

Seed coat micromorphology of some Asiatic Spermaceae (Rubiaceae)

Shu-Miaw Chaw¹ and V. V. Sivarajan²

¹ Institute of Botany, Academia Sinica, Taipei, Taiwan 11529, Republic of China

² Department of Botany, University of Calicut, Kerala 637 635, India

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Abstract. Thirteen species of four tropical genera of Spermaceae (Rubiaceae) in Asia: *Richardia* L. (1 sp.), *Diodia* L. (1 sp.), *Mitracarpus* Zucc. (1 sp.) and *Spermaceae* L. (10 spp.), were studied as to their seed coat surface patterns, using SEM. In *Richardia scabra*, the seed is fused with the mericarp and the surface pattern of the mericarp is distinctive with conspicuously striated papillae. The seeds of *Diodia virginiana* are unique in having baculate testa. In *Mitracarpus* and *Spermaceae*, the seed coats are basically reticulate, but differ in size, shape and nature of the meshes of the reticulum. Seed coat patterns may not be of great taxonomic value in the generic delimitation, which is far from clear in the Spermaceae. Yet, they might lend some support for the recognition of species in genera like *Spermaceae* when taken along with other morphological attributes.

Key words: *Diodia*; *Mitracarpus*; *Richardia*; Seed-coat pattern; *Spermaceae*; Spermaceae.

Introduction

The tribe Spermaceae with about 18 genera distributed in the tropical and temperate regions of the world excluding New Zealand (Verdcourt, 1958), is characterized by a herbaceous habit, setiferous stipules, solitary ovules attached to the middle of the septum in each cell, valvate corolla lobes, dry fruits which are valvular capsules or separating into one-seeded cocci and pluricolpate pollen grains (Verdcourt, 1958; Bremekamp, 1966; Dwyer, 1980; Chaw and Peng, 1987). Four of these genera, namely, *Spermaceae* L. (including *Borreria* G. F. W. Meyer), *Diodia* L., *Richardia* L. and *Mitracarpus* Zucc. that are usually weedy and widespread in tropical Asia have been often confused in the herbaria. Their general morphology and taxonomy have

been elucidated recently by Chaw and Peng (1987) and Sivarajan *et al.* (1987). This paper deals with the seed coat patterns of the 10 species of *Spermaceae* and one species each of *Diodia*, *Richardia* and *Mitracarpus*, available in tropical part of Asia.

Even as there is no difference of opinion as to the generic status of *Richardia* and *Mitracarpus*, the delimitation and circumscription of *Spermaceae* and *Diodia* is still inconclusive. *Spermaceae* L. and *Borreria* G. F. W. Meyer are considered to be distinct genera by many authors, based on the dehiscence of the capsule: both cocci dehiscent at apex in *Borreria*; one coccus indehiscent and adherent to the septum in *Spermaceae* (Meyer, 1818; Hepper and Keay, 1963; Backer and Bakhuizen van den Brink, Jr., 1965; Steyermark, 1972). Bremekamp (1934)

concedes that this character is too small and insufficient for generic delimitation, but retains the two genera for the "purely opportunistic reason" that it is easily recognizable. Most others, however, prefer to combine the two taxa under the generic name *Spermacoce* (Hooker, 1881; Verdcourt, 1975, 1976; Deb and Dutta, 1984; Sivarajan *et al.* 1987; Chaw and Peng, 1987).

After examining the African materials of *Spermacoce* (*sensu lato*) and *Diodia*, Verdcourt (1975) opined that he would have put them together, because their close relationship is very apparent. However, the New World materials have persuaded him to keep *Diodia* as a distinct genus. "Certainly the type species of *Diodia*, *D. virginiana* L., differs widely in its fruit structure from other species included in the genus" (Verdcourt, 1975). Lewis and Oliver (1974) have also acknowledged the fact that the circumscription of the genus *Diodia* is far from conclusive, but consider it to be more closely related to *Richardia* by their fruits which separate into indehiscent cocci. The number of carpels by which these genera are usually distinguished are variable and there are species of *Richardia* having two to four carpels, leading to confusion in the taxonomy of the group. However, the terminal capituli subtended by two to four involucreal leaves and the deciduous calyx demarcate *Richardia* from *Diodia* fairly well.

Verdcourt (1975: 317-322) has concluded that the species of *Mitracarpus* widespread in tropical Africa, S. America and Asia are all conspecific and that the correct name of the species is *M. villosus* (Sw.) DC. Consequently the name *M. verticillatus* (Schum. & Thonn.) Vatke, by which this species is invariably referred to in Indian floras, becomes a synonym of *M. villosus*.

