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Fruit and seed morphology in *Galium* L. (Rubiaceae) and its importance for taxonomic identification

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Fruit (mericarp) and seed morphology of 11 species and two subspecies of *Galium* were examined with light microscope and scanning electron microscope. Macro-and micro--morphological characters, including fruit and seed shape, colour, size, surface, epidermal cell shape, anticlinal boundaries, outer periclinal cell wall and relief of outer cell walls, are presented. Four different types of mericarp surface are described. Three types of anticlinal cell wall boundaries of seed are recognized and three different shapes of outer periclinal cell wall are described. The secondary sculpture of the cell wall varies from micro-papillate to micro-reticulate, and smooth to fine- or coarse-folded. The fruit and seed characteristics could be used for taxonomic identification.

Key words: Fruit, seed, morphology, Rubiaceae, *Galium*

Introduction

Rubiaceae form the fourth largest angiosperm family after Asteraceae, Orchidaceae and Fabaceae and comprise more than 10 000 species belonging to 640 genera. Rubiaceae are usually considered a natural monophyletic and easily circumscribed family (VERDCOURT 1958, BREMEKAMP 1966, ROBBRECHT 1988). In the flora of Egypt Rubiaceae are represented by eight genera, viz. *Kohautia*, *Oldenlandia*, *Galium*, *Valantia*, *Callipeltis*, *Crucianella*, *Pterogaillonia* and *Rubia* (BOULOS 1995, 2000). *Cruciata articulata* is reported as a new record to the flora of Egypt, so the number of genera in Rubiaceae was increased to nine (ABDEL KHALIK and BAKKER 2007).

Galium is one of the largest genera of Rubiaceae (WILLIS 1985, MABBERLEY 1987). In Egypt *Galium* is represented by 12 species (TACKHOLM 1974). BOULOS (2000) reported only 10 species of *Galium*. In the Flora orientalis, BOISSIER (1881) reported 91 species of the genus *Galium* divided into three sections: section (I) Eugalium, section (II) Aparine, and section (III) Cruciata.

Section (I) Eugalium is classified into four subsections: 1. Platygalia, 2. Leiogalia, 3. Chromogalia, and 4. Ceratocapa.

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Section (II) Aparine is classified into five subsections: 1. Camptopoda, 2. Leucaparinea, 3. Xanthaparinea, 4. Bracteata, and 5. Apera. Subsection Leiogalia includes *G. mollugo,* subsection Chromogalia includes the following species: *G. sinaicum* and *G. canum, s*ubsection Ceratocapa includes *G. ceratocarpum, s*ubsection Camptopoda includes *G. tricorne* and *G. ceratopodum,* subsection Leucaparinea includes *G. aparine* and *G. spurium, s*ubsection Xanthaparinea includes the following species: *G. parisiense G. nigricans, G. decaisnei* and *G. setaceum,*and subsection Apera includes *G. murale.*

Most taxonomists agree that the macro- and microstructure of seeds are significant for the classification of Angiosperm taxa.

During the last decades, scholars have applied SEM to morphological studies of seeds and small fruits. Micro-morphology and ultra-structural data have contributed useful information concerning the evolution and for the classification of seed plants and play an important role in the modern synthetic systems of Angiosperms (HEYWOOD 1971, DAHLGREN 1979–80).

Several studies focused on intrageneric seed coat variation (CHUANG and HECKARD 1972, HEYN and HERRNSTEDT 1977, CLARK and JERNSTEDT 1978, WOFFORD 1981, JUAN et al. 2000, ABDEL KHALIK 2006) or on variation among several closely related genera (MUSSELMAN and MANN 1976; SEAVEY et al. 1977; CHANCE and BACON 1984; MATTHEWS and LEVINS 1986; FAYED and EL NAGGAR 1988, 1996; SHANMUKHA and LEELA 1993; KANAK SAHAI et al. 1997; KARAM 1997; KOUL et al. 2000). Less commonly, SEM level variation was used to place taxa into tribes (WHIFFIN and TOMB 1972, ABDEL KHALIK and VAN DER MAESEN 2002).

More information is available concerning seed forms and internal structures (CORNER 1976, KABIL et al. 1980), seed size (ANISZEWWSKI et al. 2001), seed colours (BERGGREN 1962, DAHLGREN and CLIFFORD 1982, BARTHLOTT 1984), and epidermal cell patterns and distribution of trichomes, glands, and stomata (STACE 1965). The distribution of epidermal cells is important between species and genus level (BARTHLOTT 1981, 1984), while outer periclinal walls are also a good diagnostic for the lowest taxonomic categories (BARTHLOTT 1981, 1984, ABDEL KHALIK et al. 2002, ABDEL KHALIK 2006).

The present investigations deal with the micro- and macro-morphological (LM and SEM) characters of fruits (mericarps) and seeds for 11 species and two subspecies of the genus *Galium* from the Rubiaceae in Egypt, to show the ranges of variability in fruit (mericarp) and seed characters in order to establish their usefulness for future taxonomic work.

Materials and methods

Some of the investigated seeds were collected from mature plants in Egypt during 2004–2006. The others were taken from herbarium specimens. Only mature fruits and seeds were taken for investigation. The dried fruits were soaked in boiling water for 2–4 minutes, to compensate for shrinkage and examined by light microscope (Olympus type BH-2), with 10–15 seeds for each taxon being chosen to cover the range of variation. Measurements were standardized with ocular micrometer and fruits drawn with the aid of a camera lucida. Mature fruits and seeds were mounted on stubs with double adhesive tape. The stubs were sputter-coated with gold/palladium for 4 min. in an Apolaron E 1100 ion sputtering device. After coating, the specimens were examined with a JEOL -6300 Scanning Electron

Microscope, using accelerating voltages at 20–30 KV. All photomicrographs were taken at the SEM laboratory, Sohag University, Egypt. The terminology used here follows authors such as (ABDEL KHALIK et al 2002, ABDEL KHALIK 2006) with some modification by the author.

List of *Galium* fruit and seed specimens used in light and scanning electron microscope studies.

1. *Galium aparine* L.: Egypt, Gebel Elba, Gebel Ekwal, J. R. Shabetai 250 (K)
2. *Galium canum* Req.: Palestine, Wadi Sawaanit, in rocks, P.H. Davis 5038 (K)
3. *Galium ceratopodum* Boiss.: Egypt, Sinai, W. Gebal region, Ain Altofaha, K. Abdel Khalik 1956 (SHG).
4. *Galium mollugo* L.: Netherlands, Gelderland, NW of Wolfheze.Open low vegetation on sandy soil, C.C.H. Jongkind 5226 (WAG)
5. *Galium murale* L.: Egypt, in cultivated land, near Maruit, Letuneux 197 (K)
6. *Galium nigricanse* Boiss.: Iran, Kordestan, Sanandaj, rolling limestone hills alt. 1800, M. Jacobs 6938 (BR)
7. *Galium parisiense* L.: France, 5 km E of Cavarllon, W. J. Reijnders 1144 (L)
8. *Galium setacium* Lam. subsp. *setaceum:* Egypt, Sinai, Gebel Serbal, alt. 1684, Fayed et al. 1946–2033 (SHG).
9. *Galium setaceum* subsp. *decaisnei.*Boiss.: Egypt, Gebel Elba, Wadi Drawina, K. Abdel Khalik s.n. (SHG).
10. *Galium sinaicum* (Delile ex Decne.) Boiss.: Egypt, Sinai, Gebel Serbal, alt. 1450, Fayed et al. 2034–2035 (SHG).
11. *Galium spurium* L. subsp. *spurium*: Egypt, Sinai W. Gebel region, Ain Altofaha, K. Abdel Khalik s.n. (SHG).
12. *Galium spurium* subsp. *africanum* Verdc.: Egypt, Gebel Elba, Wadi Drawina, K. Abdel Khalik 3935 K. (SHG).
13. *Galium tricornutum* Dandy: Egypt, Alexandria, De Bullemont s.n. (BR).

