

## Scanning Electron Microscopy of some selected south Indian taxa of Marchantiales (Bryophyta: Hepaticae)

Afroz Alam<sup>1\*</sup>, Vinay Sharma, Praveen Kumar Verma<sup>3</sup> and Krishna Kumar Rawat<sup>4</sup>

<sup>1,2</sup>Department of Bioscience and Biotechnology, Banasthali University, Tonk-304 022 (Rajasthan), India

e-mail: afrozalamsafvi@gmail.com

<sup>3</sup>Rain Forest Research Institute, Deovan Sotai Ali, Post Box # 136 Jorhat – 785 001 (Assam), India

e-mail: pkverma\_bryo@yahoo.co.in

<sup>4</sup>CSIR-National Botanical Research Institute, Rana Pratap Marg, Lucknow – 226 001 (Uttar Pradesh), India

e-mail: drkkrawat@rediffmail.com

\*Corresponding author's email: afrozalamsafvi@gmail.com

**Abstract:** Scanning electron microscope analyses of sporoderm of 7 species of 3 genera of Marchantiales are presented in this study. In the studied species, sporoderm exhibit a broad range of morphology and have taxonomic specificity. The structurally rigid sporoderms have specific sculpturing and characteristic patterning. Peculiar architectures are found in different taxa, which revealed that and stable architectures may be present within an order. Sporoderms usually have specific surface markings which are often one of the best parameters to identify any species.

**Keywords:** Liverwort, Marchantiales, Scanning Electron Microscopy, Sporoderm

### 1. Introduction

In the sexual mode of reproduction of the liverwort, the spore is the first cell of the gametophytic generation. The sporoderm has a patterned and often elaborately sculptured exterior executed in that highly resistant material, sporopollenin (Taylor et al., 1974). Spore size and distinctive morphology of the sporoderm have long been used as a basis of identification of species in certain genera (Muller, 1954). In context of south India, Alam and Srivastava, (2009, 2012) studied the spores and elaters of few taxa using SEM. However, there is still lot of scope of SEM study in different taxa reported from Western Ghats (south India). Hence, we are presenting scanning electron microscope analyses of sporoderm of 7 species of 3 genera of Marchantiales from South India. In the studied species, sporoderm exhibited a broad range of morphology with taxonomic specificity. The structurally rigid sporoderm have definite sculpturing and distinctive patterning. Peculiar architectures are found in different taxa, which revealed that diversified yet stable architectures may be present within an order of liverworts. It proves that sporoderm usually has specific surface markings which can be taken as one of the best parameters to identify any taxa at species level.

## 2. Materials and Methods

### Materials:

#### Representative specimens examined:

*Asterella khasyana*: Tamil Nadu: Nilgiri Hills -Ootacamund (on way to Pykara water fall), ca. 1900m, 28.11.2001, P. K. Verma and Afroz Alam, 14463/2001 (LWU).

*Targionia lorbeeriana*: Tamil Nadu: Nilgiri Hills - Gudulur (Yellamalai); ca. 1200m; 29.09.2002; P. K. Verma and Afroz Alam; 16139/2002 (LWU).

*Targionia hypophylla*: Tamil Nadu: Nilgiri Hills - Kotagiri (Sholarmattum); ca 1900 m; 05.04.2002; P. K. Verma and Afroz Alam; 15073/02 (LWU).

*Plagiochasma cordatum*: Tamil Nadu: Nilgiri Hills - Ootacamund (on way to Love Dale), ca. 2000m, 30.11.2001, P. K. Verma and Afroz Alam, 14638/2001 (LWU).

*P. rupestre*: Tamil-Nadu Nilgiri hills – Coonoor (on way to Mettupalayam- Marapapallam); ca. 1200-1400m. 03.12.2001; P. K. Verma and Afroz Alam; 14772/2001 (LWU).

*P. udarii*: Tamil Nadu: Palni hills - Kodaikanal, ca. 1900m, October, 2000, S.C. Srivastava and Party. 13097/2000 (LWU).

*P. pterospermum*: Tamil Nadu: Nilgiri Hills -Avalanche, ca. 1900-2100m, 09.10.2000, S. C. Srivastava and party, 12582/2000 (LWU).