These above-mentioned genera can easily be distinguished as follows:

- 1a. Infl. of terminal capituli subtended by 2-4 invol. leaves; calyx deciduous....*Richardia*
- 1b. Infl. axillary and also terminal, invol. leaves absent; calyx persistent...2
- 2a. Flowers often axillary, solitary; fruits splitting into two indehiscent cocci*Diodia*
- 2b. Flowers several per axil; fruit with 1 or both cocci dehiscent3
- 3a. Capsules circumscissile*Mitracarpus*
- 3b. Capsules 2-valved*Spermacoce*

The present study is aimed at examining whether seed coat surface patterns would provide any reliable taxonomic parameter for generic delimitation or species recognition.

Materials and Methods

Seeds prepared for scanning electron microscopy (SEM) were collected from herbarium sheets using a dissecting light microscope (LM). After cleaning with a fine Chinese brush dipped in distilled water, the seeds were dehydrated through an ethanol series and acetone, critical-point dried with CO₂, coated with gold, and examined with a Zeiss 950 SEM at an accelerating voltage of 15 KV.

Three to eighteen seeds of each sample were mounted and examined. Little variation within samples was obvious. The seed photographed was in all cases judged to be typical for the sample. All voucher numbers of specimens examined are listed after the description of each species. Voucher numbers are given in the legends of Figs. 1-39 only when more than one collection was observed.

Observations

Richardia scabra L.

Figs. 1-3.

Here the seeds are inseparable from the mericarp as they enlarge and fuse with the latter. The mericarps or cocci are obovoid or ellipsoid, 2.5-3.5×1.5-2 mm, and the surface is papillose. The papillae occur in various sizes

and shapes, some of them appearing as buckled protuberances on the mericarp surface; under higher magnifications, the entire surface is seen to be covered by conspicuous, thick, sinuate striations.

Specimen examined: *Chaw 221* (HAST).

Diodia virginiana L. Figs. 4-6.

Fruits separating into 2 indehiscent cocci, but unlike in *Richardia* seeds are fused with the mericarp. Seed much larger than in other taxa, 4.3-5.0×2-3 mm, with a longitudinal ventral groove; surface reticulate under LM, but densely baculate under SEM, the bacula characterized by striations.

Specimen examined: *Hsieh & Chaw 1501* (HAST).

Mitracarpus villosus (Sw.) DC. [= *M. verticillatus* (Schum. & Thonn.) Vatke] Figs. 7-9.

Seeds rhomboid or squarish, with almost truncate ends, brownish, 0.8-1×0.4-0.6 mm with 4 short ventral grooves from the angles connecting the central, longitudinal groove; surface almost smooth under LM, but distinctly reticulate under SEM; meshes not in regular rows, polygonal, isodiametric and depressed, granulate or with closely parallel, faint striations; walls of the reticulum thick and raised.

Specimen examined: *Nair 26799* (CALI).

Spermace pusilla Wall. [= *Borreria pusilla* (Wall.) DC.] Figs. 10-11.

Seeds oblong-ellipsoid with rounded ends, dark brown, or brownish black, 1-1.8×0.5-0.7 mm, with longitudinal ventral groove; surface smooth under LM, but faintly reticulate under SEM; meshes not in regular rows, polygonal, flat or slightly depressed, isodiametric or longish, smooth without granules or striations; walls thin, faint and a little raised.

Specimen examined: *Joseph 2799, Sivarajan 68* (CALI).

S. ramanii Sivar. & Nair. [= *Borreria stricta* auct. non (L. f.) K. Schum.] Figs. 12-13.

Seeds oblong-ellipsoid, light brown with rounded ends, ca. 1.5×8 mm, with a longitudinal, ventral groove; surface smooth under lower magnifications, faintly reticulate at higher magnification; meshes polygonal, not in regular rows, irregularly shaped, not depressed, smooth without granules or striations, walls raised, thin, but more conspicuous than in *S. pusilla*.

Specimen examined: *Sivarajan 625* (CALI).

S. malabarica (Sivar. & Mani.) Sivar. et al. [*Borreria malabarica* Sivar. & Mani.] Figs. 14-16.

Seeds brownish, ellipsoid with rounded ends, ca. 2.4×1.2 mm, with a ventral groove; seed surface minutely reticulate, meshes polygonal, not in regular rows, isodiametric or longish, depressed and with closely parallel, faint striations. Walls fairly thick and raised.

