

THE SEED ATLAS OF PAKISTAN-II GREWIOIDEAE

AFSHEEN AATHER, RUBINA ABID AND M. QASIER*

Department of Botany, University of Karachi, Karachi-75270, Pakistan

*Federal Urdu University of Arts, Science and Technology, Karachi, Pakistan.

Abstract

Seed morphology of 20 taxa belonging to the subfamily Grewioideae of the extended family Malvaceae, was examined using light and scanning electron microscopy (SEM). Seed macro and micro morphological characters were found useful to delimit the taxa both at the generic and specific levels and for designing a Seed atlas of Pakistan.

Introduction

Family Malvaceae is a large family divided into 9 subfamilies viz., Grewioideae, Byttnerioideae, Tilioideae, Dombeyoideae, Malvoideae, Bombacoideae, Helicteroideae, Sterculioideae and Brownlowioideae (Judd & Manchester, 1997; Alverson *et al.*, 1999; Bayer & Kubitzki, 2003). The subfamily Grewioideae mainly includes most of the genera of the former family Tiliaceae (Bayer & Kubitzki, 2003). Some reports are available on the seed morphology of the subfamily Grewioideae. Different seed characters were used by various workers such as Hooker (1876), Ghafoor (1974), Corner (1976) and Ya *et al.*, (2007) who have given the importance to the seed shape, type of cotyledons and position of embryo. Whereas, Ahmed & Qaiser (1989), Kirkbride *et al.*, (2006) and Moeaha *et al.*, (2009) observed that the surface pattern of seeds was also found useful to delimit the various taxa of the subfamily Grewioideae. In the present study, seeds of the subfamily Grewioideae were examined under light and scanning electron microscopy to evaluate the taxonomic decisions and to design a Seed atlas of Pakistan.

Material and Methods

Mature seeds of 20 taxa of the subfamily Grewioideae were collected from herbarium specimens (Appendix I) and examined for seed morphological characters under stereomicroscope (Nikon XN Model), compound microscope (Nikon type 102) and scanning electron microscope (JSM-6380A). For scanning electron microscopy dry seeds were directly mounted on metallic stub using double adhesive tape and coated with gold for a period of 6 minutes in sputtering chamber and observed under SEM. The terminology used is in accordance to Bergreen (1981) and Stearn (1983) with slight modifications. The characters of seed viz., shape, colour, size, surface, position of ridge, margin and hilum were studied.

Observations

General seed characters of the subfamily Grewioideae: Seeds 2.5-2.7 x 1.7-1.9mm, angular or non angular, globose-sub globose, obliquely truncate with rounded or sharp edges, rhomboidal, wedge shaped, cuneiform, elliptic-pyriform, ovate, plano-convex and ovoid with or without membranous margin, ridge dorsal or lateral, oblique or straight or absent. Colour varies from light brown to dark brown, reddish brown, blackish brown, greenish brown, yellowish brown, yellowish golden, yellowish green, black and greenish black. Glabrous, rugose, alveolate, reticulate, sulcate, striate, rugosely striate, sparsely-densely lineate, lineately rugose, ruminate, appressedly ruminate or ruminate foveate-alveolate. Hilum distinct or indistinct, basal or sub basal (Table 1; Figs. 1-3).

Appendix I. List of the voucher specimens.

