plates 23-28 µm broad, margin crenate ……...……...…… *S. arenosa*
- Plates 50-70 µm broad, margin serrate ……...…….. *S. falconariana*

Spergulla L.

Seeds 1-1.5 x 1- 1.5 mm, winged, elliptic pyriform, black and shiny, surface puncticulate and sparsely tuberculate, hilum basal (Table 1; Fig. 7H-I).

It is represented by a single species viz., *Spergula fallax* (Lowe) E.H.L.

Spergularia J. &C. Presl

Seeds 0.4-1.5x 0.3-1mm, winged or without wing, elliptic pyriform or cuneate, light brown, sunshiny surface ruminate-granulate or papillate-granulate, hilum sub-central or indistinct (Table 1; Fig. 7J-P).

It comprises of 3 species viz., *Spergularia diandra* (Guss.) Heldr. & Sart., *S. marina* (L.) Griseb. and *S. media* (L.) Presl.

Key to the species

1 + Seeds winged ……...……. *S. media*
- Seeds not winged ……...……..… 2
2 + Seeds elliptic pyriform, surface ruminate-granulate ……...…… *S. marina*
- Seeds cuneate, papillate-granulate ……...……..… *S. diandra*

Stellaria L.

Seeds 0.4-2 x 0.5-3mm, reniform, brown-dark brown, unshiny, surface plates 120-240 x 31-104 µm, plate surface granulate or smooth, margin smooth, undulate, dentate or papillate, plates depressed centrally or not, hilum central (Table 1; Figs. 7Q-R, 8A-E).

It comprises 3 species viz., *Stellaria kotschyana* ssp., *glabra* Fenzl. ex Boiss., *S. media* (L.) Vill., *S. montioides* (Edgew. & Hook.f.) S.A. Ghazanfar.

Key to the species

1 + Seeds 3mm broad, surface plates with smooth- undulate margin ...…… *S. kotschyana* ssp., *glabra*
- Seeds 0.5-1.5 mm broad, surface plates with dentate-papillate margin 2
2 + Plates granulate ……...……...……. *S. media*
- Plates non-granulate ……...……...……. *S. montioides*

Vaccaria Med.

Seeds 1.5-2 x 1.4-1.5 mm, angular and globose-sub globose, dark brown and shiny surface covered with linear-round plates, margin crenate, surface rugose 87-140 x 42-90.3 hilum indistinct (Table 1; Fig. 8F-H).

It is represented by a single species viz., *Vaccaria hispanica* (Miller) Rauschert.