Specimen examined: *Sivarajan 36428* (CALI).

S. articularis L. f. [= *Borreria articularis* (L. f.) Williams]

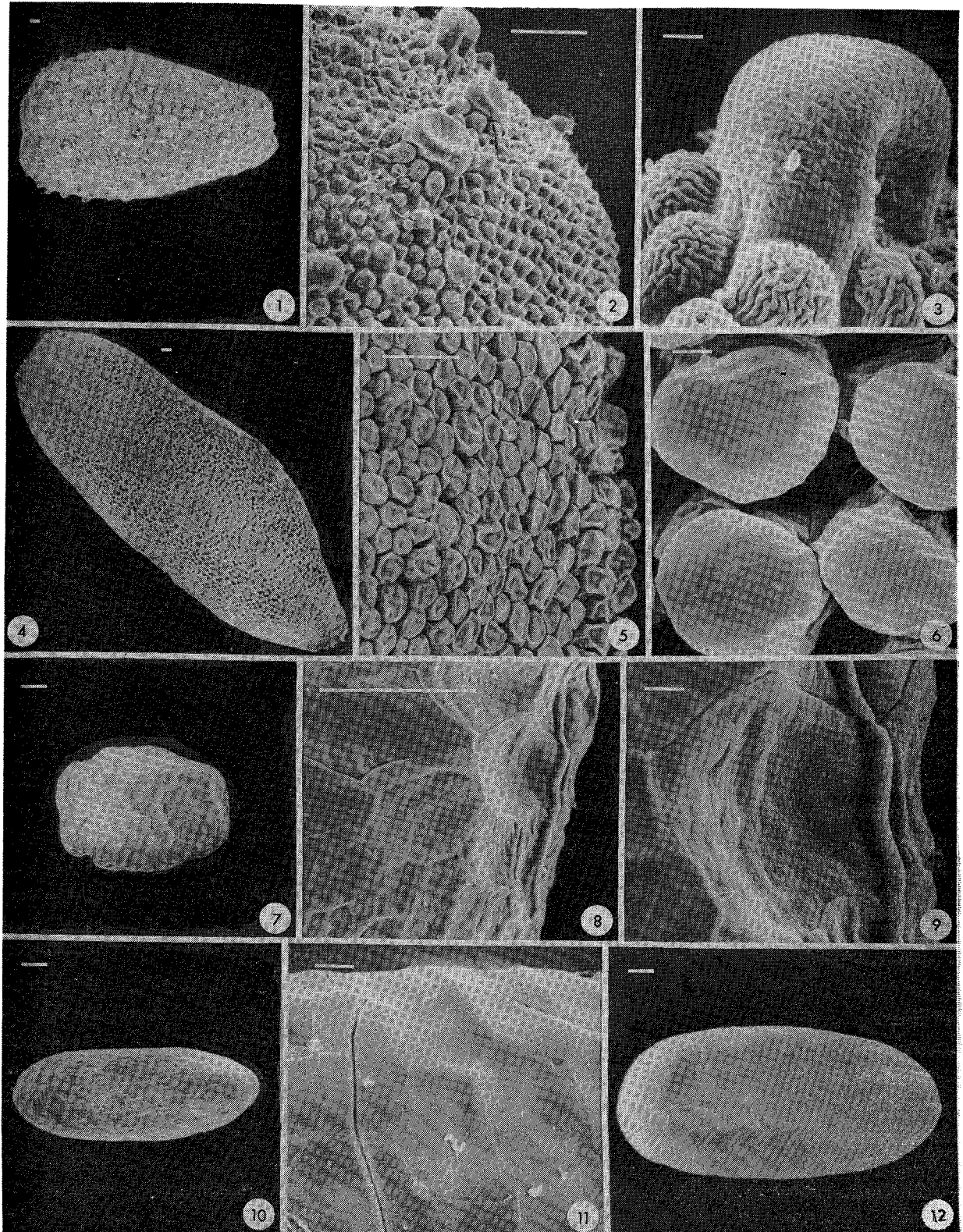
TYPE I. Figs. 17-18.

Seeds ellipsoid with rounded ends and a longitudinal ventral groove, brownish, ca. 2.5-3.5×1.4-1.8 mm, smooth under LM, reticulate under SEM; meshes polygonal, not in regular rows, isodiametric or longish, deeply depressed, almost smooth; walls raised, thick, merging with the mesh.