Results

Fruit (mericarp) characters

Fruit characters are very important for interspecies distinction in the genus *Galium*. Fruits are of a schizocarpic nature, composed of two mericarps, each mericarp having only one seed.

Mericarp shape

Mericarp shape among the investigated taxa showed a large variation. Mericarps vary from cylindrical or globose, globose to sub globose or reniform (Tab. 1). Mericarp shape showed significant differences among species, but no significant differences among subspecies or varieties. However they are cylindrical in *Galium murale* (Figs. 2.5, 4.5); globose in *G. aparine* and *G. tricornutum*, (Figs.1.1, 4.1, 3.13, 5.14); sub globose in *G. ceratopodum*, *G. spurium* subsp. *spurium* and subsp. *africanum* (Figs. 1.3, 4.3, 3.11, 3.12, 5.12, 5.13); reniform in *G. canum*, *G. mollugo, G. nigricans*, *G. parisiense*, *G. setacium* subsp. *setacium* and subsp. *decaisnei*, *G. sinaicum* (Figs. 1.2, 1.4; 2.6, 2.7, 2.8, 3.10).

Tab. 1. Mericarp and seed description of ***Galium*** species.

Taxon	seed shape	Seed size(mm)	Seed color	Epidermal cell shape	Anticlinal boundries	Periclinal cell wall	Mericarp size (mm)	Meri-carp color.	Mericarp surface
G. aparine L.	Globose	2.8–4.5 × 2.8–4.5	Dark brown	5,6 gonals, elongate in one direction	Raised-channeled, straight, smooth	Flat to slightly con-cave, micro-papillate	3 –5 × 3–5	Dark	Hooked hairs arising from tubercle-like base
G. canum Req.	Reniform	0.5–0.8 × 0.3–0.5	Dark brown	Polygonal, elongate in one direction	Slightly raised, straight to slightly sinous, smooth	Flat, smooth	0.7–1 × 0.5–0.7	Dark brown	Long white simple straight hairs
G. ceratopodum Boiss.	Globose to sub globose	1.6–2.2 × 1.3–2.0	Dark brown	Polygonal	Raised, straight to sinous, smooth.	Flat to slig-htly concave, fine folds	1.7 –2.5 × 1.5 –2.5	Dark brown	Tuberculate
G. mollugo L.	Reniform	1.0–1.6 × 0.8–1.0	Yellow brown	Isodiametric, 5,6 gonals, elongate in one direction	Raised, straight to slightly sinous, smooth to fine folded	Flat to slightly concave, micro-papillate	1.4–1.8 × 1.0–1.2	Dark pinkish	Micropapillate
G. murale L.	Slender	1–1.5 × 0.2–0.4	Black	Isodiametric, 4,5 gonals elongate in one direction	Raised, straight, with coarse folds.	Flat to slightly concave, smooth.	1.3–2.0 × 0.3–0.5	Dark brown	Hooked hairs
G. nigricans Boiss	Rreniform	07–1.3 × 0.3–0.5	Black	4,5 gonals, elongate in one direction.	Raised, straight to slightly sinuous, fine folds.	Flat to slightly concave, smooth	0.9–1.5 × 0.4–0.6	Dark brown	Hooked hairs
G. parisiense L	Reniform	0.7–1.2 × 0.3–0.9	Dark brown	Polygonal	Raised, sinuous, course folds	Flat, course folds	0.8 –1.2 × 0.5 –1.0	Dark brown	Micropapillate

Tab. 1. – continued

Taxon	seed shape	Seed size(mm)	Seed color	Epidermal cell shape	Anticlinal boundries	Periclinal cell wall	Mericarp size (mm)	Meri-carp color	Mericarp surface
G. setacium. Lam subsp. *setacium*	Reniform	0.4–0.9 × 0.2–0.4	Yellow brown	4,5 gonals	Raised, slightly sinuous, smooth to folded	Flat to slightly concave, fine folds	0.5–1.0 × 0.3–0.5	Brown	Hooked hairs
*G. setaceum.*L am subsp. *decaisnei* Boiss	reniform	0.4–0.7 × 0.4–0.6	Yellow brown	Isodimetric, polygonal,	Channeled, sinuous, cours folds	Convex, radiate course folds	0.5–0.8 × 0.4–0.7	Purple to brown	Depressed hairs or short papillae
G. sinaicum (Delile ex Decne.)Boiss.	Reniform	0.8–1.1 × 0.4–0.8	Yellow brown	Isodiametric, 5,6 gonals, elongate in one direction	Slightly raised, straight, smooth	Flat to slightly concave, micro--papillate.	1.0–1.5 × 0.4–0.9	Dirty yellow to brown	Micropapillate.
G. spurium L. subsp. spurium	Globose to sub globose	1.5–2.3 × 1.5–2.2	Dark brown	5,6 gonals	Raised, straight to slightly sinous, fine folded	Flat to slightly concave, mico--papillate.	1.9–2.5 × 1.5–2.5	Dark brown	Hooked hairs not tuberculated at the base
G. spurium.L subsp. africa-num Verdc.	Globose to sub globose	1.8–2.0 × 1.5–2.0	Dark brown	5,6 gonals.	Raised, straight to slightly sinuous, smooth.	Flat to slightly concave, course folds.	1.8– 2.5 × 1.5–2.5	Dark	Long hooked hairs
G. tricornutum Dandy.	Globose	2.0–3.7 × 2.0–3.7	Dark brown	Isodiametric, 4,5 gonals	Raised, straight to slightly sinuous, coarse folds.	Flat to slightly concave, micro reticulate.	2–4 × 2–4	Dark brown	Densely tuberculated

