

SEM STUDIES ON THE SPERMODERM PATTERNS OF SOME SPECIES OF *NICOTIANA* L.

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Abstract

Scanning electron microscopic studies were made on the patterns of spermoderm surface of eight species of *Nicotiana* L. (Solanaceae) seeds, viz., *Nicotiana alata* Link & Otto, *N. amplexicaulis* N. T. Burbidge, *N. benthamiana* Domin, *N. debneyi* Domin, *N. glauca* R. Grah., *N. glutinosa* L., *N. gossei* Domin and *N. hesperis* N. T. Burbidge. Under SEM they show reticulate pattern but vary from each other in minute details which proved useful in their identification.

Introduction

A considerable amount of work on the spermoderm structure of some solanaceous seeds with the help of light microscope (Corner, 1976) and SEM (Heywood, 1969; Mayer & Poljakoff-Mayber 1975; Edmonds 1983; Farooqui & Bahadur, 1985; Gunn & Gaffney, 1974 and Srivastava & Bagchi, 1986) were made to mark certain characteristic features of diagnostic value.

Seeds of *Nicotiana* species are minute and provide little morphological variation and are difficult to identify them as such or even under Light microscope (LM). The SEM study made by Farooqui and Bahadur (1985) on the spermoderm microsculpturing of some *Nicotiana* spp. has little relevance to taxonomic value since the grouping of various seeds into thick and thin radial walls are not based on actual measurements and thus, give no idea of its thickness.

In the present SEM study spermoderm structure of eight exotic species of *Nicotiana* were examined eg. *N. alata* Link & Otto, *N. amplexicaulis* N. T. Burbidge, *N. benthamiana* Domin, *N. debneyi* Domin, *N. glauca* R. Grah., *N. glutinosa* L., *N. gossei* Domin and *N. hesperis* N. T. Burbidge.

Material and Methods

Fully mature and dried authentic seed samples of all the above mentioned species of *Nicotiana* were obtained from the Central Tobacco Research Institute, Rajahmundry, Andhra Pradesh. Seeds were washed with pure acetone, dried and subsequently mounted on stubs with silver paint and coated with a thin layer of gold for 3 minutes at 10 mA and 1 kV in a Edwards coating unit. To ensure uniformity, the flat middle wide area of at least five seeds in each case were scanned using Phillips 420 Transmission/Scanning electron microscope at 20 kV.

Observations

Seeds of *Nicotiana* species are variously shaped and are usually flat, round, oval, triangular or Kidney-shaped. They are minute, the average seed size of the studied seeds are

$0.707 \pm 0.118 \times 0.482 \pm 0.097$ mm. Seeds are usually covered with dried mucilage which appear like fibrils, precipitate or crust under SEM. All *Nicotiana* seeds studied show reticulate pattern with sinuate radial walls under light microscope and scanning electron microscope. However, under SEM minute variations in the spermoderm ornamentation become more clear, which are of immense importance for seed identification as well as in taxonomic and phylogenetic evaluation. The diagnostic characters of eight species of *Nicotiana* as observed under LM and SEM are :

N. alata (Pl. 1, figs. 1, 2)—Seeds are oval or kidney-shaped and $0.865 \pm 0.092 \times 0.67 \pm 0.57$ mm in size. The radial walls of the reticulum are 14-17 μm thick and wavy. Surface of the cavity is granular. There is heavy deposition of mucilage in the form of fibrillar structures (shown by arrow in Pl. 1, fig. 2) on the spermoderm surface.

N. amplexicaulis (Pl. 1, figs. 3, 4)—Seeds are mostly kidney-shaped and $0.716 \pm 0.067 \times 0.466 \pm 0.038$ mm in size. The radial walls of the reticulum are 26-29 μm thick and wavy. The cavity as well as walls have heavy but uneven deposition of precipitate like mucilage (shown by arrow in Pl. 1, fig. 4) without fibrillar structures.

N. benthamiana (Pl. 1, figs. 5, 6)—Seeds are kidney-shaped and measure $0.516 \pm 0.04 \times 0.379 \pm 0.036$ mm in size. The radial walls are wavy and 21-25 μm thick. Joints of the walls have triangular pits (shown by arrowheads in Pl. 1, fig. 6) and middle lamellar region is grooved (shown by arrow in Pl. 1, fig. 6). Unlike *N. amplexicaulis* the radial walls are mostly smooth. Precipitate like mucilage depositions are mostly confined to the cavity area, the surface of which has small irregular tubercles.