### Method:

A slightly modified method of Ireland (1992) was used. Spores of selected species were obtained from mature capsules collected from various localities of Nilgiri and Palni hills, South India. Voucher specimens are deposited in hepatic herbarium, University of Lucknow (LWU). The air dried capsules were incised; spores were dusted on aluminum stubs and glow-discharged with a thin layer of metallic gold. After coating, the mounted specimens were either stored in desiccators or examined immediately using SEM.

## 3. Observations

1. *Asterella khasyana* (Griff.) Pande, K.P. Srivast. & Sultan Khan, *J. Hattori Bot. Lab.* 11:7 (1954) 'khasiana'; Long, *Bryophyt. Biblioth.* 63: 169 (2006). – *Fimbralaria khasyana* (Griff.) Mitt. *J. Proc. Linn. Soc. Bot.*, London 5: 126 (1861) 'khasiana'. *Fimbralaria butleri* Steph., *Sp. Hepat.* 6: 12 (1917), - *Asterella butleri* (Steph.) Pande *et al.* ex Parihar, *University of Allahabad Studies*, Botany Section 1961-62: 27 (1962). – *Fimbralaria papulosa* Steph., *Sp. Hepat.* 6: 16 (1917). – *Asterella papulosa* (Steph.) Pande *et al.* ex Kachroo, *J. Hattori Bot. Lab.* 19: 4 (1958). – *Asterella blumeana* auct. plur. non (Nees) Kachroo, *J. Hattori Bot. Lab.* 12: 36-38 (1954); Grolle, *Flora of E. Himalaya* 3: 241 (1975); Kachroo, *J. Indian bot. Soc.* 56: 73 (1977); Gupta & Udari, *Bryophyt. Biblioth.* 29:69 (1986). – *Fimbralaria blumeana* auct. plur. non Nees; Stephani, *Sp. Hepat.* 89 (1899); Kashyap, *Liverw. W. Himal. Panjab pl.* 1: 62 (1929-1934). (**Plate 1, figs. A,B**)

#### Spore under SEM:

Spore trilete, 85-100 µm in size. Distal face with large, primary reticulation, alveolae with shallow pits, 25-30 µm in diameter, containing small secondary reticulation, alveolae and pits. The proximal face has prominent triradiate mark distinct in the central area, faint near the equatorial rim. Large primary reticulations absent, small secondary reticulation with numerous alveolae and pits, covering entire proximal surface. Equatorial rim prominent, without equatorial pore.

**Note:** The size of primary large reticulations on distal surface is notable. Long (2006), in his world monograph on *Asterella*, mentioned their diameter range as 11-23 µm, whereas in our sample, it ranged 25-30 µm. However, the size of spores corresponds to the size mentioned by Long (2006).

2. *Targionia lorbeeriana* Müll., *Rabenh. Krypt*; f1. Ed. 3, 6: 761-320. (1940); Udari & Gupta, *J. Indian Bot. Lab.* 6 (2); 215-219 f1 43 (1983). (**Plate 1, Figs.: C, D**)

#### Spores under SEM:

Spores spherical, 30-35 µm in diameter, distal face has large reticulate lamellae, forming complete to rarely incomplete reticulations. Alveolae 3-6 across the diameter, surface faintly granulate to

striate; Equatorial rim present, small, undulating; proximal face with irregular small to large lamellae, surface granulate, triradiate mark usually inconspicuous, rarely with folded lamellae.

**3. *Targionia hypophylla*** L., *Spec. Plant* 1136. (1753); Steph., *Spec. Hep.* 1: 61:1(1900); Macv., *Std. Handb. Brit. Hep.* 1: 33. F. 1-3. 1926; Kash., *Livr. W. Himalaya & Punjab Pl.* 1: 57 (1929). - *Targionia michellii* Corda in *Opiz Beitr.* 1: 649 (1829). (**Plate: 1, Figs.: E, F**)

**Spores under SEM:**

Spores spherical, trilete, 45-55 µm in diameter, distal face double sculptured, primary reticulations with large alveolae, reticulations mostly complete, rarely incomplete; lamellae surface with minute secondary reticulation, large alveolae 3-6 across diameter, deeply pitted with large, shallowly pitted to flat secondary reticulations in pits with smooth to granulate surface texture; equatorial rim inconspicuous to small, undulating; proximal face with numerous, small, irregular lamellae, seldom forming small reticulations; triradiate mark inconspicuous.