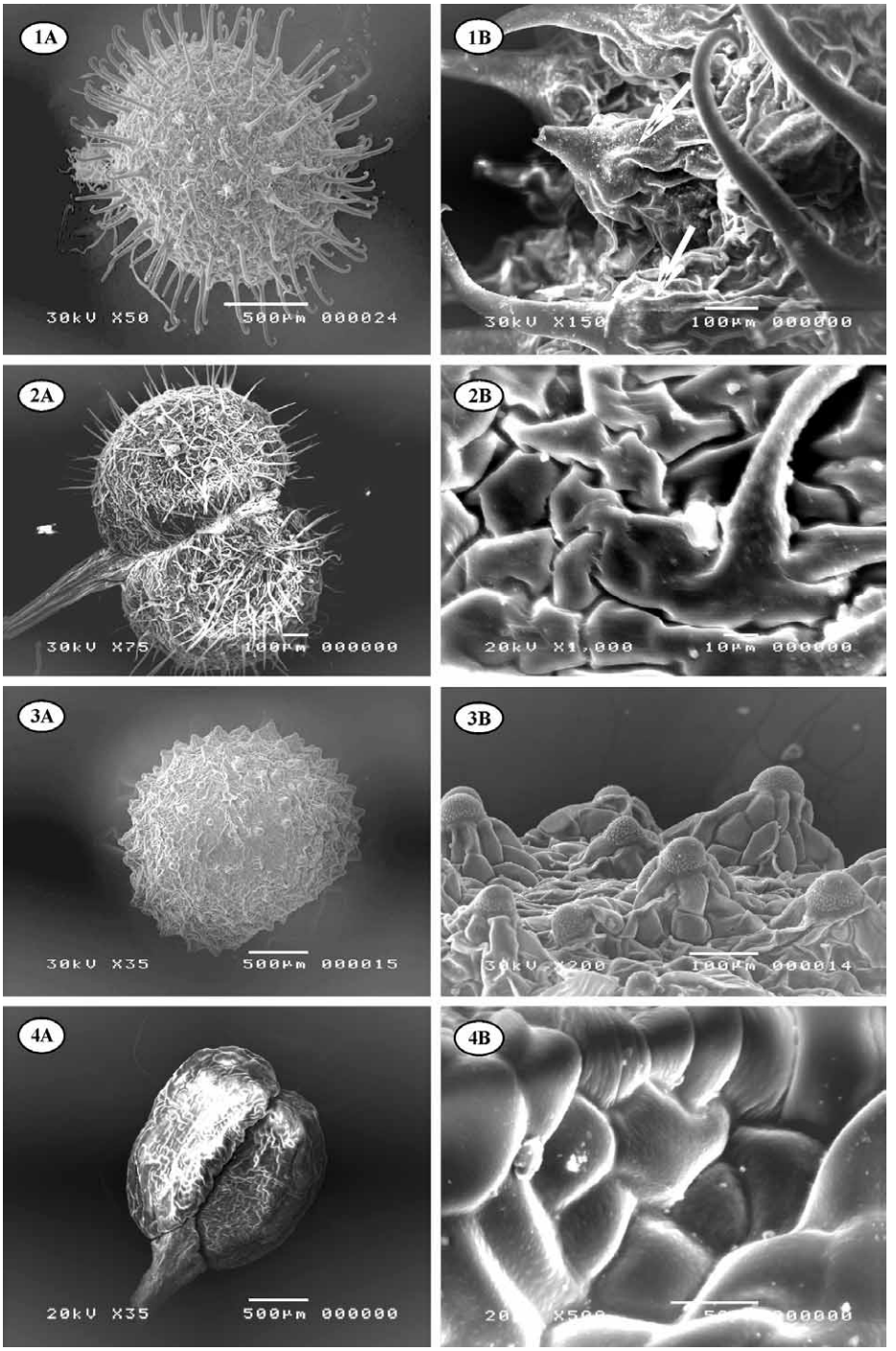

Fig. 1. SEM photographs of mericarps. A – entire mericarps, B – enlargement of mericarp coat, 1 – *Galium aparine*. 2 – *G. canum,* 3 – *G. ceratopodum*. 4 – *G. mollugo*.

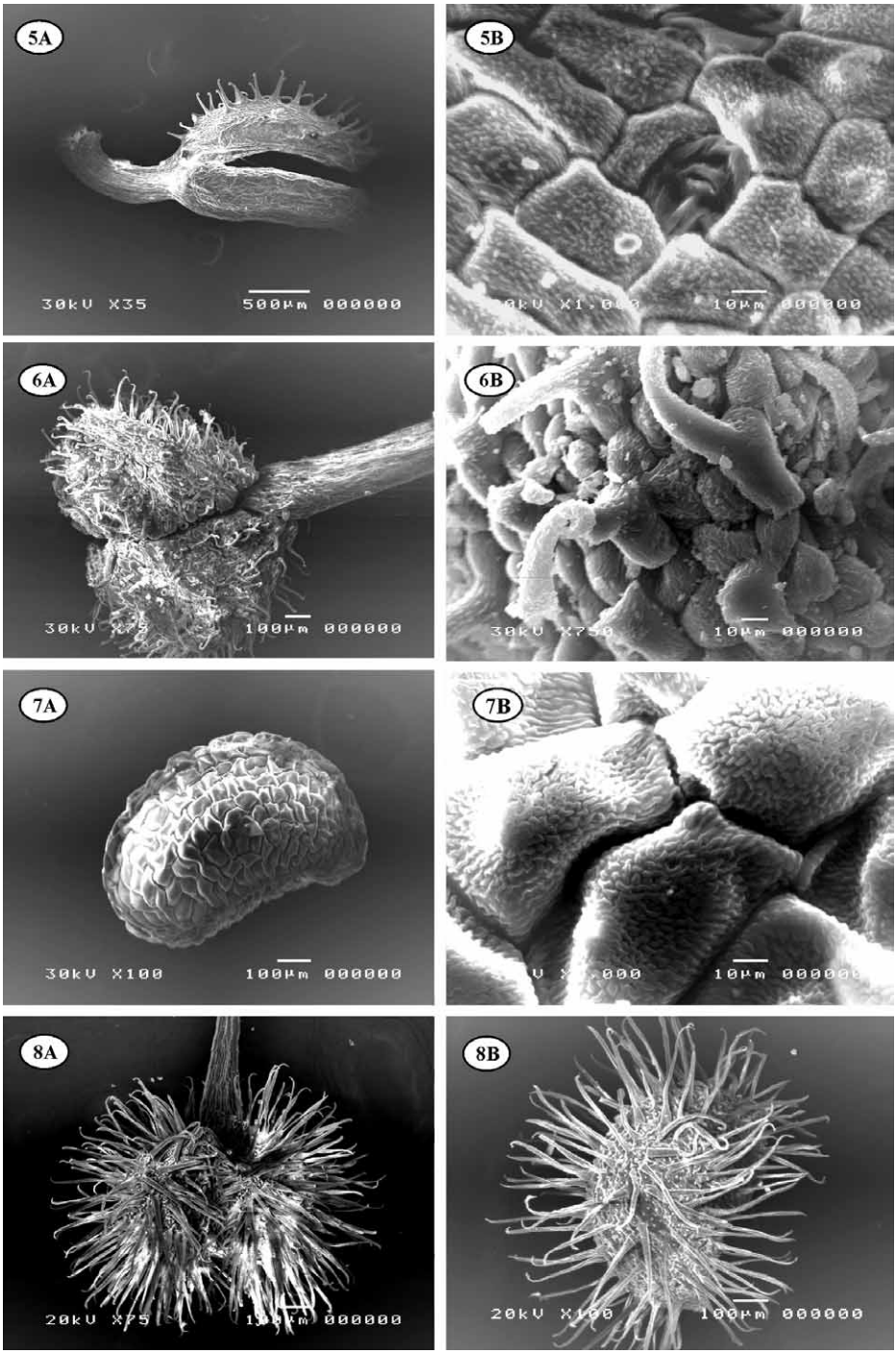

Fig. 2. SEM photographs of mericarps. A – entire mericarps, B – enlargement of mericarp coat. 5 – *Galium murale*, 6 – *G. nigricans*, 7 – *G. parisiense*, 8 – *G. setaceum* subsp. *setaceum*.