N. debneyi (Pl. 2, figs. 7, 8)—Seeds are oval to triangular, $0.707 \pm 0.055 \times 0.483 \pm 0.062$ mm in size. The radial walls of the reticulum are relatively less wavy and 29-33 μm thick. Joints of the radial walls possess protuberance (shown by arrow in Pl. 2, fig. 8). Cavity surface possesses mucilage in the form of crust.

N. glauca (Pl. 2, figs. 9, 10)—Seeds are mostly triangular to kidney-shaped, measuring $0.649 \pm 0.045 \times 0.42 \pm 0.046$ mm in size. Reticula of the spermoderm are mostly elongated, the radial walls are 14-17 μm thick and smooth. Middle lamella is grooved (shown by arrow in Pl. 2, fig. 10) and joints of the walls have triangular pits (shown by arrow head in Pl. 2, fig. 10). Cavity surface is granular and occasionally bears small tubercles. Mucilage on the spermoderm is scanty and as in *N. alata* it is present in the form of fibrillar and globular structures.

N. glutinosa (Pl. 2, figs. 11, 12)—Seeds are usually triangular to rectangular in shape and are $0.67 \pm 0.06 \times 0.408 \pm 0.029$ mm in size. The walls of the reticulum are 17-21 μm thick and wavy. Joints of the walls may sometimes possess triangular pits (shown by arrow in Pl. 2, fig. 12). Cavity surface as observed in *N. amplexicaulis*, is granular and coated with mucilage in the form of precipitate.

N. gossei (Pl. 3, figs. 13, 14)—Seeds are mostly kidney-shaped and measure $0.878 \pm 0.086 \times 0.578 \pm 0.073$ mm in size. The radial walls of the reticulum are less wavy, 25-29 μm thick and ridged at joints (shown by arrow in Pl. 3, fig. 14). The spermoderm surface is covered by a heavy deposition of crust-like mucilage.

N. hesperis (Pl. 3, figs. 15, 16)—Seeds are kidney shaped, $0.657 \pm 0.043 \times 0.449 \pm 0.054$ mm in size. Reticulate radial walls of the spermoderm are 19-22 μm thick and less wavy. Wall surface and the cavity surface both have tubercles which are irregular in shape (shown by arrow in Pl. 3, fig. 16). The surface also has coating of mucilage in the form of precipitate.

Discussion

Seeds of *Nicotiana* species, under study, are minute and exhibit minor variations making their identification difficult. However, SEM studies proved useful in elaborating details of the seed surface. The seeds range in size between $0.516 \pm 0.04 \times 0.379 \pm 0.036$ mm (eg. *N. benthamiana*) and $0.878 \pm 0.086 \times 0.578 \pm 0.073$ mm (eg. *N. gossei*), whereas rest of the species vary in between these ranges. The seeds are usually flat, globular, oval or kidney-shaped, finely pitted and yellowish brown to brownish-black in colour. The hilum is located at the extreme anterior end on one of the lateral sides of the seed.

Variations in the spermoderm ornamentation of various seeds become more distinct when observed under SEM. All the seeds of *Nicotiana* species studied show reticulate pattern of their radial walls and the surface has a mucilage coating. Thickening of the radial wall takes place during the development of the seed. The outer epidermal cells of the testa elongate radially and gradually thickens from inside, while the distal walls of the epidermis remains thin (Corner, 1976). Maturation of seed is followed by dehydration and during this period distal part of the cell wall disintegrates and become mucilaginous. The mucilage and the cuticle collapse on the surface of the epidermal radial walls as a covering and form a particular type of ornamentation on the seed surface.