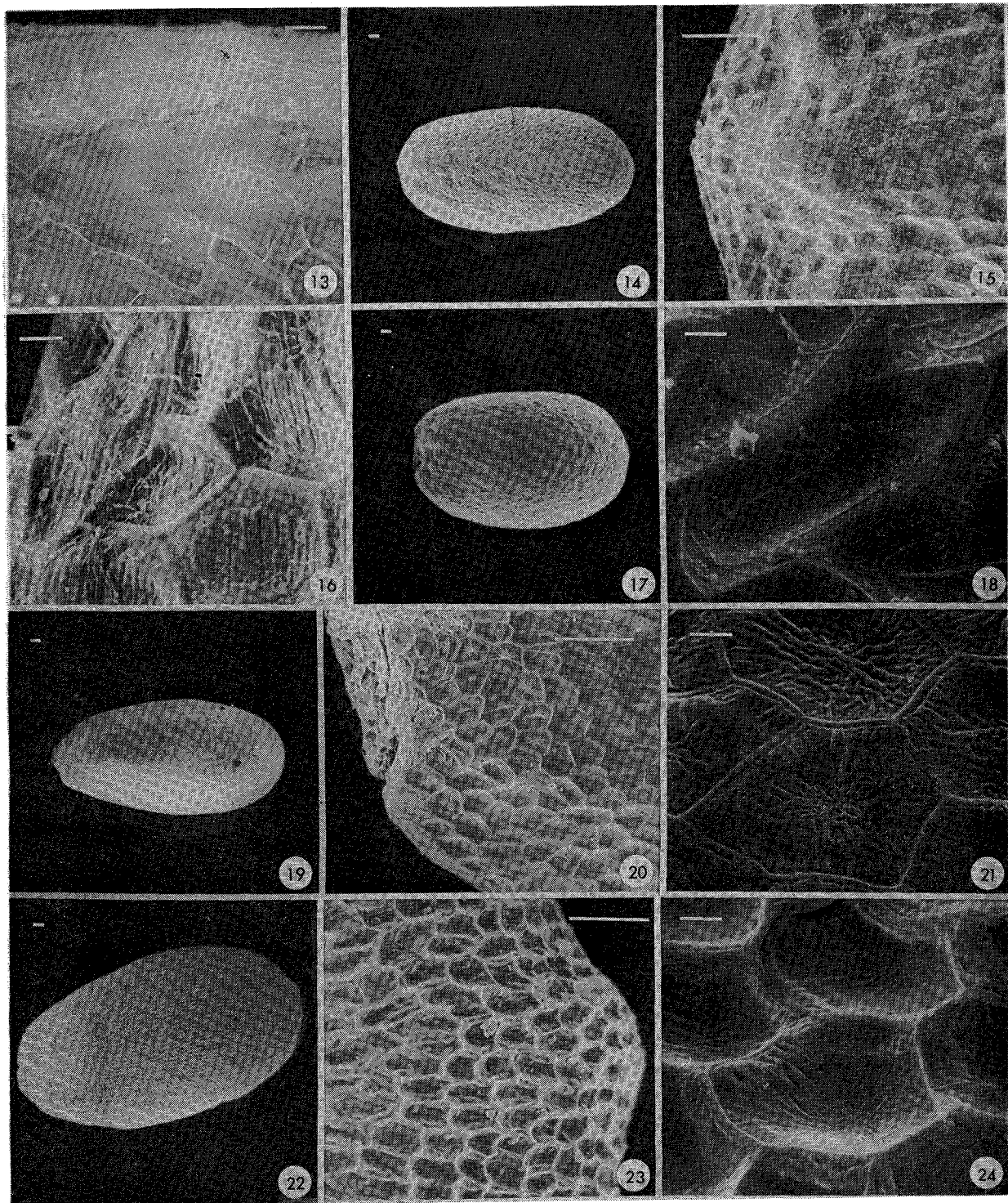
Specimens examined: *Chaw & Peng 321, Chaw 403, 469, 481* (HAST); *Sivarajan 1484* (CALI).

TYPE II. Figs. 19-21.