Mericarp size

Mericarp dimensions vary greatly among the examined taxa. The largest mericarps in *Galium aparine* and *G. tricornutum* have a diameter of 2–5 × 2–5 mm, and the smallest mericarp measures 0.5–1.5 × 0.3–1 mm in *G. canum*, *G. nigricans*, *G. parisiense*, *G. setacium* and *G. sinaicum*, while the rest of the species have a slightly larger mericarp, 1.3–2.5 x 0.3–2.5 mm. Mericarp size was found useful to distinguish the four species *G. aparine* and *G. spurium,* and *G. ceratopodum* and *G. tricornutum*. However in *G. aparine* mericarp diameter is (3–5 × 3–5 mm), but in *G. spurium* 1.9–2.5 × 1.5–2.5 mm; in *G. tricornutum* too it is 2–4 × 2–4 mm but in *G. ceratopodum* 1.7–2.5 × 1.5–2.5 mm.

Mericarp indumentum

Mericarp indumentum forms one of the most important characters in the classification of *Galium* at different level (specific and infraspecific level). It varies from hooked hairs arising from the tuberculate base in *Galium aparine* (Figs. 1.1, 4.1); hooked hairs without tuberculate base in *G. murale*, *G. nigricans, G. setacium* subsp. *setacium* and *G. spurium* (Figs. 2.5, 4.5, 2.6, 2.8, 5.8, 3.12, 5.12, 13); long simple straight hairs in *G. canum* (Figs. 1.2, 4.2); depressed hairs or short papillae in *G. setacium* subsp. *decaisnei* (Figs. 3.9, 5.9); it is micro-papillate in *G. mollugo*, *G. parisiense* and *G. sinaicum* (Figs. 1.4, 4.4, 2.7, 5.7, 3.10, 5.10); densely tuberculate in *G. ceratopodum* and *G. tricornutum* (Figs. 1.3, 4.3, 3.13, 5.14) to glabrous in *G. spurium* subsp. *spurium* var. *spurium* (Figs. 3.12, 5.12).

Mericarp colour

The colour of the mericarp is of high diagnostic and systematic value at different levels. The colour of mericarps varies from dark, dark brown, dark- pinkish, brown, purple to brown to dirty yellow to brown. However, they are dark in *G. aparine* and *G. spurium* subsp**.** *africanum*, dark brown in *G. canum*, *G. ceratopodum*, *G. murale*, *G. nigricans*, *G. parisiense*, *G. spurium* subsp. *spurium* and *G. tricornutum*, dark-pinkish in *G. mollugo*, brown in *G. setacium* subsp. *setacium*, purple to brown in *G. setacium* subsp. *decaisnei*, dirty yellow to brown in *G. sinaicum*.

The colour is also used to distinguish between subspecies of *G. spurium*, for it is dark brown in subsp. *spurium* and dark in *G. spurium* subsp. *africanum*. It also assists in the distinction of the subspecies of *G. setacium*, being brown in *G. setacium* subsp. *setacium* and purple to brown in *G. setacium* subsp. *decaisnei*.

Seed characters

Seed characters are very important for distinction of species in the genus *Galium*.

Seed shape

The shape of seeds among the investigated taxa showed a large variation. Most seeds vary from cylindrical or globose, globose to sub-globose or reniform (Tab. 1); however they are cylindrical in *Galium murale* (Fig. 7.5); globose in *G. aparine* and *G. tricornutum* (Figs. 6.1, 8.13); sub globose in, *G. ceratopodum*, *G. spurium* subsp *spurium* and subsp *africanum* (Figs. 6.3, 8.11, 8.12); reniform in *G. canum*, *G. mollugo, G. nigricans*e, *G. pari-*

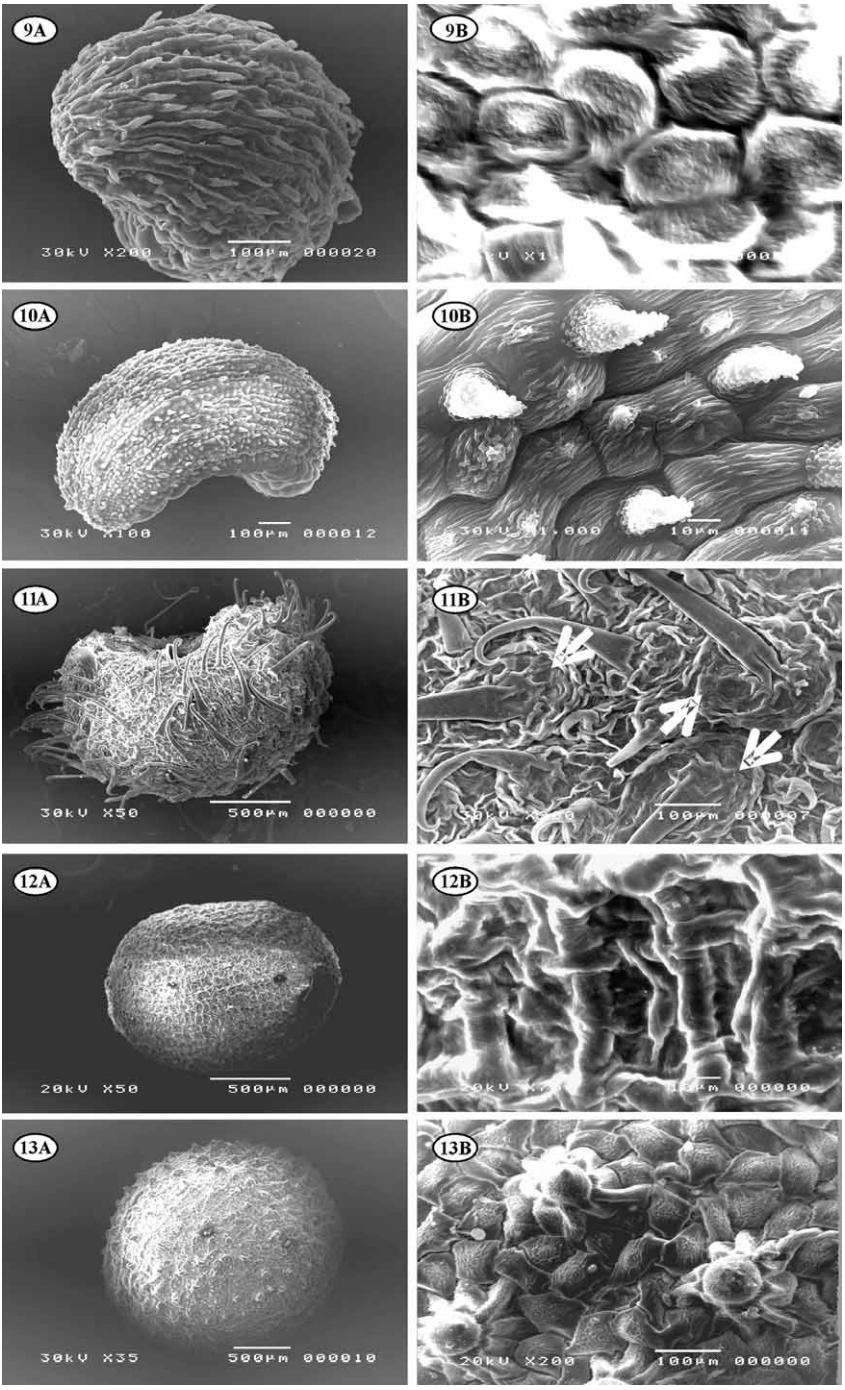

Fig. 3. SEM photographs of mericarps. A – entire mericarps, B – enlargement of mericarps coat. 9 – *Galium setaceum* subsp. *Decaisnei*, 10 – *G. sinaicum,* 11 – *G. spurium* subsp. *spurium*. va.r *hirsutum,* 12 – *G. spurium* subsp. *spurium*. var. *spurium, 13.*– *G. tricornutum.*