**4. *Plagiochasma cordatum*** Lehm. et Lindenb., *Nov. Strip. Pug.* 4: 13 (1832); Kashyap, *Livr. W. Himalaya and Punjab Plains.* I: 81 (1929). (**Plate: 2, Figs.: A, B**)

**Spore under SEM:**

Spores trilete, 50-65 µm in diameter, with or without equatorial pores. Distal face bulging, with coarse, wavy, irregularly distributed ridges, surface minutely papillate. Equatorial rim prominent, more pronounced in proximal view, slightly wavy, minutely papillate on margins also. Proximal face with distinct triradiate mark with lamellae reaching near the periphery. In each spore facet, a pronounced single lamellae present parallel to the margin of spore, minute papillae are present mostly near the centre and equatorial rim.

**5. *Plagiochasma rupestre*** (G. Forst.) Steph., *Sp. Hepat.* 1:80 (1899); *Aytonia rupestris* Forst., *Char. Gen. Pl.*: 148 (1776); *Plagiochasma nepalense* (Lehm. et Bisch.) Steph., *Spec. Hep.* 1:81. (1898). *Plagiochasma simlense* Kash. (*P. simlensis*) *J. Bombay. Nat. Hist. Soc.* 25: 279 (1917). (**Plate: 2, Figs.: C, D**)

**Spore under SEM:**

Spores trilete, 65-75 µm in size, dimorphic; conspicuously folded and spinulate, somewhat anisopolar, distal surface with coarse, wavy, irregular lamellae, seldom forming reticulation. Equatorial rim present, prominent in proximal view, without equatorial pores, minutely lamellate on margin. Proximal surface with prominent triradiate mark and one or more prominent lamellae on each facet parallel to equatorial rim, surface minutely papillate, mostly near centre and equatorial rim.

**6. *Plagiochasma udarii*** Alam, A & S.C. Srivast. *Indian Journal of Forestry* vol. 32(4): 623-634. (2009). (**Plate: 2, Figs.: E, F**)

**Spore under SEM:**

Spores spherical to triangular, trilete, 55-62 µm in diameter; distal surface coarsely lamellate with large reticulations and deeply pitted alveolae; equatorial rim inconspicuous, without equatorial pores, margin minutely lamellate; proximal face with distinct, coarse lamellae forming large pitted alveolae, triradiate mark prominent with conspicuously raised lamellae, surface minutely papillate (Alam and Srivastava, 2009; 2012)

**7. *Plagiochasma pterospermum*** Mass., *Mem. Acad. Agrie Verona* 73:46 (1897); *P. articulatum* Kashyap, *New Phytol.* 13: 320 (1914). (**Plate: 2; Figs.: G, H**)

**Spore under SEM:**

Spores spherical- tetrahedral, trilete, 41-52 µm in diameter. distal face coarsely lamellate, forming large reticulations, alveolae deeply pitted. Equatorial rim prominent, wavy, with equatorial pores. Minutely lamellate on margin. Proximal surface minutely papillate with distinct folded triradiate

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mark, reaching up to the periphery. Each spore facet with coarse lamellae forming reticulated, pitted alveolae.