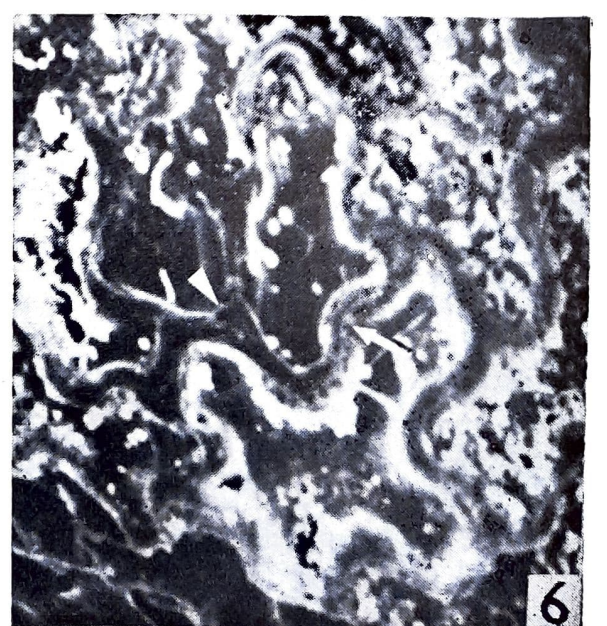
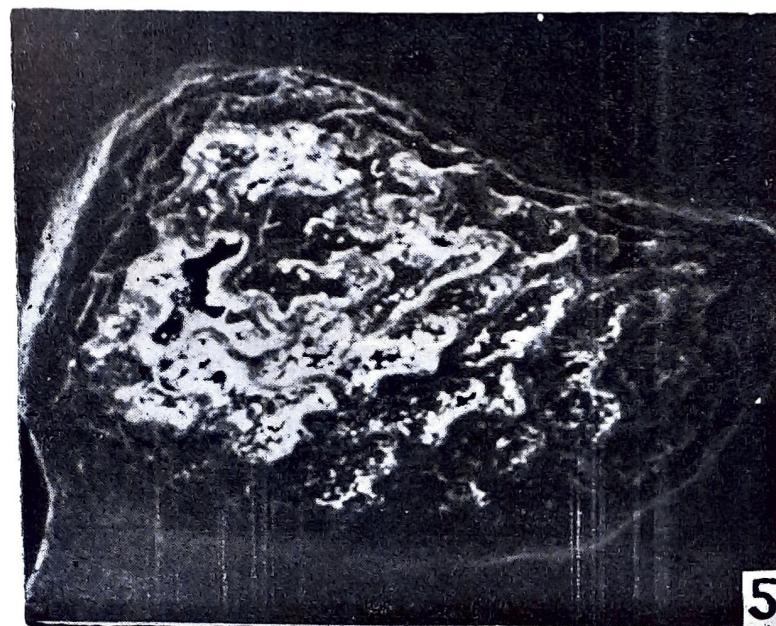
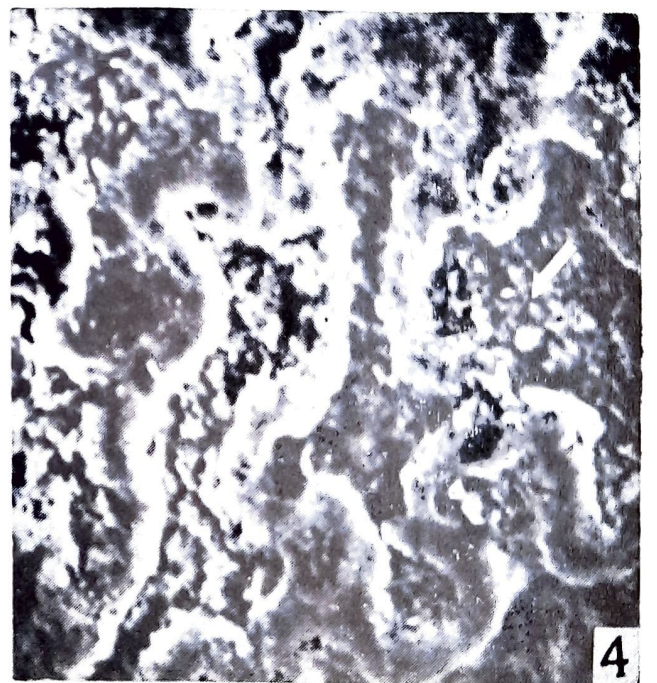
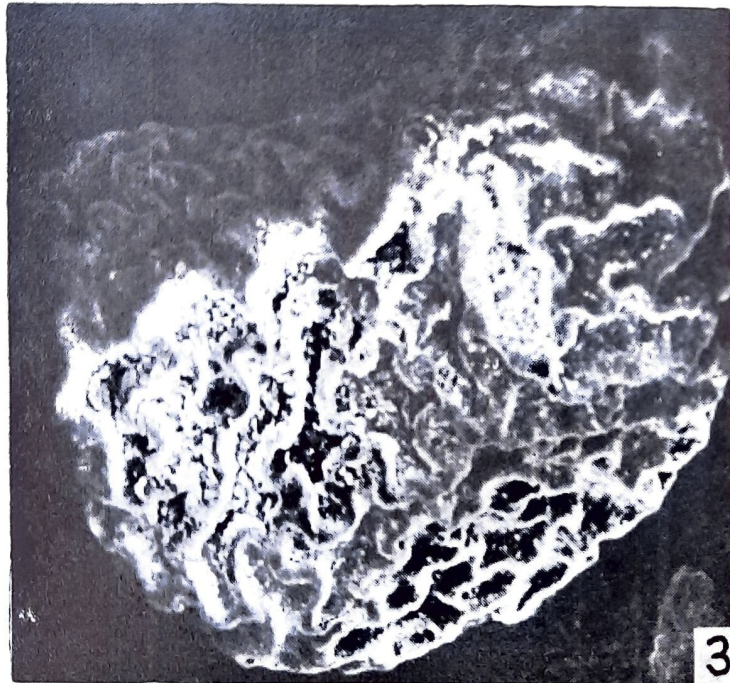
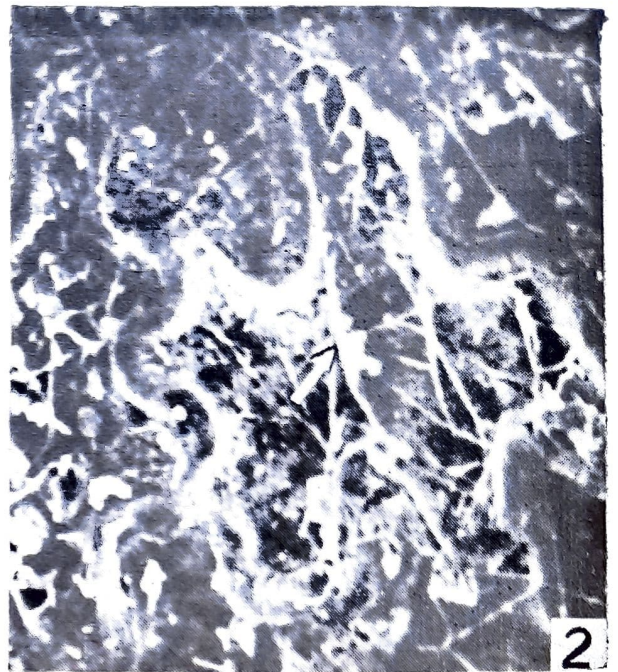
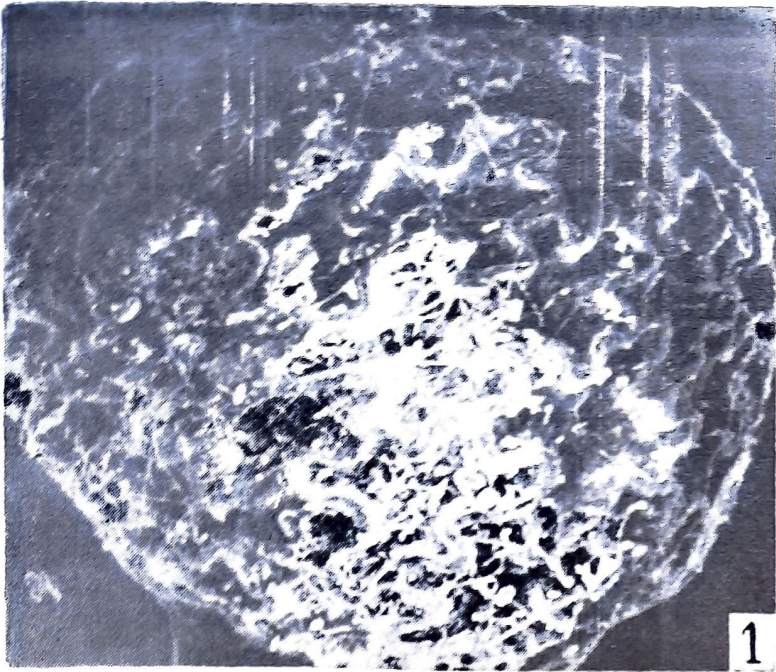
Farooqi and Baġadui (1985), while studying the spermoderm structure of *N. plumbaginifolia*, *N. triglyphylla*, *N. alata*, *N. langsdorhi*, *N. clevelandii* and *N. nudicaulis*, categorised them in two main groups consisting of thin and thick radial walls but without any data in terms of measurement. Spermoderm of a seed may show minor variations in thickness and sinuation of radial walls of a reticulum. On an average, thinnest (14-17 μm) and thickest (29-33 μm) radial walls were recorded in *N. alata*, *N. glauca* and *N. debneyi* respectively, while radial walls of medium thickness (17-25 μm) were found in *N. benthamiana*, *N. glutinosa* and *N. hesperis*. Some of the features such as formation of triangular, pit-like structure at a meeting point of three radial walls (eg. *N. benthamiana*, *N. glauca*, *N. glutinosa*) and grooved middle lamella (eg. *N. benthamiana*, *N. glauca*) have taxonomic significance in their identification. Further, occurrence of mucilage in various form on seeds eg. fibre like (*N. alata*, *N. glauca*), precipitate like (*N. amplexicaulis*, *N. benthamiana*, *N. glutinosa*, *N. hesperis*) and crust like (*N. debneyi*, *N. gossei*) are also complementary to other characters of diagnostic value.