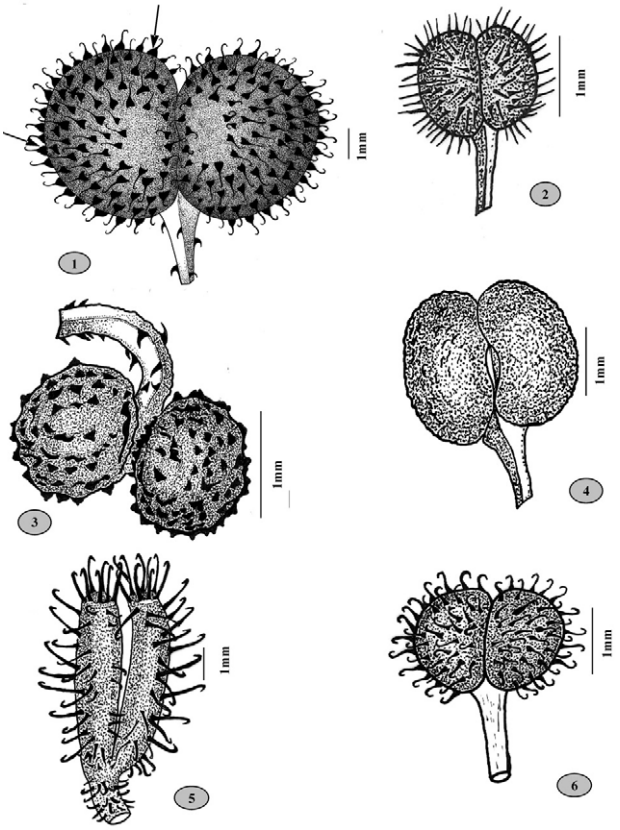

Fig. 4. Mericarp shape of *Galium* species. 1 – *G. aparine*. 2 – *G. canum*, 3 – *G. ceratopodum*, 4 – G. *mollugo*, 5 – G. *murale*, 6 – *G. nigricans*.

siense, *G. setacium* subsp. *setacium, G. setacium* subsp. *decaisnei* and *G. sinaicum* (Figs. 6.2, 6.4, 7.6, 7.8, 8. 9, 10). Seed shape showed significant differences among species, but no significance difference among subspecies or varieties.

Seed size

Seeds sizes vary greatly among the examined taxa. The largest seeds in *Galium aparine* and *G. tricornutum* have a diameter 2.0–4.8 × 2.0–4.8 mm (Figs. 6.1A, 8.13A) and the smallest seeds measure 0.4–1.3 × 0.2–0.9 mm in *G. canum*, *G. nigricanse*, *G. parisiense*, *G. setacium* and *G. sinaicum* (Figs. 6.2A, 7.6A, 7A, 8A, 8.9A, 10A), while the rest of the species have slightly larger seeds, 1.0–2.3 × 0.2–2.3 mm, *G. ceratopodum, G. mollugo, G. murale* and *G. spurium* (Figs. 6.3, 4, 7.5, 8.11, 12).

Seed colour

The colours of seeds are of less diagnostic and systematic value among species. The colour of seeds varies from black, dark brown, yellow brown. They are black in *Galium mu-*

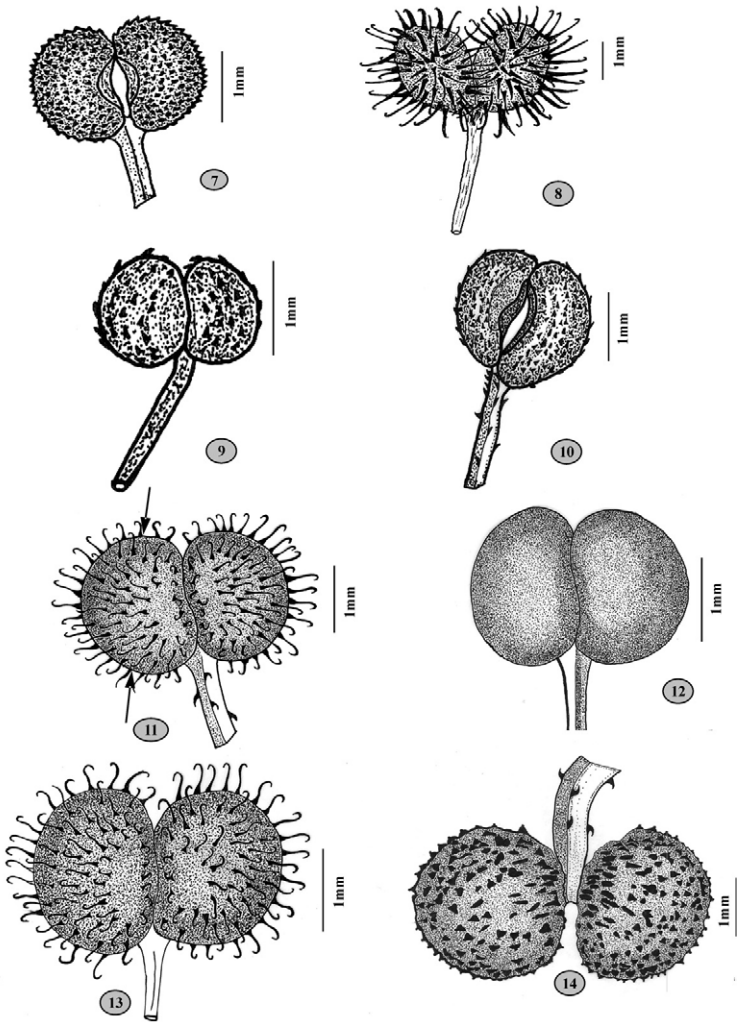

Fig. 5. Mericarp shape of *Galium* species..7 – *G. parisiense*, 8 – *G. setacium* subsp. *setacium*, 9 – *G. setacium* subsp. *decaisnei*, 10 – G. *sinaicum*, 11 – G. *spurium* subsp. *spurium* var. *hirsutum*, 12 – *G. spurium subsp*. *spurium* var. *spurium* 13 – *G. spurium* subsp. *africanum*, 14 – *G. tricornutum*.

rale and *G. nigricanse,* dark brown in *G. aparine*, *G. canum*, *G. parisiense*, *G. spurium* and *G. tricornutum*, yellow brown in *G. setacium* and *G. sinaicum*. It's difficult to distinguish between species depending on the colour of seed, because of the narrow range of colour degree between species, and most species have the same seed colour.