#### 4. Discussion and Conclusion

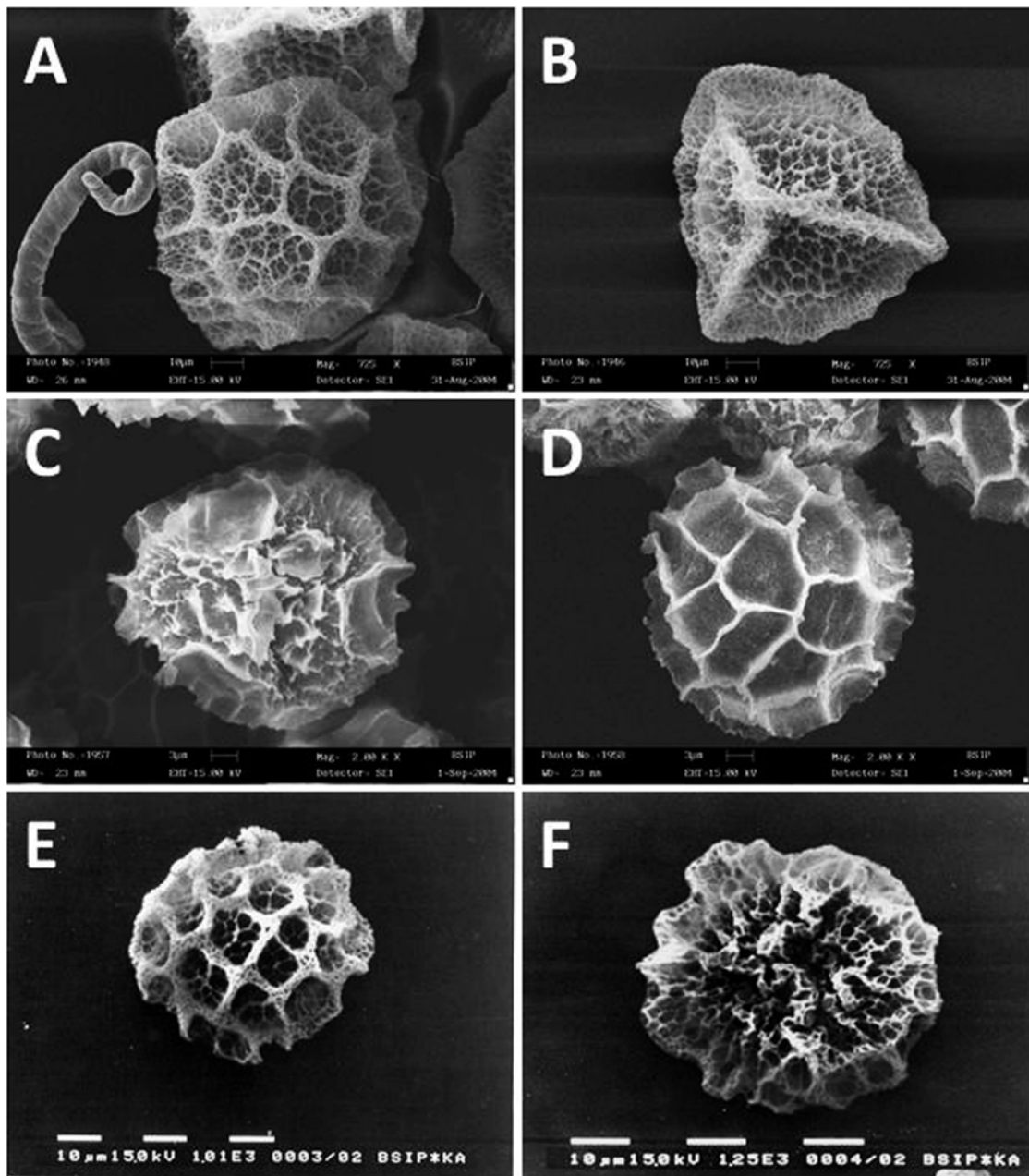
SEM technique is among the vital tools for taxonomists available nowadays to analyze and measure the investigative features at micro-morphological level. It is also evident that SEM technique is appropriate and effectual for characterization of liverwort sporoderm. The elucidation of the SEM observations of varied architectural designs of sporoderm has been immensely helpful in species identification of closely related taxa.