Taxa	Collector, Number & Herbarium
<i>Corchorus aestuans</i>	S. A Farooqi & M. Qaiser 3201(KUH); M. Qaiser <i>et al.</i> , 3791, 3728 (KUH); Sultan ul Abedin & A. Ghafoor 4051 (KUH); M. Tasnif s.n. (KUH); Jafri 4109 (KUH). Khush Gul s.n. (KUH).
<i>C. capsularis</i>	
<i>C. depressus</i>	S. I. Ali & Sultan ul Abedin 332 (KUH); Sultan ul Abedin 3745 (KUH); Jafri 809 (KUH); S. A. Farooqi & Sultan ul Abedin 486 (KUH); M. Qaiser & A. Ghafoor 3951 (KUH).
<i>C. fascicularis</i>	S. M. H. Jafri 1027, 1060 (KUH).
<i>C. olitorius</i> var. <i>olitorius</i>	M. Qaiser & A. Ghafoor 4664 (KUH); M. Qaiser & Saifullah 2152 (KUH); M. Qasier <i>et al.</i> , 3621 (KUH).
<i>C. olitorius</i> var. <i>incisifolius</i>	Sultan ul Abedin 7738 (KUH).
<i>C. pseudo-olitorius</i>	Coll. Ignat. s.n. (KUH).
<i>C. tridens</i>	S. A. Farooqi & M. Qaiser 2892 (KUH); Jafri 2565, 2566 (KUH); M. Qaiser & A. Ghafoor 3948 (KUH); S. A. Farooqi s.n. (KUH); M. Qaiser <i>et al.</i> , 3665 (KUH); Sultan ul Abedin 3746 (KUH).
<i>C. trilocularis</i>	S. Khatoon 358, 401 (KUH); Tahir Ali 680 (KUH); Sultan ul Abedin 355 (KUH); M. Qaiser & A. Ghafoor 4957, 4959 (KUH); M. Qaiser <i>et al.</i> , 611 (KUH).
<i>Grewia asiatica</i>	Saida Qasier s.n. (KUH); Amtul Waheed s.n. (KUH); Ruqaya Islam s.n. (KUH).
<i>G. damine</i>	M. Qaiser & S. A Farooqi 236 (KUH).
<i>G. elastica</i>	Ulfat Hussain Zargar 786456 (KUH).
<i>G. glabra</i>	M. Qasier 2685 (KUH).
<i>G. helicterifolia</i>	Tahir Ali & S. Nadeem Ahsan 1937 (KUH).
<i>G. optiva</i>	M. Qaiser & A. Ghafoor 4842 (KUH).
<i>G. tenax</i>	M. Qaiser & A. Ghafoor 4463 (KUH); Jafri 1420 (KUH); S.I. Ali <i>et al.</i> , 1853 (KUH); Rizwan Yusuf & A. Hussain 56 (KUH); A. Ghafoor & Tahir Ali 3678 (KUH); Rizwan Yusuf 75 (KUH); A. Hussain 1023 (KUH); Sultan ul Abedin 3670 (KUH).
<i>G. villosa</i>	S. I. Ali <i>et al.</i> , 308 (KUH); M. Qaiser & A. Ghafoor 4459 (KUH); M. Qaiser, A. Ghafoor & A. Hussain 4056 (KUH); S.A. Farooqi & M. Qaiser 2742, 2733 (KUH); A. Hussain s.n. (KUH).
<i>Triumfetta pentandra</i>	M. Qaiser <i>et al.</i> , 4004 (KUH); S. A. Farooqi & M. Qaiser 3184 (KUH).
<i>T. rhomboidea</i>	M. Qaiser 7063 (KUH).
<i>T. rotundifolia</i>	S.I. Ali 1501 (KUH); S.I. Ali <i>et al.</i> , 4473 (KUH); M. Qaiser & A. Ghafoor 3959(KUH).

Table1. Seed morphological characters of the sub family Grewioideae.