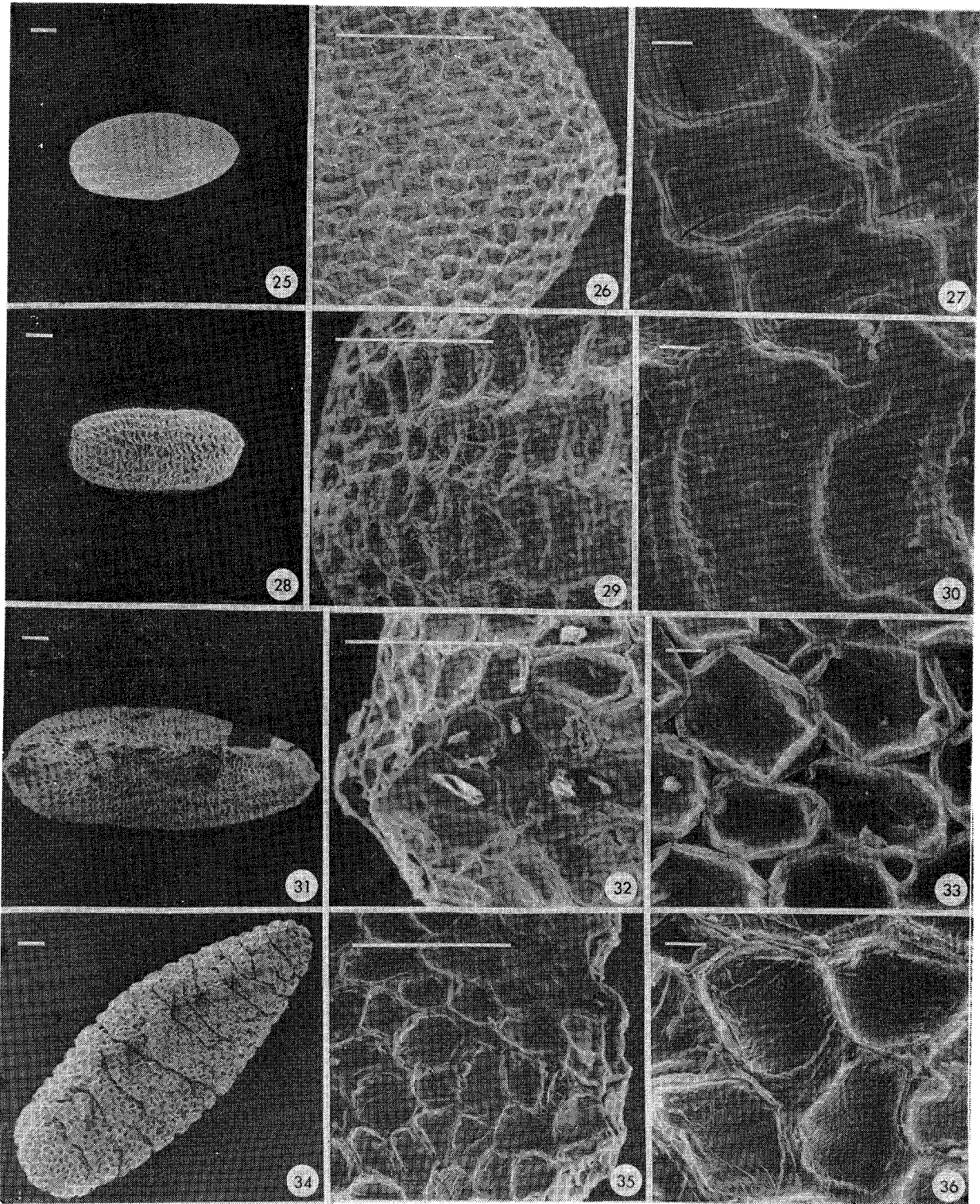
Seeds oblong-ellipsoid with rounded ends, light brown, ca. 2-2.5×1-1.3 mm, surface smooth under LM, but reticulate under SEM; meshes depressed, polygonal, not in regular rows, isodiametric or longish, often with conspicuous, radiating striations from the centre; walls



Figs. 1-12. Scanning electron micrographs of seeds of Spermacoceae. 1-3, *Richardia scabra*; 4-6, *Diodia virginiana*; 7-9, *Mitracarpus villosus*; 10-11, *Spermacoce pusilla*, from Joseph 2779; 12, *S. ramanii*. Scale bars equal 100 μm except that those of 3, 6, 9 and 11 are 10 μm .



Figs. 13-24. Scanning electron micrographs of seeds of *Spermaceae*. 13, *S. ramanii*; 14-16, *S. malabarica*; 17-18, *S. articularis*, type I, from Sivarajan 1484; 19-21, *S. articularis*, type II, from Meenabai 4818; 22-24, *S. hispida*, from CU 24657. Scale bars equal 100 μm except that those of 13, 16, 18, 21 and 24 are 10 μm .