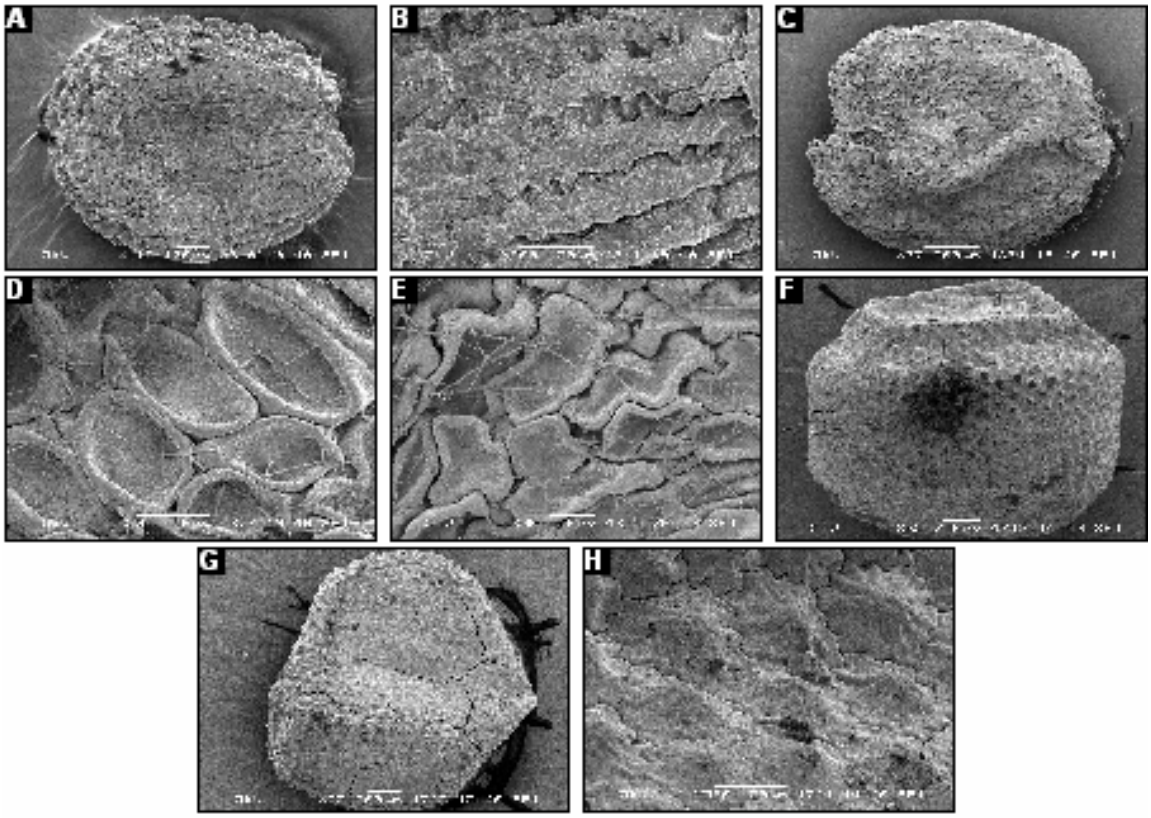

Fig. 8. Scanning electron micrographs: *Stellaria media*: A, seed; B, surface. *S. montioides*: C, seed; D, E, surface. *Vaccaria hispanica*: F, G, seed; H, surface. (Scale bars: C=500 µm; F, G=200 µm; A=100 µm; B, D, E, H=50 µm).

Results and Discussion

The seed morphological data is significant enough to correlate the taxonomic delimitation of the family Caryophyllaceae at various levels. The subfamily Paronychioideae remains distinct from rest of the two sub families by having stipulate leaves and dorsal subapical appendiculate sepals (Bittrich, 1993) and seeds without specific surface plates. Bergreen (1981) used the term "Armadillo plates" for these ornamentations and this specific feature was found to be very useful for generic and specific delimitations. However, most of the previous and some recent workers did not give any attention to this specific feature and they had just used the terms tuberculate, sulcate, striate or colliculate for these plates (Wofford, 1981; Ghazanfar & Nasir, 1986; Yildiz, 2002, 2006; Ataslar & Ocak, 2005; Perveen, 2009; Fawzi *et al.*, 2010).The remaining two subfamilies viz., Alsinoideae and Caryophylloideae are characterized by having exstipulate leaves and sepals without dorsal sub apical appendages and seeds are with specific surface plates except that of the genus *Cucubalus* of the tribe Sileneae where psilate seeds are observed. Beside this, the genus *Cucubalus* also remains distinct from rest of the Caryophylloid genera by having indehiscent berry (Ghazanfar & Nasir, 1986; Bittrich, 1993). Similar to that of the palynological findings (Perveen & Qaiser, 2003, 2006) the two sub families could not be further separated on the basis of seed character alone. While the genera of the subfamily Paronychioideae viz., *Spergulla*, *Spergularia* and *Polycarpea* of the tribe Polycarpeae, and the genera *Cometes* and *Pterantus* of the tribe Paronychieae were grouped together into *Spergulla arvensis*-type with 3-10 colpate pollen grains except that of the genus *Herniaria* where porate pollen grains were found. Presently the generic delimitation of the family Caryophyllaceae is well correlated with seed morphology as the genus *Holosteum* can be easily distinguished from rest of the genera by having umbel inflorescence (Ghazanfar & Nasir, 1986; Bittrich, 1993) and ventrally ridged oblong seeds, while the remaining genera have cymes or solitary flower. Among them *Cerastium* can be distinguished from other genera due to the presence of cylindrical capsule and cuneate-elliptic pyriform seeds, while the remaining genera viz., *Arenaria*, *Leprodiclis*, *Minuartia*, *Myosoton*, *Stellaria* and *Silene* have globose-ovoid capsule (Ghazanfar & Nasir, 1986) and reniform-sub reniform or transversely cuneate seeds. The genus *Myosoton* and *Sagina* can be coupled due to the presence of 4-5 styles but they can be further separated as lanceolate-spathulate leaves (Ghazanfar & Nasir, 1986) and reniform, sub-reniform or elliptic pyriform seeds are observed in the genus *Myosoton*. While the genus *Sagina* have linear leaves (Ghazanfar & Nasir, 1986; Bittrich, 1993) and triangular seeds, these findings are also supported by the earlier findings of Crow (1979) where triangular seeds were also reported. The genus *Vaccaria* remains distinct due to the presence of winged calyx (Ghazanfar & Nasir, 1986) and globose-sub globose seeds, similarly, the two genera *Dianthus* and *Petrorhagia* can be coupled due to the presence of non winged calyx (Ghazanfar & Nasir, 1986) and ovate seeds, both these

genera remains distinct from each other due to presence of bracteoles (Ghazanfar & Nasir, 1986) and different seed size and plate margin.