Name of taxa	Size (mm)	Shape	Colour	Ridge	Membranous margin	Surface	Hilum	Position of hilum
<i>Cochrorus aestuans</i>	1-1.7 x 0.7-1	Angular, obliquely truncate at both ends, rounded edges	Blackish brown	Dorsal and oblique	Absent	Ruminant foveate to alveolate	Indistinct	Towards the ridge
<i>C. capsularis</i>	2.5-2.6 x 1.4-1.5	Angular, cuniform, rounded edges	Dark reddish brown	Dorsal and oblique	Absent	Rugose	Indistinct	Towards the ridge
<i>C. depressus</i>	1.4-1.5 x 0.5-0.6	Angular, obliquely truncate, sharp edges	Light brown to blackish brown	Dorsal and oblique	Absent	Ruminant	Indistinct	Towards the ridge
<i>C. fascicularis</i>	1-1.3 x 0.9-1	Angular, wedge shape, obliquely truncate, sharp edges	Reddish brown to black	Lateral and straight	Absent	Rugose	Indistinct	Towards the ridge
<i>C. olitorius</i> var. <i>olitorius</i>	1.9-2 x 1-1.3	Angular, rhomboidal, obliquely truncate, sharp edges	Dark brown, greenish black	Dorsal and straight	Absent	Rugose	Indistinct	Towards the ridge
<i>C. olitorius</i> var. <i>insicifolius</i>	1 x 0.5-0.8	Angular, obliquely truncate at both ends, sharp edges	Dark brown	Dorsal and straight	Absent	Reticulate	Indistinct	Towards the ridge
<i>C. pseudo-olitorius</i>	1.4-1.5 x 0.9-1	Angular, cuniform, rounded edges	Black & dark brown at edges	Lateral and straight	Absent	Alveolate	Indistinct	Towards the ridge
<i>C. tridens</i>	1.2-1.3 x 0.6-0.7	Angular, obliquely truncate at both ends, sharp edges	Blackish brown	Dorsal and straight	Absent	Appressedly ruminant	Indistinct	Towards the ridge
<i>C. trilocularis</i>	1.2-1.5 x 0.8-0.9	Angular, obliquely truncate at both ends, rounded edges	Black	Dorsal and oblique	Absent	Ruminant	Indistinct	Towards the ridge
<i>Grewia asiatica</i>	3.5-3.8 x 2.5-2.8	Elliptic-pyriform - sub globose	Light brown	Absent	Entire	Rugosely striate	Distinct	Basal
<i>G. damine</i>	2.8-3 x 2-2.5	Elliptic-pyriform	Yellowish golden	Absent	Entire	Striate	Distinct	Sub basal
<i>G. elastica</i>	2.4-2.5 x 2.2-2.3	Elliptic-pyriform - sub globose	Yellowish green	Absent	Entire	Densely linearly striate	Distinct	Sub basal - basal
<i>G. glabra</i>	3.5-3.6 x 3-3.1	Ovate	Yellowish brown	Absent	Entire	sparsely linearly striate	Distinct	Sub basal
<i>G. helicterifolia</i>	3.3-3.4 x 2.5-2.8	Globose	Light brown-yellowish golden	Absent	Entire	Sulcate	Distinct	Basal
<i>G. optiva</i>	3-3.2 x 2-2.3	Ovate	Greenish brown	Absent	Entire	Rugose	Distinct	Sub basal
<i>G. tenax</i>	4-4.3 x 3-3.3	Ovate	Yellowish brown	Absent	Wavy	Densely linearly striate	Distinct	Sub basal
<i>G. villosa</i>	3.3-3.5 x 2.2-2.5	Elliptic-pyriform	Yellowish golden	Absent	Wavy	Lineately rugose	Distinct	Sub basal
<i>Triumfetta pentandra</i>	2.9-3 x 2-2.1	Angular, ovoid	Reddish brown	Absent	Absent	Rugose	Distinct	Basal
<i>T. rhomboidea</i>	3-3.1 x 1.9-2	Angular, ovoid	Dark brown	Absent	Absent	Alveolate	Distinct	Sub basal
<i>T. rotundifolia</i>	2.9-3 x 1.8-2	Angular, ovoid, plano-convex	Dark brown	Absent	Absent	Rugose	Distinct	Basal

Key to the genera

- 1 + Seeds globose, sub globose, elliptic-pyriform, ovoid or ovate. Hilum distinct, basal-sub basal 2

- Seeds obliquely truncate. Hilum indistinct and towards the ridge *Corchorus*

2 + Seeds non angular with membranous margin *Grewia*

- Seeds angular with out membranous margin *Triumfetta*

***Corchorus* L.**

Seeds angular, rhomboidal, wedge shaped, cuneiform and obliquely truncate with sharp to rounded edges. Colour light brown, dark brown, reddish brown, blackish brown, black and greenish black, 1.43-1.6 x 0.85-0.93mm, glabrous, rugose, alveolate, reticulate, ruminate, appressedly ruminate and ruminate foveate-alveolate. Hilum indistinct, present towards the ridge. Ridge dorsal or lateral, oblique to straight (Table 1; Figs. 1 A-O, 2 A-C).