Figs. 25-36. Scanning electron micrographs of seeds of *Spermacoce*. 25-27, *S. latifolia*, from *Sivarajan 464*; 28-30, *S. mauritiana*, from *CU 15904*; 31-33, *S. ocymoides*; 34-36, *S. assurgens*, from *TUA 1698*. Scale bars equal 100 μm except that those of 27, 30, 33 and 36 are 10 μm .

thick, raised and not merging with meshes.

Specimens examined: *Sowmini 1087, Meenabai 4818, Mohanan 11590* (CALI).

S. hispida L. [= *Borreria hispida* (L.) K. Schum.]

Figs. 22-24.

Seeds ovoid or ellipsoid with rounded ends, dark brown or blackish, reticulate under LM and SEM with a longitudinal ventral groove, ca. $2.3-3.2 \times 1.6-1.8$ mm. Meshes not in regular rows, polygonal, isodiametric or longish, deeply depressed, occasionally with faint, radiating striations; walls thick, raised and distinct from meshes.

Specimens examined: *Sivarajan 318, CU 24657* (CALI).

S. latifolia Aubl. [= *Borreria latifolia* (Aubl.) K. Schum.]

Figs. 25-27.

Seeds ellipsoid or ovoid with rounded ends, brownish with a longitudinal, ventral groove, $1.7-2.7 \times 1-1.5$ mm; surface reticulate, meshes polygonal, not in regular rows, deeply depressed, smooth with granules or striations inside; walls thick, raised, often flexuous resulting in meshes of various shapes.

Specimens examined: *Chaw 220, Chaw & Peng 255* (HAST); *Sivarajan 464* (CALI).

S. mauritiana Gideon ex Verdc. [= *Borreria repens* DC.]

Figs. 28-30.

Seeds oblong-ellipsoid with rounded ends, brownish, $0.9-1.1 \times 0.4-0.6$ mm, smooth under LM, distinctly reticulate under SEM; meshes in regular, longitudinal rows, rectangular or polygonal, broader than long, deeply depressed inside with granulate or bullate protuberances; walls thick, raised and distinct.

Specimens examined: *Chaw & Peng 257* (HAST); *Sivarajan 320, CU 15904* (CALI).

S. ocymoides Burm. f. [= *Borreria ocymoides* (Burm. f.) DC.]

Figs. 31-33.

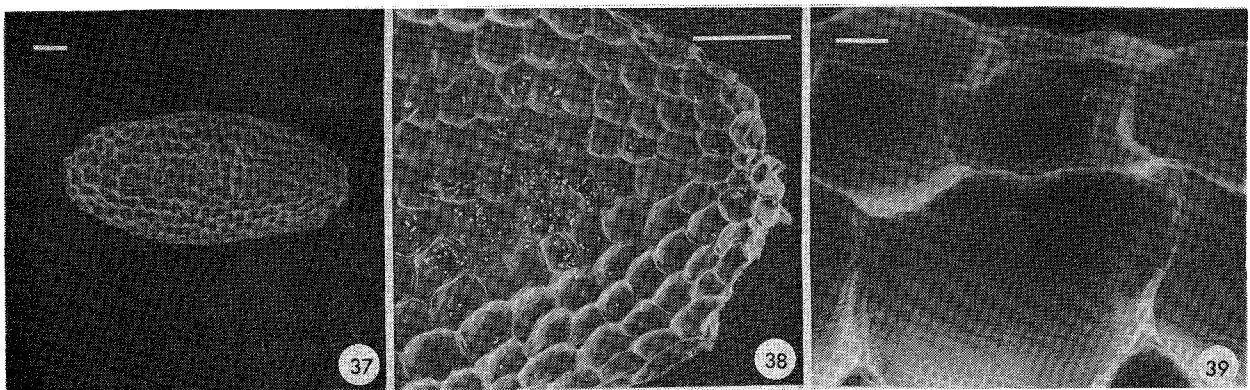
Seeds brownish, oblong-ellipsoid with rounded ends, ca. 1.7×0.7 mm, with a longitudinal, ventral groove; testa appear reticulate at lower magnifications, but as areolate under higher magnifications; meshes or areoles polygonal, not in regular rows, isodiametric or longish, smooth or with fine striations, flat or deeply depressed; walls thick, merging with the meshes.