Epidermal cells

The cellular shapes can be of considerable diagnostic and systematic value. Epidermal cells of seed coats exhibit random arrangement or arrangement in parallel rows. They exhi-

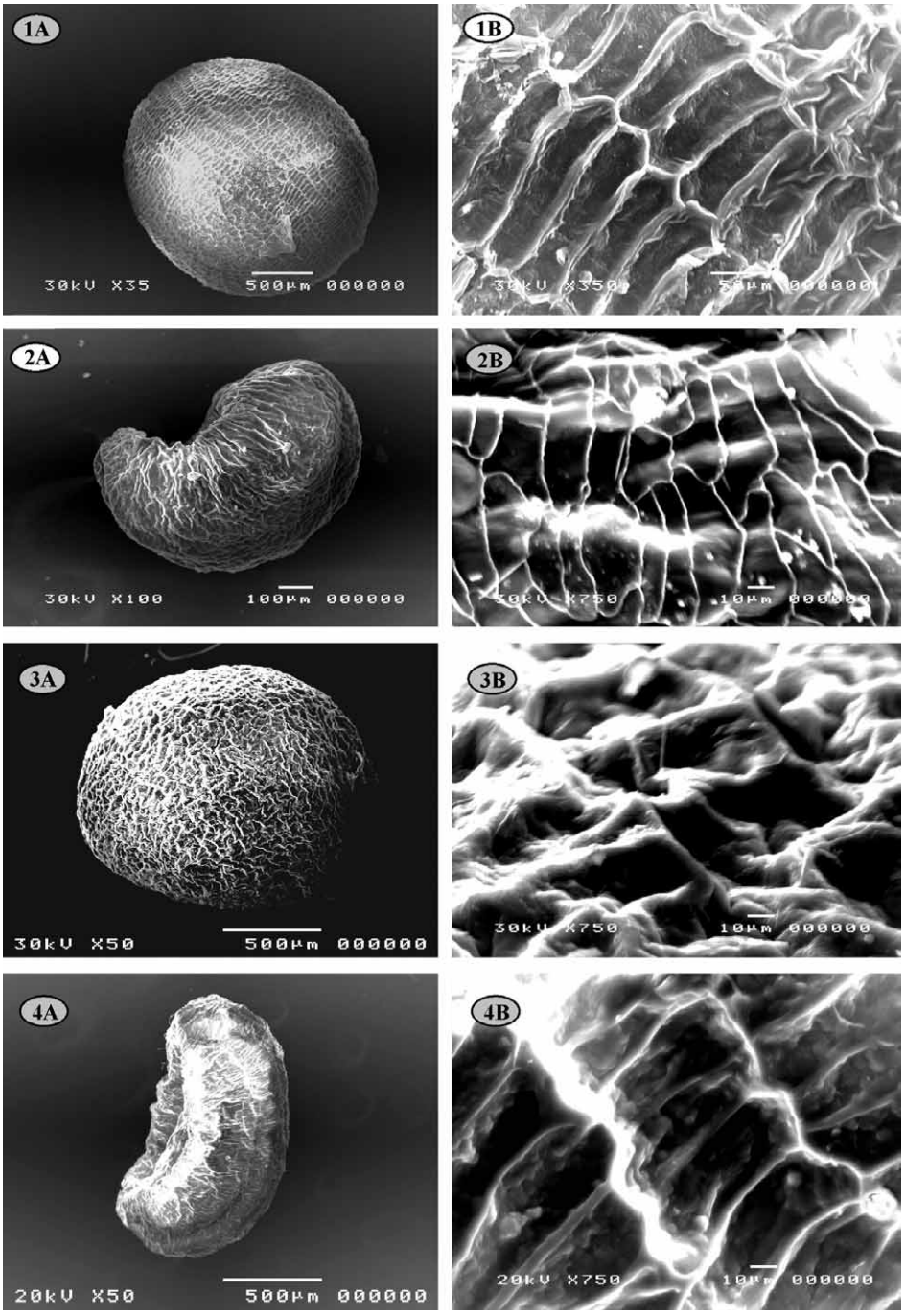

Fig. 6. SEM photographs of seeds. A – entire seeds, B – enlargement of seed coat. 1 – *Galium aparine*, 2 – *G. canum*, 3 – *G. ceratopodum,* 4 – *G. mollugo*.

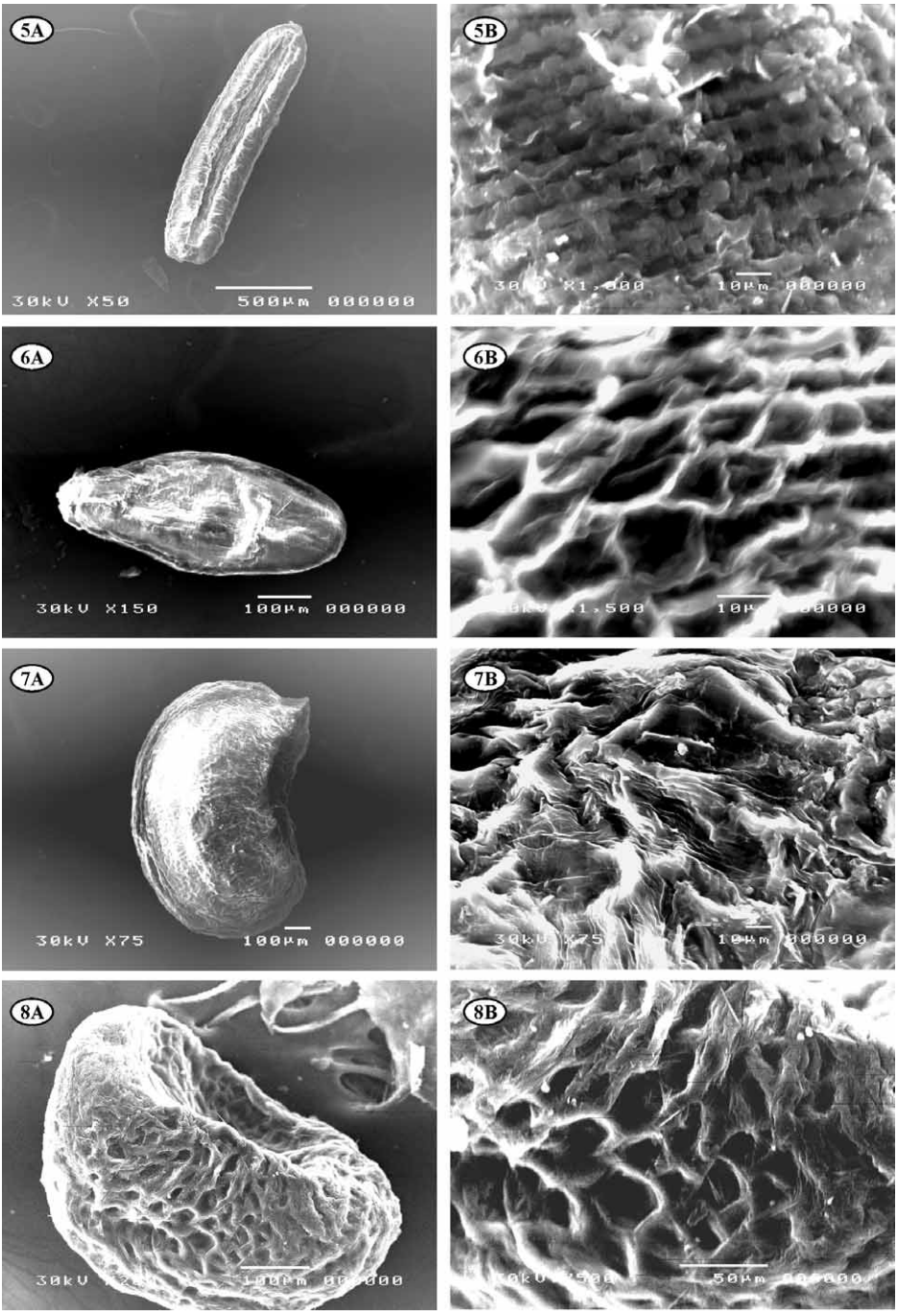

Fig. 7. SEM photographs of seeds. 5 – *Galium murale*, 6 – *G. nigricans*, 7 – *G. parisiense*, 8 – *G. setaceum* subsp. *setaceum*.