Represented by 9 taxa viz., *C. aestuans* L., *C. capsularis* L., *C. depressus* (L.) Stocks, *C. fascicularis* Lam., *C. olitorius* L. var. *olitorius*, *C. olitorius* L. var. *incisifolius* Aschers. & Schwein, *C. pseudo-olitorius* Islam & Zaid, *C. tridens* L. and *C. trilocularis* L.

Key to the species

- 1 + Seeds obliquely truncate with sharp edges 2

 - Seeds obliquely truncate with rounded edges or cuneiform 5

2 + Seeds with straight ridge 3

 - Seeds with oblique ridge *C. depressus*

3 + Seeds reddish brown-black or dark brown-greenish black, rugose 4

 - Seeds blackish brown, appressedly ruminate *C. tridens*

4 + Seeds wedge shaped, reddish brown-black, ridge towards the lateral side *C. fascicularis*

 - Seeds rhomboidal, dark brown, greenish black, ridge towards the dorsal side *C. olitorius*

5 + Ridge oblique on dorsal side 6

 - Ridge straight on lateral side *C. pseudo-olitorius*

6 + Seeds blackish brown, black, 1-1.7 x 0.7-1 mm 7

 - Seeds reddish brown, 2.5-2.6 x 1.4-1.5 mm *C. capsularis*

7 + Seeds blackish brown, ruminate foveate-alveolate *C. aestuans*

 - Seeds black, ruminate *C. trilocularis*

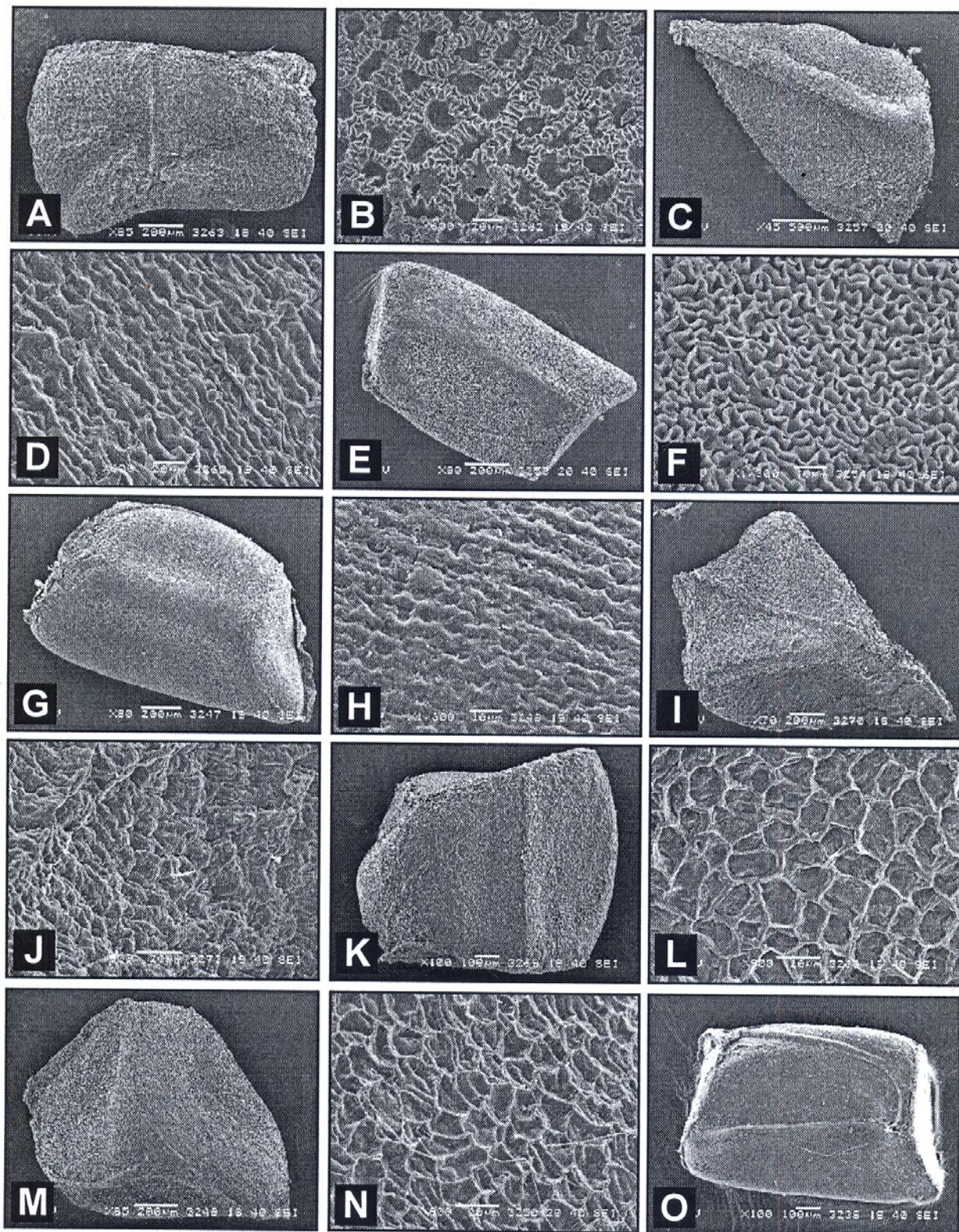


Fig. 1. Scanning Electron micrographs. *C. aestuans*: A, seed; B, surface. *C. capsularis*: C, seed; D, surface. *C. depressus*: E, seed; F, surface. *C. fascicularis*: G, seed; H, surface. *C. olitorius* var. *olitorius*: I, seed; J, surface. *C. olitorius* var. *insicifolius*: K, seed; L, surface. *C. pseudo-olitorius*: M, seed; N, surface. *C. tridens*: O, seed (Scale bar: A, E, G, I, M= 200μm; B, D, J, L, N= 20μm; C= 500μm; F, H= 10μm; K, O= 100μm).