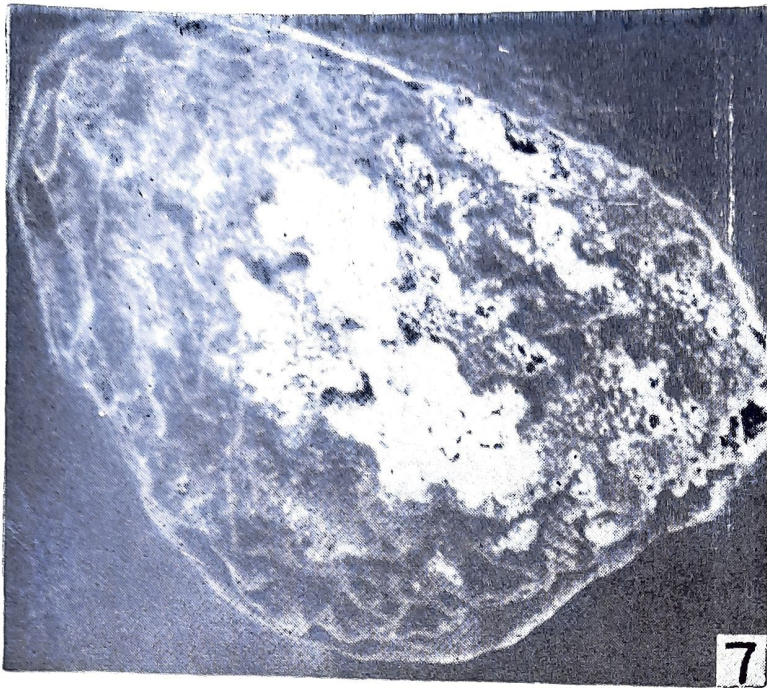
On the basis of characters observed by SEM, the studied seeds of *Nicotiana* species can be categorised for identification as follows :