Specimen examined: *Sowmini 1272* (CALI).

S. assurgens Ruiz & Pavon [= *Borreria laevis* sensu auct. permult.]

Figs. 34-36.

Seeds oblong-ellipsoid with rounded ends, brownish with a longitudinal ventral groove;



Figs. 37-39. Scanning electron micrographs of seeds of *Spermaceae verticillata*. Scale bars of 37 and 38 equal $100 \mu\text{m}$, 39 equals $10 \mu\text{m}$.

seed surface prominently reticulate with transverse grooves; meshes polygonal, mostly isodiametric, deeply depressed with fine striations inside; walls thick, raised and distinct from the mesh.

Specimens examined: *Chaw & Peng 260* (HAST), *TUA 1698* (CALI).

S. verticillata L. [= *Borreria verticillata* (L.) G. F. W. Meyer] Figs. 37-39.

Seeds oblong-ellipsoid, ends rounded, 1.8-2 × 0.8 mm, brownish, ca. 1 × 0.5 mm; surface distinctly reticulate; meshes in almost regular, vertical rows, polygonal, mostly isodiametric, deeply depressed with very faint striations inside; walls thick, raised and distinct from the mesh.

Specimen examined: *Francis 1046* (CALI).

Discussion

SEM studies on the seed coat patterns of these four Asiatic genera show that not much light can be shed on "generic boundaries (which) are far from clear in the Spermaceae" (Lewis and Oliver, 1974). The genus *Richardia* stands apart from all others in regards to the surface patterns of the mericarp. Lewis and Oliver (1974) described the mericarps as "densely papillose and vertically strigillose". Observations at higher magnifications have now contributed another interesting aspect: the papillae themselves are beautifully ornamented by conspicuous, sinuate striations.

Both *Mitracarpus* and *Spermaceae*, the seeds of which were studied, displayed basically similar, reticulate seed coat patterns. At the species level, however, this may be useful along with other characteristics. The seed surface reticula in different species vary in terms of shape of cells: they are either isodiametric, transversely or longitudinally elongated, depressed or flat, striated, granulate or smooth

meshed. Walls can be thick or thin, straight, curved or flexuous; they can rise abruptly from the seed surface or merge with the mesh, becoming structurally indistinct.

The genus *Diodia* L. has only a single representative in Asia, i. e. *D. virginiana*, the type species of the genus. While reporting this species from Taiwan, Hsieh and Chaw (1987) have described its seeds as reticulate. However, the present study reveals that the largest seeds of *D. virginiana* are unique among the four Asiatic genera in their baculate surface cells. Verdcourt (1975, pl. 42) has provided SEM micrographs of the seed coat patterns of another species of the genus *D. aulacosperma*. This closely resembles the transverse groove pattern of *S. assurgens*. However these grooves are regularly spaced and extended all around the seed except for the ventral groove, whereas the grooves of *S. assurgens* are not regular nor do they completely encircle the seed.

In *Spermaceae*, the linear-lanceolate-leaved *S. pusilla* and *S. ramanii* have a distinctive type of reticulum with very faint walls, rising abruptly from an otherwise smooth seed surface. While the rest, i. e. the broad-leaved species, the walls are thick and meshes depressed to varying degrees. *S. malabarica*, however, agrees more with the broadleaved species rather than the linear-lanceolate-leaved ones. *Spermaceae articularis* L. f. is a variable species, even after the exclusion of *S. hispida* L. It displays almost continuous variations from an erect or prostrate form with non-flexuous leaves and long tubular flowers (*Meenabai 4818*, CALI) to a sub-erect or prostrate form with slightly flexuous leaves and short, campanulate flowers (*Sivarajan 1484*, CALI). The present study also points to a possible high degree of infraspecific variability. The former form has testa cells almost isodiametric and the meshes of the reticulum are often beset with radiating striae (Figs. 19-21), while the latter has fairly

elongated meshes without such striae (Figs. 17 and 18). Perhaps more extensive sampling and analysis of the intermediate populations might reveal more of variations in seed-coat patterns also.

Spermaceae hispida and *S. articularis* are closely related and have been even considered conspecific, until Sivarajan and Nair (1986) segregated them. In external morphology one may relate the two species through the flexuous-leaved form with short campanulate flowers. However, the seed-coat pattern studied shows otherwise, i. e. the prostrate form with non-flexuous leaves and long tubular flowers, display a more closely related seed-coat pattern to *S. hispida*, rather than the other form which is otherwise more similar to the latter.