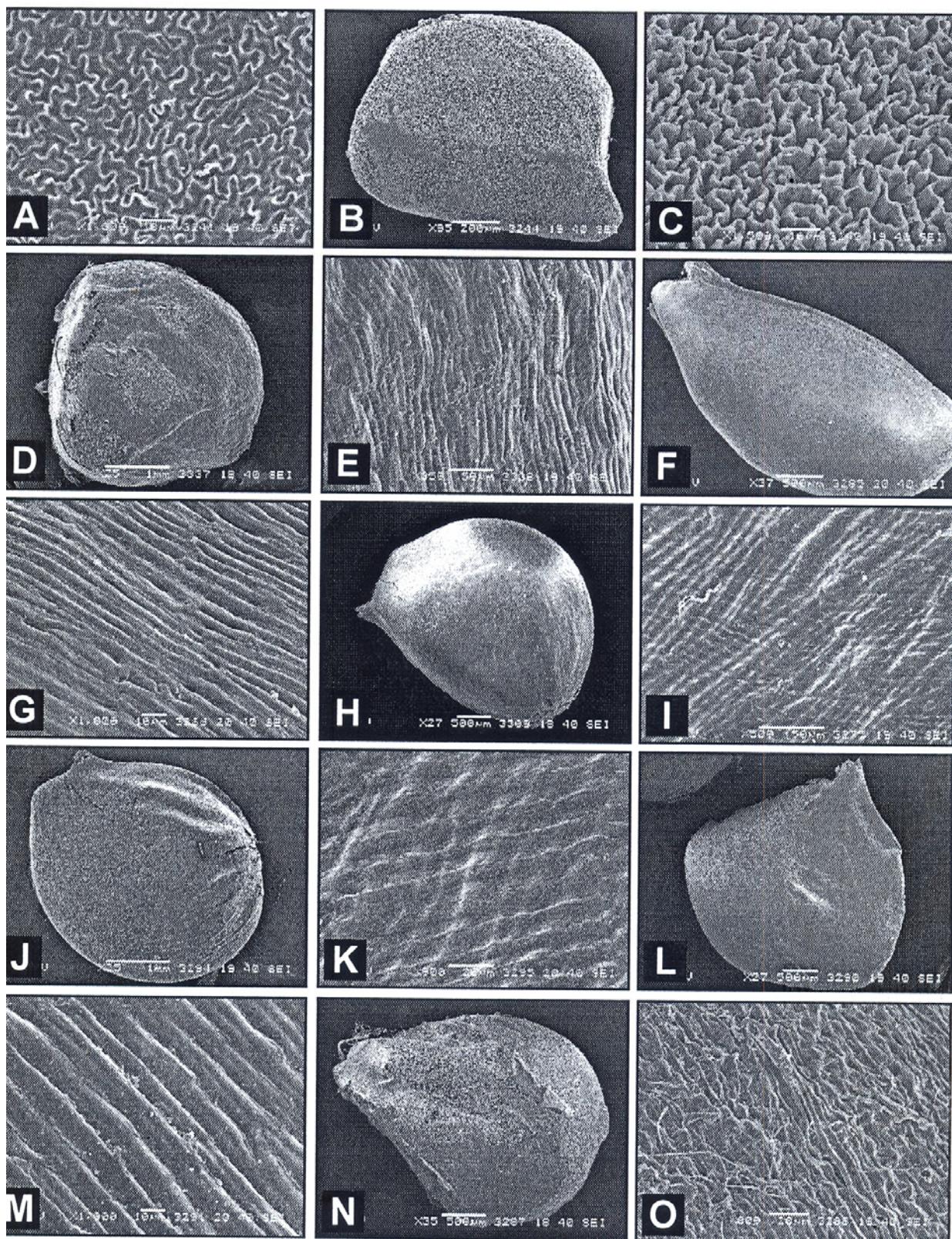


Fig. 2. Scanning Electron micrographs. *Corchorus tridens*: A, surface. *C. trilocularis*: B, seed; C, surface. *Grewia asiatica*: D, seed; E, surface. *G. damine*: F, seed; G, surface. *G. elastica*: H, seed; I, surface. *G. glabra*: J, seed; K, surface. *G. helicterifolia*: L, seed; M, surface. *G. optiva*: N, seed; O, surface (Scale bar: A, C, D, G, J, M= 10 μ m; B= 200 μ m; E, I= 50 μ m; F, H, L, N= 500 μ m; K, O= 20 μ m).