Key-1

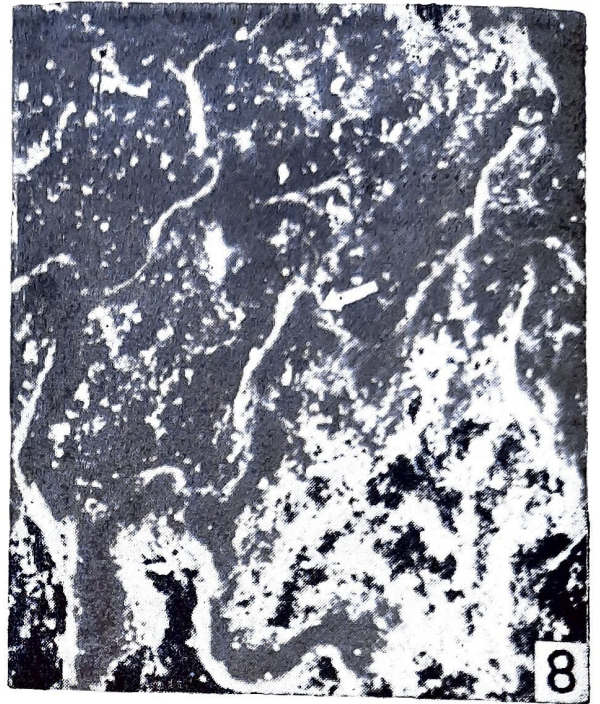
Seeds $0.707 \pm 0.118 \times 0.482 \pm .079$ mm in size, having mucilage coating, spermoderm with reticulate pattern of radial walls.