bit arrangement in parallel rows in the following species, *Galium aparine, G. canum, G. mollugo, G. murale, G. nigricans* and *G. sinaicum* (Figs. 6.1B, 2B, 4B, 7.5B, 6B, 8.10B), but in other species they are randomly arranged (Figs. 6.3B, 7.7B, 8.9B, 11B,12B,13B). The cell shapes show significant variation among the species. They are isodiametric, elongate in one direction and 4–6-polygonal, isodiametric in *Galium mollugo*, *G. setacium* subsp. *decaisnei*, *G. sinaicum* and *G. tricornutum* (Figs. 6.4B, 8.9B, 10B, 13B), elongate in one direction in *G. aparine, G. canum, G. mollugo, G. murale, G. nigricans,* and *G. sinaicum* (Figs. 6.1B, 2B, 4B, 7.5B, 6B, 8.10B), 4–5 gonal in *Galium murale*, *G. nigricans*, *G. setacium* subsp. *setacium* and *G. tricornutum* (Figs. 7.5B, 6B, 8B, 8.13B), 5–6 gonal in *Galium aparine*, *G. mollugo*, *G. sinaicum* and *G. spurium* (Figs. 6.1B, 4B, 8.10B, 11B,12B) and polygonal in *G. canum*, *G. ceratopodum*, *G. parisiense* and *G. setacium* subsp. *decaisnei* (Figs. 6.2B, 3B, 7.7B, 8.9B).

Anticlinal cell wall boundaries

There are 3 types of cell wall boundaries: 1. Raised-channelled, straight, smooth in *Galium aparine* (Fig. 6.1); 2. Channelled, sinous, coarse folds in *G. setacium* subsp. *decaisnei* (Fig. 8.9); 3. Raised, straight to sinous, smooth to folded in the rest of the taxa.

Outer periclinal cell wall

Curvature can serve as a good diagnostic character for the lowest taxonomic categories (ABDEL KHALIK et al. 2002). There are three different shapes of outer periclinal cell wall; 1. Convex in *G. setacium* subsp. *decaisnei* (Fig. 8.9); 2. Flat in *G. canum* and *G. parisiense* (Figs. 6.2, 7.7); 3. Flat to slightly concave in the rest of the species (Figs. 6.1, 3, 4, 7.5, 6, 8.10, 11, 12, 13).

Secondary cell wall sculpture

The surface of the outer cell wall shows a great variation among species and subspecies within the genus *Galium* in Egypt. There are 5 different shapes for the surface of the outer cell wall: 1. smooth in *G. canum*, *G. murale* and *G. nigricans* (Figs. 6.2, 7.5, 6); 2. fine folds in *G. ceratopodum* and *G. setacium* subsp. *setacium* (Figs. 6.3, 7.8); 3. coarse folds in *G. parisiense*, *G. setacium* subsp. *decaisnei* and *G. spurium* subsp. *africanum* (Figs. 7.7, 8.9, 12); 4. micro-papillate in *Galium aparine*, *G. sinaicum* and *G. spurium* subsp. *spurium* (Figs. 6.1, 8.10, 11); 5. micro-reticulate in *G. tricornutum* (Fig. 8.13).

Key to the studied species based on seed characters

1a. Seed slender.....*Galium murale*
1b. Seed globose, sub globose or reniform.....2
2a. Seed globose or sub-globose.....3
2b. Seed reniform.....7
3a. Seed globose (2.8–4.5 × 2.8–4.5); anticlinal boundaries raised channelled.....*Galium aparine*
3b. Seed globose or sub globose; anticlinal boundaries raised..... 4

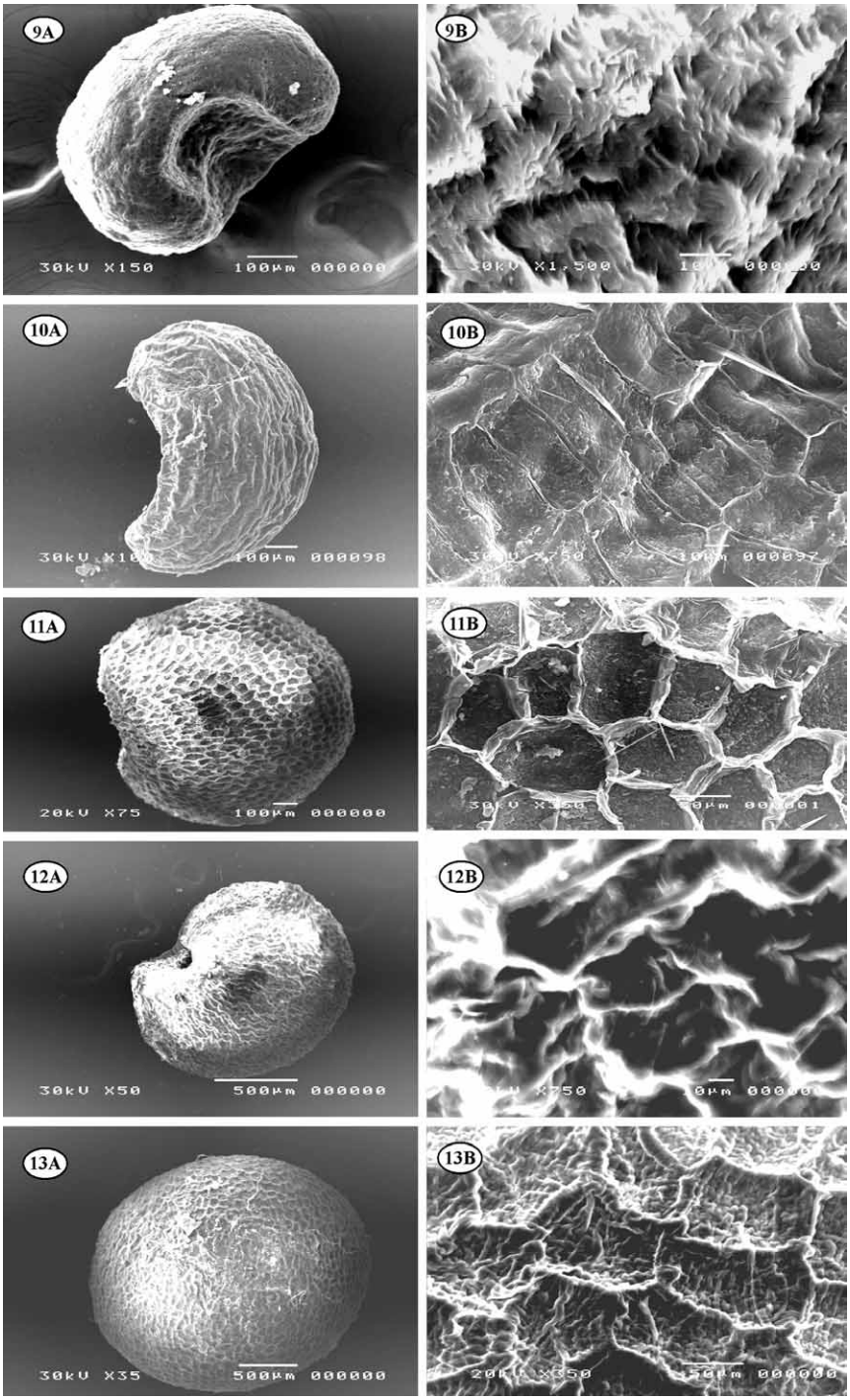

Fig. 8. SEM photographs of seeds. 9 – *Galium setaceum* subsp. *Decaisnei*, 10 – *G. sinaicum,* 11 – *G. spurium* subsp. *spurium*, 12 – *G. spurium* subsp. *Africanum*, 13 – G. *tricornutum*.