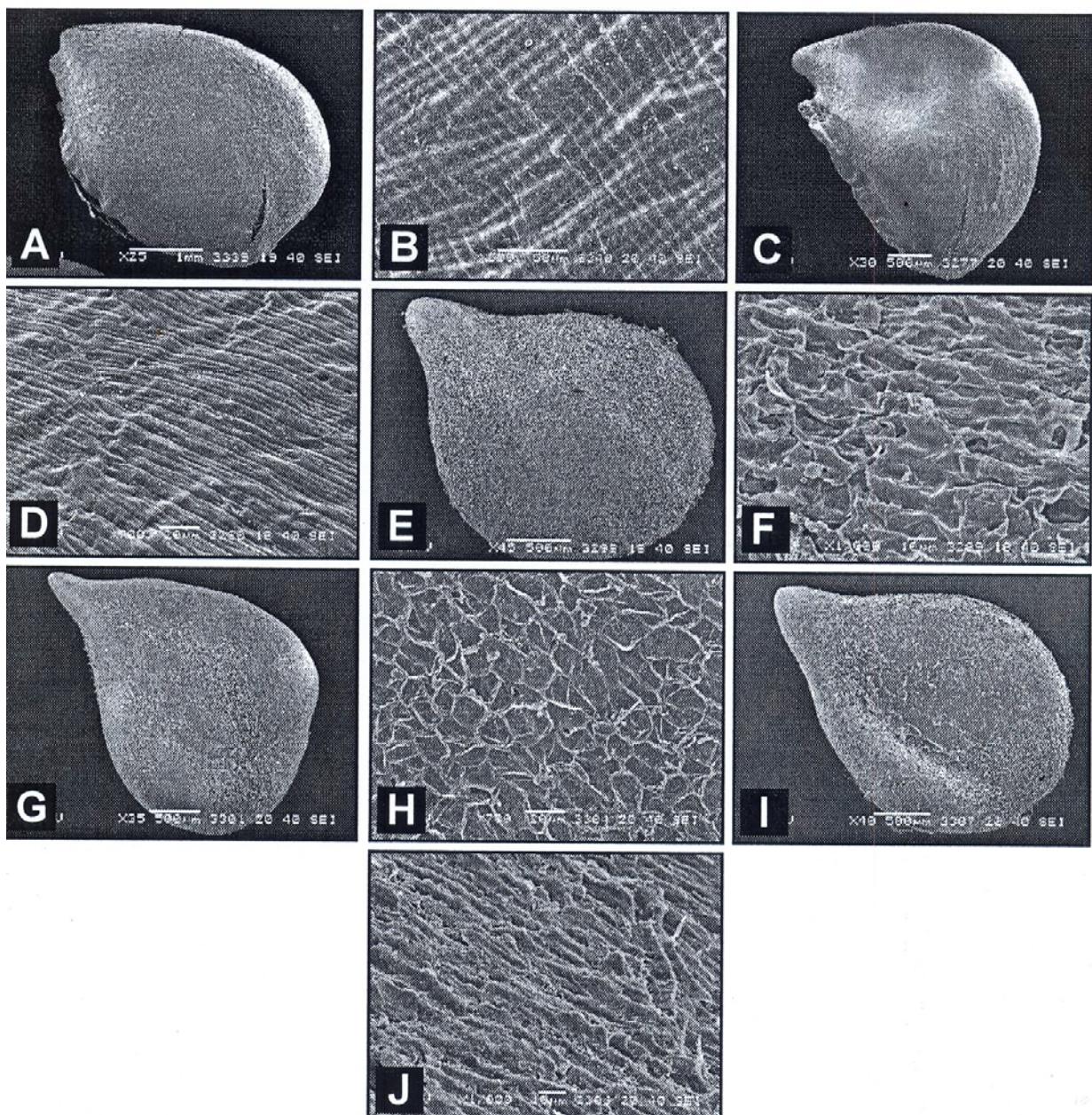


Fig. 3. Scanning Electron micrographs. *Grewia tenax*: A, seed; B, surface. *G. villosa*: C, seed; D, surface. *Triumfetta pentandra*: E, seed; F, surface. *T. rhomboidea*: G, seed; H, surface. *T. rotundifolia*: I, seed; J, surface (Scale bar: A, F, J= 10μm; B= 50μm; C, E, G, I= 500μm; D, H= 20μm).

Grewia L.

Seeds non angular, globose to sub globose, elliptic-pyriform or ovate with membranous wavy or entire margin, light brown, yellowish golden, yellowish brown, yellowish green and greenish brown, 3.2-3.4 x 2.4-2.7mm, glabrous, rugose, lineately rugose, sulcate, striate, rugosely striate, sparsely-densely lineate. Hilum distinct, basal-sub basal (Table 1; Figs. 2 D-O, 3 A-D). Represented by 8 species viz., *G. asiatica* L., *G. damine* Gaertn., *G. elastica* Royle, *G. glabra* Bl., *G. helicterifolia* Wall. ex G.Don, *G. optiva* Drum. ex Burret., *G. tenax* (Forsk.) Fiori and *G. villosa* Willd.

Key to the species

- 1 + Seeds with entire membranous margin 2
 - Seeds with wavy membranous margin 7

- 2 + Seeds elliptic-pyriform to sub globose 3
 - Seeds ovate or globose 5

- 3 + Seeds yellowish golden, yellowish green, hilum basal-sub basal 4
 - Seeds light brown, hilum basal *G. asiatica*

- 4 + Seeds striate *G. damine*
 - Seeds lineately striate *G. elastica*

- 5 + Seeds ovate, greenish brown or yellowish brown 6
 - Seeds globose, light brown-yellowish golden *G. helictrifolia*

- 6 + Seeds rugose *G. optiva*
 - Seeds lineately striate *G. glabra*

- 7 + Seeds ovate, yellowish brown, 4-4.3 x 3-3.3mm, lineately striate *G. tenax*
 - Seeds elliptic-pyriform, yellowish golden 3.3 -3.5 x 2.2-2.5mm, lineately rugose *G. villosa*

***Triumfetta* L.**

Seeds angular, ovoid and plano-convex, dark brown-reddish brown, glabrous 2.9-3.3 x 1.9-2.3 mm, rugose or alveolate. Hilum distinct, basal-sub basal (Table 1; Fig. 3 E-J).