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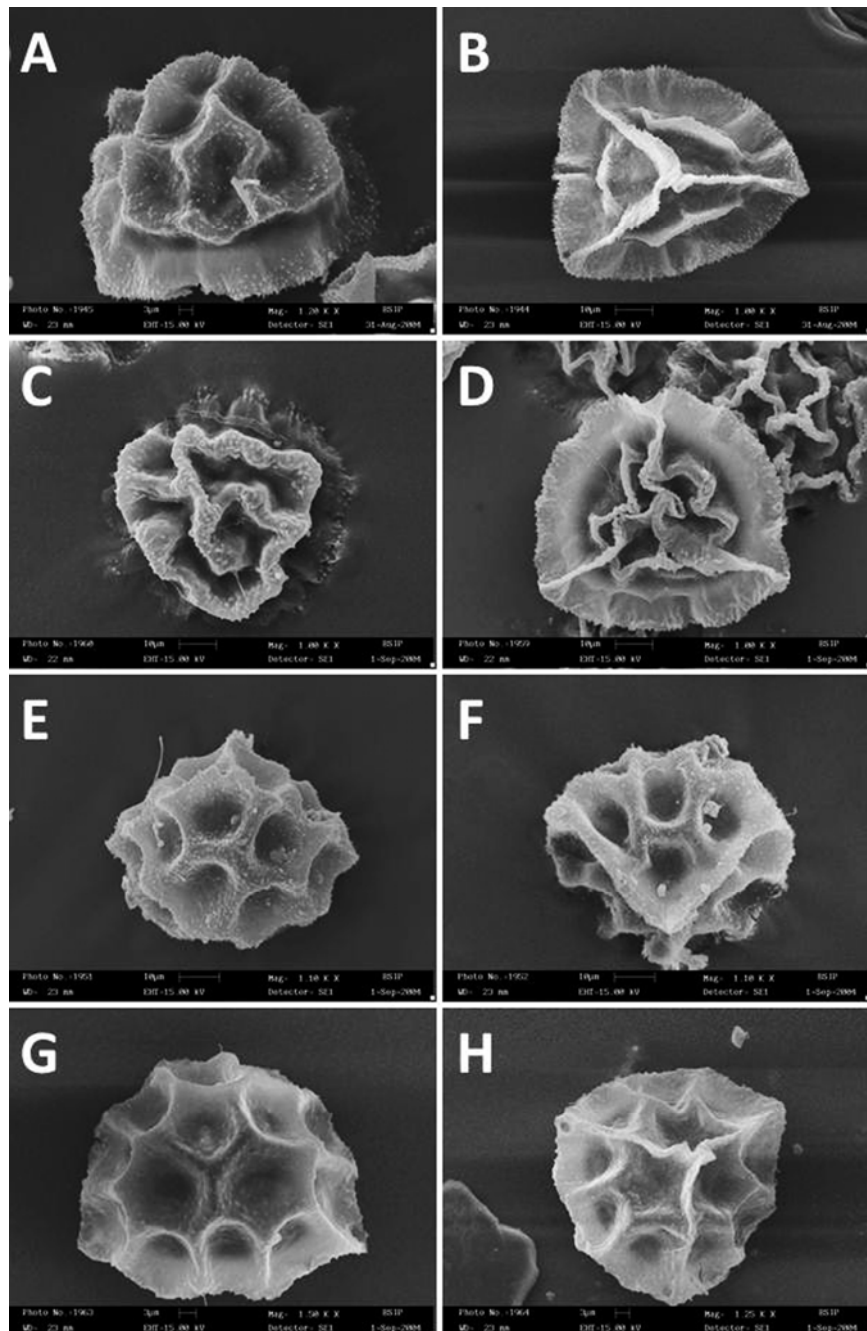
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#### 6. Literature

- ALAM, A & SRIVASTAVA, S. C. (2009). Current Status of Genus *Plagiochasma* in Nilgiri and Palni hills with SEM details of Spores. *Indian Journal of Forestry*, 32(4): 623-634.
- ALAM, A & SRIVASTAVA, S. C. (2012). Hepaticae of Nilgiri hills, Western Ghats India (India), Vol. I: Terrestrial Diversity. LAP-Lambert Academic Publishers, Germany. pp. 1-374.
- IRELAND, R. R. (1992). Scanning electron microscopy of spores of *Herzogiella*, *Lindbergia* **16**: 169-179.
- LONG, D. G. (2006). Revision of the genus *Asterella* P. Beauv. in Eurasia. *Bryophytorum Bibliotheca* 63. J. Cramer, Berlin, Stuttgart, pp. 1-299.
- MULLER, K (1954). Die Lebermoose Europas. In: Rabenhorst's *Krytog. Fl. Deutschl.* **6**: 481-640.
- TAYLOR, J., PEGGIE J. H., & BIGELOW, C (1974). Scanning Electron Microscopy of Liverworts spores and elaters, *The Bryologist* **77**: 281-327.



**Plate- 1. Figs.:** A-F. A. *Asterella khasyana* (distal view), B. *Asterella khasyana* (proximal view). C. *Targionia lorbeeriana* (distal view), D. *Targionia lorbeeriana* (proximal view), E. *Targionia hypophylla* (distal view), F. *Targionia hypophylla* (proximal view).



**Plate- 2. Figs.: A-H.** A. *Plagiochasma cordatum* (distal view), B. *Plagiochasma cordatum* (proximal view). C. *Plagiochasma rupestre* (distal view), D. *Plagiochasma rupestre* (proximal view). E. *Plagiochasma udarii* (distal view), F. *Plagiochasma udarii* (proximal view), G. *Plagiochasma pterospermum* (distal view), H. *Plagiochasma pterospermum* (proximal view).