Similar to that of the generic delimitation seed morphological characters are also found useful for the specific delimitation. The genus *Silene* is the largest genus of the family Caryophyllaceae, seed surface plates of the genus *Silene* have played an important role in the delimitation of the taxa at specific level (Bergreen, 1981) however, surface plates were totally ignored and simple striate, tuberculate or scabrid surfaces were observed (Perveen, 2009). Similarly, all the species of the genus *Silene* can be distinguished quite easily from each other due to seed coloration, shape and surface plates margins. Among the species of the genus *Arenaria*, *A. neelgerensis* is distinguished from other species by having centrally depressed seeds, while in remaining species seeds are not centrally depressed. Similarly, *A. orbiculata* can be distinguished by having brown seeds with tuberculate plates, While in *A. serpyllifolia* and *A. leptoclades* seeds are chocolate brown with non tuberculate surface plates, but these two species can be separated from each other by having different seed size. These present findings are in contrast to that of the earliest findings of Wofford (1981) who reported colliculate surface in *A. orbiculata* and *A. serpyllifolia*. On the basis of seed morphology the genus *Cerastium* can be divided in two main groups i.e., seeds angular and cuneate or seed non-angular and elliptic pyriform. The group with angular and cuneate seeds includes *C. glomeratum*, *C. pusillum*, *C. fontanum* subsp. *trivale* and these taxa could not be further separated by sharing common seed characters. While the remaining species have non-angular and elliptic pyiform seeds, where *C. cerastioides* can be easily distinguished due to yellowish brown seeds by having surface plates with papillate margin, while in the remaining two species, *C. dichotomum* and *C. tomentosum*, seeds are brown and surface plates have smooth margin. These two species can be further distinguished as sub central hilum is found in *C. dichotomum* and *C. tomentosum* is characterized by the presence of sub basal hilum.

Similarly, all the species of the genus *Gypsophila*, can be divided into two group such as seeds elliptic pyriform or reniform-transversely cuneate. *G. alsinioides* and *G. floribunda* can be grouped together by having elliptic seeds, but still these species can be distinguished from each other by having surface plates with dentate margin in *G. alsinioides* and surface plates with serrate margin are found in *G. floribunda*. Among the species having elliptic pyriform-transversely cuneate seeds, *G. makaranica* is characterized by having light brown seeds and surface plates with serrulate margin, while in the remaining two species *G. cerastioides* and *G. bellidifolia* seeds are dark brown and surface plates have smooth or serrate margin, but these two species remains distinct from each other by having granulated plates with serrate margin in *C. cerastioides* and non-granulated plates with smooth margin in *G. bellidifolia*. Moreover, the genus *Holosteum* is characterized by having papillate seed surface as also noticed out by Bergreen (1981) but these findings are not in accordance with Dequan & Rabeler (2001) who observed rugolose seed surface in *Holosteum.* Among the species of *Leprodiclis*, *L. tenera* remains distinct from rest of the species due to the presence of grey seeds, while seeds are dark brown in *L. holosteoides* and *L. stellaroides* but both these species can be distinguished by having elliptic pyriform seeds in *L. holosteoides*, while *L. stellaroides* have reniform seeds.

The species of the genus *Minuartia* can be separated into two groups such as seeds with central hilum or with sub-central hilum, *M. hybrida* and *M. kashmirica* can be coupled by having seeds with central hilum, but they can be separated from each other, as seeds are centrally depressed and surface plates have smooth margins in *M. hybrida* but in *M. kashmirica* seeds are not centrally depressed and surface plates have serrulate margin. Similarly, *M. biflora* and *M. meyeri* have seeds with sub-central hilum, and they can be distinct from each other by granulate and irregularly arranged plates in *M. biflora* and smooth with regularly arranged plates in *M. meyeri*.

The two species of genus *Polycarpea* can be easily distinguished by the presence of elliptic seeds in *P. spicata* and sub-reniform seeds in *P. coryombosa.* Likewise, the two species of *Saponaria* can be easily distinguished from each other as seeds with central-sub central hilum and surface plates with smooth margin has been found in *S. griffithiana*, while in *S. subrosularis* seeds having marginal hilum and surface plates are with serrate margin.

Within the genus *Spergularia*, *S. media* remains distinct from rest of the species by having winged seeds, while the other two species have non-winged seeds, and these species remains distinct from each other by having elliptic pyriform seeds with papillate-granulate surface in *S. marina*, while *S. diandra* is characterized by the presence of cuneate seeds with pappilate-granulate surface. However, Ahmad & Qaiser (1989) reported tuberculate seed surface instead of papillate surface.

Shilong & Rabeler (2001) observed smooth or tuberculate seed surface in the genus *Stellaria*, but in the present study seed surface with plates has been observed. The species of the genus *Stellaria* can be further separated on the basis of seed surface plate margins.

It is concluded that seed morphological data also supports the earlier pollen morphological findings (Perveen, 2003, 2006) as the sub family Paronychioideae can be clearly differentiated from rest of the two sub families viz., Alsinoideae and Caryophylloideae. However, both the subfamilies Alsinoideae and Caryophylloideae cannot be separated on the basis of seed morphology as the seeds in both the subfamilies are quite similar. However, it strongly supports the delimitation of taxa at the generic and specific levels.

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