4a. Seed globose (2.0–3.7 × 2.0–3.7); sculpture of periclinal cell wall micro-reticulate..... *Galium tricornutum*
4b. Seed subglobose; sculpture of periclinal cell wall fine or coarsely folded.....5
5a. Epidermal cell shape polygonal.....*Galium ceratopodum*
5b. Epidermal cell shape 5,6 gonal.....6
6a. Sculpture of periclinal cell wall micro-papillate.....*Galium spurium* subsp. *spurium*
6b. Sculpture of periclinal cell wall coarsely folded.....*Galium spurium* subsp. *africanum*
7a. Anticlinal boundaries channelled.....*Galium setacium* subsp. *decaisnei*
7b. Anticlinal boundaries raised.....8
8a. Epidermal cell shape polygonal.....12
8b. Epidermal cell shape 4,6 gonal.....9
9a. Periclinal cell wall flat to slightly concave, micro-papillate.....10
9b. Periclinal cell wall flat to slightly concave, smooth or fine folds.....11
10a. Anticlinal boundries raised, straight to slightly sinous, smooth to fine folded.....*Galium mollugo*
10b. Anticlinal boundries slightly raised, straight, smooth.....*Galium sinaicum*
11a. Seed black; epidermal cell shape 4, 5 gonal, elongate in one direction; sculpture of periclinal cell wall smooth.....*Galium nigricans*
11b. Seed yellow brown; epidermal cell shape 4, 5 gonal; sculpture of periclinal cell wall fine folded…..... *Galium setacium* subsp. *setacium*
12a. Epidermal cell shape polygonal, elongate in one direction; anticlinal boundaries smooth; sculpture of periclinal cell wall smooth.....*Galium canum*
12b. Epidermal cell shape polygonal; anticlinal boundries coarse folded; sculpture of periclinal cell wall coarse folded.....*Galium parisiense*

Discussion

The scanning electron microscope has been used by various workers during the past few years for viewing details of the surface structure of pollen and seed. ECHLIN (1968) showed the effectiveness of SEM in illustrating the seed coat of two subspecies of *Arenaria ciliata*. Scanning electron microscopy of seeds would become a routine technique for furnishing information on seed coat morphology (HEYWOOD 1969). ABDEL KHALIK (2002) recommended SEM in his studies of the structure of seed and pollen ornamentation in the Brassicaceae as being useful for taxonomic studies.

Our results seems to prove that there exists a correspondence between the mericarps and seed characters and the traditional classification within the subsections. The sections of *Galium*, however, seem to be quite artificial. They have been based on traditional characters of high practical value, but of low relevance concerning phylogenetic relationships, such as life span, position of inflorescence, pedicel shape, stem characters, corolla colour. In our investigations we show the importance of the ornamentation of the mericarps and seed coats.

Based on the main external fruit morphology, two groups have traditionally been distinguished: glabrous and hispid fruits (BOISSIER 1881). In our investigations four types of mericarp surface were observed, one type of which was represented by more than one species generally belonging to different subsections. For example, the hooked-hairs type was dis-

cerned in *G. aparine* which belongs to the subsection Leucarparina (BOISSIER 1881), while *G. murale, G. nigricanse,* and *G. setacium* belong to the subsections Apera and Xanthaparinea**.** Moreover *G. mollugo, G. sinaicum* and *G. parisiense* have micropapillate fruit surface and these taxa belong to different sections: Eugalium and Aparine respectively. On the other hand *G. ceratopodum* and *G. tricornutum* have a tuberculate fruit surface, and they belong to the same subsection, Camptopoda. These results are incongruent with those of NATALI et al. (1995) who concluded that *Galium* species are a monophyletic group based on DNA sequence of the chloroplast atpB-rbcL intergene region.

Mericarp hairs were found useful to distinguish between two closely similar species, based on the base of the hair; however, in *G. aparine* the hooked hair rises from a tuberculate base (Fig. 1.1B, 4.1), but in *G. spurium* from a flat base (Figs. 3.11B, 5.11). Furthermore, it was found useful to separate the subspecies and varieties. However, in *G. setacium* subsp. *setacium* the mericarp was covered with hooked hairs (figs. 2. 8, 5. 8), but in *G. setacium* subsp. *decaisnei* with depressed hairs (Figs. 3.9, 5.9). Moreover in *G. spurium*, it is very significant to distinguish between var. *spurium* with glabrous mericarp (Figs. 3.12, 5.12) and var. *hirsutum* with hooked hairs on the mericarp.

Seed morphological characters were helpful in distinguishing various species (Tab. 1). They do not confirm the sectional classification of the genus *Galium*.

Seed size was found useful to distinguish between *G. aparine* and *G. spurium,* species which have similar morphological characters (. Seeds measure 2.8–4.8 × 2.8–4.8 mm in *G. aparine* but 1.5–2.3 × 1.5–2.2 mm in *G. spurium*. In *G. tricornutum* the diameter is 2.0–3.7 × 2.5–3.5 mm, but in *Galium ceratopodum* 1.6–2.2 × 1.3–2.0 mm.

Our results identify three different types of anticlinal cell wall boundaries and three types of periclinal boundaries, which does not distinguish the sections and subsections proposed by BOISSIER (1881). Furthermore the anticlinal cell wall boundaries showed a great variation between species and subspecies, varying from raised-channelled, straight, smooth in *G. aparine* (Fig. 6.1); slightly raised, straight or slightly sinous, smooth in *G. canum* and *G. sinaicum* (Figs. 6.2, 8.10); raised, straight to sinous, smooth to fine folds in *G. ceratopodum*, *G. mollugo*, *G. nigricans*, *G. setacium* subsp *setacium* and *G. spurium*, (Figs. 6.3, 4, 7.6, 8.11, 12); raised, straight to sinuous, coarse folded in *G. murale*, *G. parisiense* and *G. tricornutum* (Figs. 7.5, 7, 8.13); channelled, sinuous, coarsely folded in *G. setacium* subsp. *decaisnei* (Fig. 8.9). At subspecies level, this character separates two subspecies of *G. setacium*: it is raised, slightly sinuous, smooth to folded in subspecies *setacium* (Fig. 7.8), to channeled, sinous, coarsely folded in subspecies *decaisnei* (Fig. 8.9).

The surface of the outer cell wall was found useful for the distinction of the subspecies of *G. setacium*; there are fine folds in subspecies *setacium* and coarse folds in subspecies *decaisnei*. There is a similar situation with the subspecies of *G. spurium*; the surface is micro-papillate in subspecies *spurium* (Fig. 8.11), and coarsely folded in subspecies *africanum* (Fig. 8.12).

The developmental variations of seed and fruit morphology are worth taking into account, not only because they give us a better comprehension of sculpture development but also for preparing an identification key.

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