Represented by 3 species viz., *T. pentandra* A. Rich., *T. rhomboidea* Jacq., and *T. rotundifolia* Lam.

Key to the species

- 1 + Seeds rugose, hilum basal 2
 - Seeds alveolate, hilum sub basal *T. rhomboidea*

- 2 + Seeds ovoid, planoconvex, dark brown *T. rotundifolia*
 - Seeds ovoid, reddish brown *T. pentandra*

Result and Discussion

The subfamily Grewioideae has quite diverse and stable seed morphological characters which could be easily used to delimit the taxa both at generic and specific levels. The genus *Corchorus* is characterized by having angular and obliquely truncate seeds (Ghafoor, 1974; Ahmed & Qaiser, 1989; Ya *et al.*, 2007). The genus *Grewia* has non-angular seeds with membranous margin, while the genus *Triumfetta* could be distinguished from rest of the genera by having angular, ovoid and plano-convex seeds (Ghafoor, 1974). However, the importance of seed morphology at specific level is also

evident in *Corchorus*, *Grewia* and *Triumfetta* where the species of the genus *Corchorus* are divided into two groups such as seeds with sharp edges includes *C. depressus*, *C. tridens*, *C. fascicularis* and *C. olitorius*. Second group having seeds with rounded edges including *C. aestuans*, *C. capsularis*, *C. pseudo-olitorius* and *C. trilocularis*. Amongst the taxa of first group *C. depressus* is distinguished from rest of the species by having the seeds with ruminate surface, indistinct hilum on dorsal and oblique ridge. While in remaining species rugose, reticulate or appressedly ruminate surface along with the indistinct hilum on dorsal or lateral and straight ridge was observed. However, present findings of *C. depressus* are not in accordance with the earlier findings of Ahmed & Qaiser (1989) where oblong and reticulately striate seeds with circular and lateral hilum were observed. Furthermore, *C. tridens* is characterized by having appressedly ruminate surface and rest of the two species have rugose surface but still both the species remain distinct by having wedge shaped and rhomboidal seed in *C. fascicularis* and *C. olitorius* respectively. Within the second group, *C. pseudo-olitorius* is distinguished by the presence of straight ridge on lateral side and rest of the species have oblique ridge on dorsal side. Amongst them *C. capsularis* is separated on the basis of reddish brown seeds and remaining ones have blackish brown-black seeds and both could be further distinguished by having ruminate foveate-alveolate surface in *C. aestuans* and ruminate surface in *C. trilocularis*. Similarly the two varieties of *C. olitorius* could also be separated from each other by having seeds with rugose surface in *C. olitorius* var. *olitorius* while, *C. olitorius* var. *insicifolius* has reticulate surface.

All the species of *Grewia* are also distinguished from each other as *G. tenax* and *G. villosa* are separated by having seeds with wavy margin while remaining species having seeds with entire membranous margin. Both the species remain distinct by having ovate seeds with striate and densely lineate surface in *G. tenax* while, *G. villosa* is characterized by having elliptic-pyriform seeds with lineately rugose surface. However, the present findings are in contrast to that of the findings of Ahmed & Qaiser (1989) where oblong-ovate seeds with psilate surface has been observed in *C. villosa*. Species having seeds with entire margin remain distinct by having different surface patterns and the position of hilum. Amongst all the species of the genus *Triumfetta*, *T. rhomboidea* is the only species having alveolate seeds with sub basal hilum. While, seeds with rugose surface and basal hilum were found in *T. rotundifolia* and *T. pentandra* and both the species could be differentiated by having different seed colours.

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