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Literature Cited

- Backer, C. A. and R. C. Bakhuizen van den Brink, Jr. 1965. Flora of Java. Vol. 2. Groningen.
- Bremekamp, C. E. B. 1934. Notes on the Rubiaceae of Surinam. Rec. Trav. Bot. Neerl. 31: 248-308.
- Bremekamp, C. E. B. 1966. Remarks on the position, the delimitation and the subdivision of the Rubiaceae. Acta Bot. Neerl. 8: 479-481.
- Chaw, S. M. and C. I. Peng. 1987. Remarks on the species of Spermaceae (Rubiaceae) of Taiwan. J. Taiwan Mus. 40: 71-83.
- Deb, D. B. and R. Dutta. 1984. Revision of the genus *Spermaceae* (Rubiaceae) in India and adjoining regions. J. Econ. Tax. Bot. 5: 1037-1063.
- Dwyer, J. D. 1980. Flora of Panama, Family 179. Rubiaceae—Part I. Ann. Missouri Bot. Gard. 67: 1-256.
- Hepper, F. N. and R. W. J. Keay. 1963. Rubiaceae. In J. Hutchinson and J. M. Dalziel (eds.), Flora of West Tropical Africa. Vol. 2. London, pp. 218-222.
- Hooker, J. D. 1881. The Flora of British India. Vol. 3. Kent, pp. 199-201.
- Hsieh, C. F. and S. M. Chaw. 1987. *Diodia virginiana* L. (Rubiaceae) in Hsinchu: New to Taiwan. Bot. Bull. Acad. Sinica. 28: 43-48.
- Lewis, W. H. and R. L. Oliver. 1974. Revision of *Richardia* (Rubiaceae). Brittonia 26: 271-301.
- Meyer, G. F. W. 1818. Primitiae Florae Essequiboensis. Göttingen, p. 79.
- Sivarajan, V. V. and R. Vasudevan Nair. 1986. Taxonomic and nomenclatural notes on the *Spermaceae hispida-articularis* complex and *S. stricta-pusilla* complex (Rubiaceae). Taxon 35: 363-369.
- Sivarajan, V. V., R. Vasudevan Nair, and T. U. Ahmed Kunju. 1987. The genus *Spermaceae* L. (Rubiaceae) in India. Proc. Indian Acad. Sci. (Plant Sci.) 97: 347-358.
- Steyermark, J. 1972. 39. *Borreria* G. F. W. Meyer. The Botany of the Guyana Highland—Part IX. Mem. New York Bot. Gard. 23: 805-831.
- Verdcourt, B. 1958. Remarks on the classification of the Rubiaceae. Bull. Jard. Bot. Brux. 28: 209-281.
- Verdcourt, B. 1975. Studies in the Rubiaceae-Rubioideae for the Flora of Tropical East Africa: I. Kew Bull. 30: 247-326.
- Verdcourt, B. 1976. Rubiaceae I. In R. M. Polhill (ed.), Flora of Tropical East Africa. London, pp. 339-379.

亞洲茜草科鴨舌癩舅族植物之種皮微細形態

趙淑妙¹ V. V. Sivarajan²¹中央研究院植物研究所 ²印度 Calicut 大學植物系

茜草科的鴨舌癩舅族 (Spermacoceae) 約有18屬，除紐西蘭外，廣佈於世界的熱帶和亞熱帶地區。本文以掃描式電子顯微鏡觀察亞洲地區的4屬13種該族植物——擬鴨舌癩屬 (*Richardia* L.) 1種，鈕扣草屬 (*Diodia* L.) 1種，*Mitracarpus* Zucc. 屬1種，鴨舌癩舅屬 (*Spermacoce* L.) 10種等的種皮形態，以探討本族內的屬和種之間的界定。

擬鴨舌癩的種子和分果并結合，且果皮的乳頭狀突起有明顯的條紋而別於其他的3屬。大鈕扣草 (*D. virginiana* L.) 的種皮細胞為圓柱狀，相當特別。其餘的2屬，基本上種皮細胞呈網狀，但不同種之間網格的大小和形狀則有所差異。*Mitracarpus* 屬的種子為長斜方體，其腹面的四個頂角有溝槽和中心的縱溝滙合。以種皮的性狀配合其他形態特徵可以容易的區分鴨舌癩舅屬內的種類。鴨舌癩舅 (*S. articularis* L. f.) 的種子有2型，是否另有一變種存在，須再作進一步的研究。