- | | | |
|--------------------------------------------------|----|-----------------------|
| | .. | <i>Nicotiana</i> |
| 1. Radial walls less than 25 μm thick | | |
| 2. Radial walls less than 17 μm thick | | |
| 3. Middle lamella grooved | .. | <i>N. glauca</i> |
| 3. Middle lamella not grooved | .. | <i>N. alata</i> |
| 2. Radial Walls more than 17 μm thick | | |
| 4. Middle lamella grooved | .. | <i>N. benthamiana</i> |
| 4. Middle lamella not grooved | | |
| 5. Wall smooth | .. | <i>N. glutinosa</i> |
| 5. Wall rough | .. | <i>N. hesperis</i> |

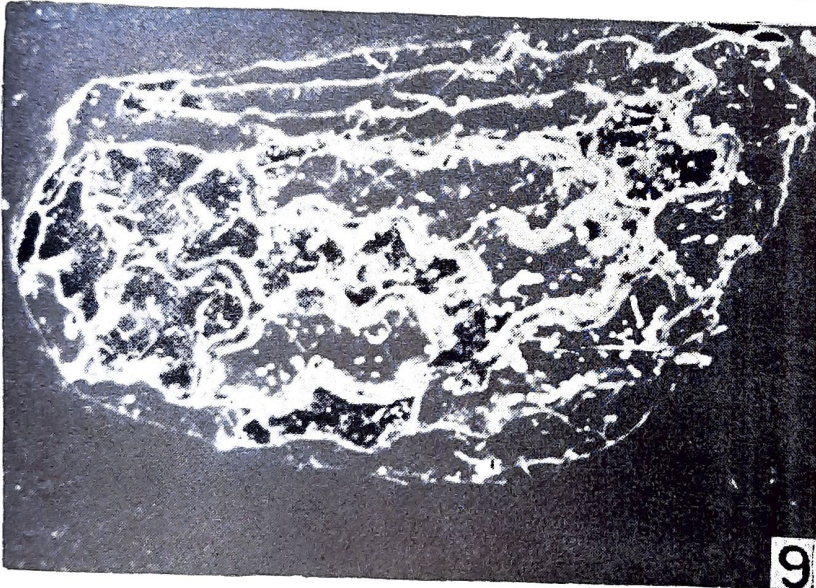




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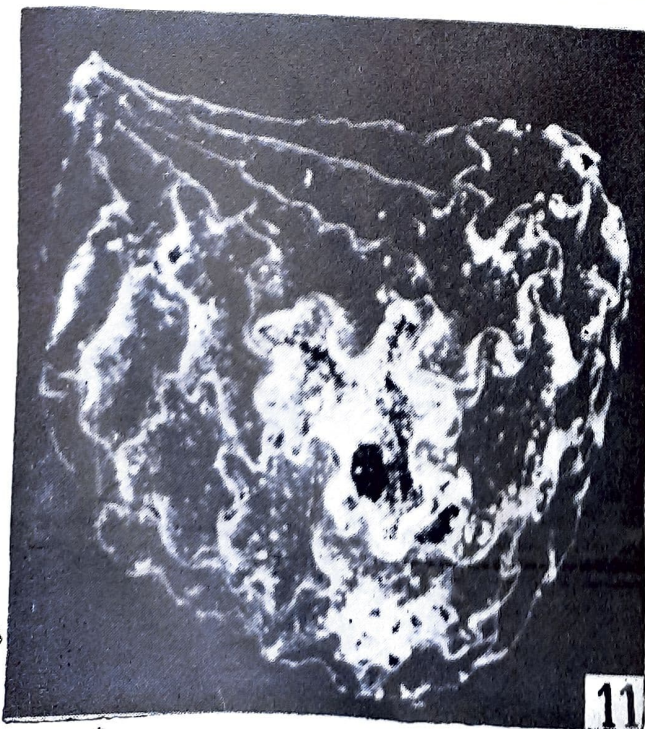
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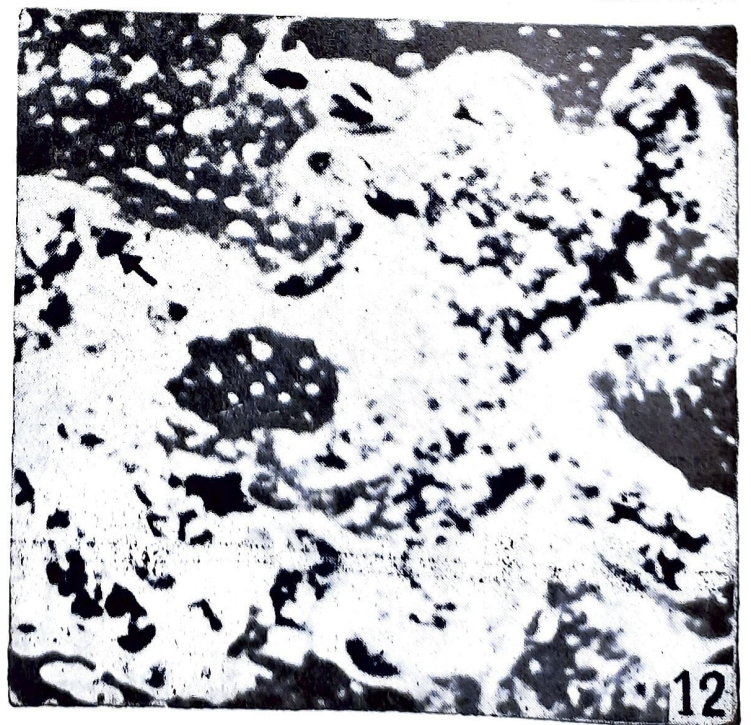
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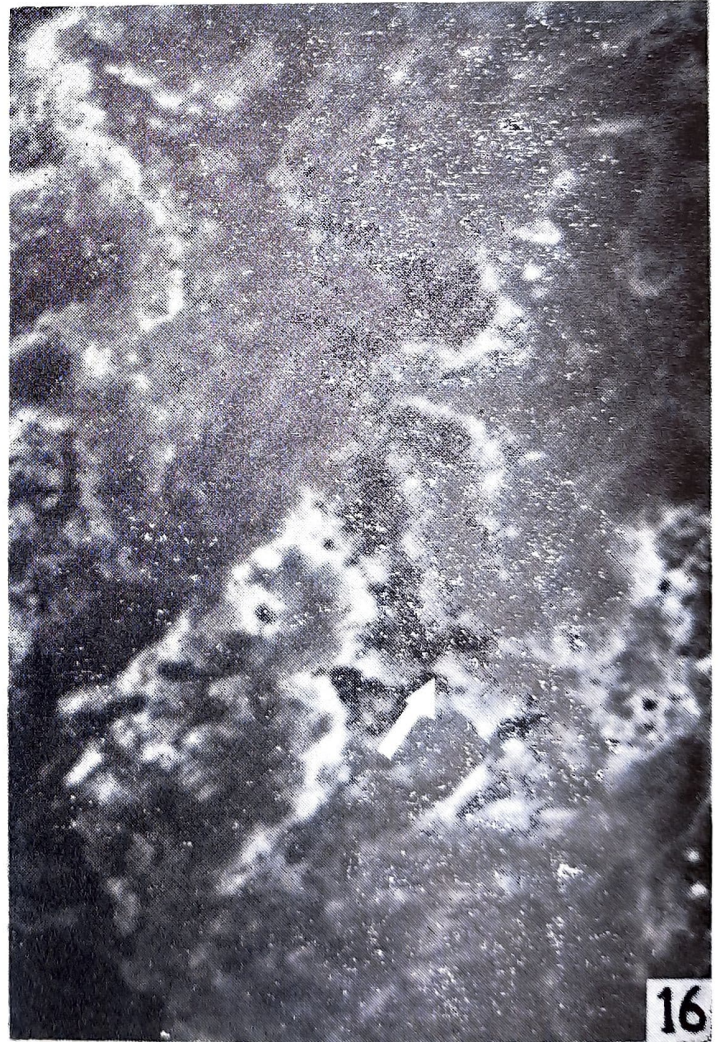
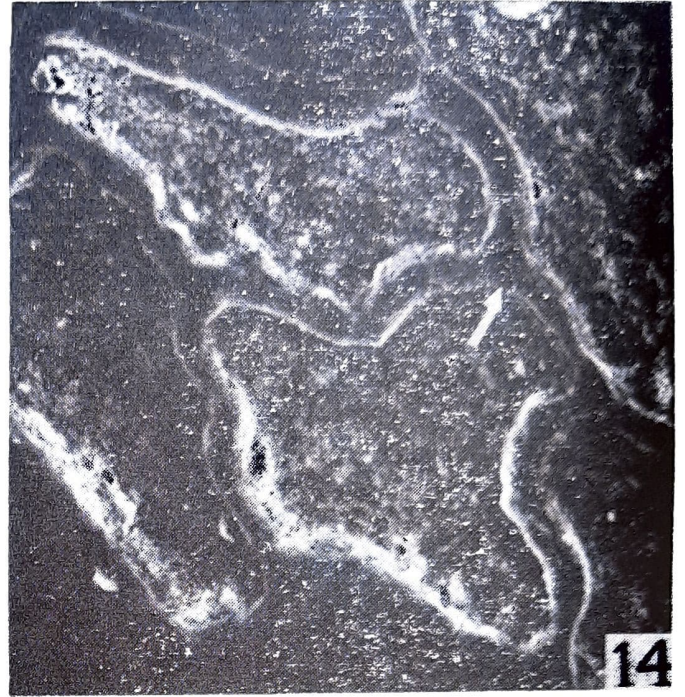
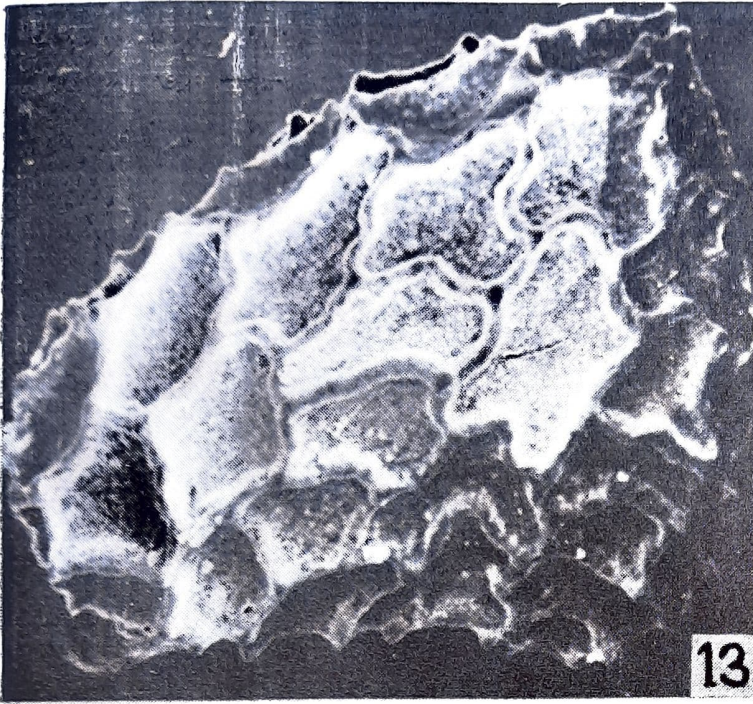
10



11



12



- | | |
|----------------------------------------------------|----------------------------|
| 1. Radial walls more than 25 μm thick | |
| 6. Joints of walls lack protuberance, walls rough | .. <i>N. amplexicaulis</i> |
| 6. Joints of walls have protuberance, walls smooth | |
| 7. Wall distinct, less wavy | .. <i>N. gossei</i> |
| 7. Wall less distinct, wavy | .. <i>N. debneyi</i> |

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Explanation of Plates

Plate 1

Figures 1-6 SEM microphotographs showing spermoderm structure of *Nicotiana* species.

- 1, 2. *N. alata*, arrow indicates fib:illar mucilage deposition in fig. 2 (1 \times 140, 2 \times 350).
- 3, 4. *N. amplexicaulis*, arrow indicates precipitate type mucilage deposition in fig. 4 (3 \times 200, 4 \times 450).
- 5, 6. *N. benthamiana*, arrow indicates grooved middle lamella and arrow head indicates pit in fig. 6 (5 \times 200, 6 \times 450).

Plate 2

Figures 7-12. SEM microphotographs showing spermoderm structure of *Nicotiana* species.

- 7, 8. *N. debneyi*, arrow indicates the protuberance in joint of walls in fig. 8 (7 \times 200, 8 \times 400).
- 9, 10. *N. glauca*, arrow indicates grooved middle lamella and arrow head indicates pit in fig. 10 (9 \times 200, 10 \times 600).
- 11, 12. *N. glutinosa* arrow indicates pit in fig. 12 (11 \times 200, 12 \times 540).

Plate 3

Figures 13-16. SEM microphotographs showing spermoderm structure of *Nicotiana* species.

- 13, 14. *N. gossei*, arrow indicates the protuberance in joint of walls in fig. 14 (13 \times 160, 14 \times 330).
- 15, 16. *N. hesperis*, arrow indicates the rough wall in figs. 15 and 16 (15 \times 250, 16 \times 525).