
LM and SEM pollen studies in Indian Alangiaceae

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The pollen grains of Indian *Alangium* Lam. show interesting morphological features. In the present paper, the pollen of four species, viz., *Alangium alpinum* (Clarke) Smith, *A. barbatum* (R. Br.) Baill. ex Kuntze, *A. chinense* (Lour.) Harm. and *A. salvifolium* (L.) Wangerin have been studied under light and scanning electron microscopes. *Alangium alpinum* distributed in the eastern Himalaya, has 3-4 colpi pollen with characteristic striate sculptured exine. Whereas, *A. chinense* is distributed throughout the Himalaya as well as the plains has rugulate-reticulate sculptured exine with aperture similar to *A. alpinum*. *A. barbatum* is sporadic in Assam and has 3-colporate pollen with faint striate sculpturing. *A. salvifolium* happens to be the most widely distributed species extending from Himalayan foothills to extreme south of the Indian Peninsula as well as Andaman Island and has 3-6 porate-porate-brevicolporate apertures. Pollen grains of *A. salvifolium* subsp. *sundanum* (Miq.) Bloembergen show gemmate-granulate sculpturing whereas, in *A. salvifolium* subsp. *salvifolium* (L.) Wangerin the sculpturing is rugulate-verrucate-gemmate-granulate. SEM studies reveal secondary ornamentation in *A. alpinum*, *A. barbatum* and *A. salvifolium* and interesting variations in the orientation of muri in case of *A. chinense*.

Key-words—Palynology, Morphology, *Alangium*, Alangiaceae.

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सारांश

भारतीय एलेन्जिएसी कुल के परागकणों का प्रकाश एवं क्रमवीक्षण इलेक्ट्रान सूक्ष्मदर्शी द्वारा अध्ययन

छाया शर्मा एवं आशा गुप्ता

भारतीय एलेन्जियम लेम. के परागकण विशेष आकारिकीय लक्षण प्रदर्शित करते हैं। इस शोध-पत्र में *ए. अल्पाइनम*, *ए. बार्बेटम*, *ए. चाइनेन्से* एवं *ए. साल्वीफोलियम* नामक चार जातियों के परागकणों का प्रकाश एवं क्रमवीक्षण इलेक्ट्रान सूक्ष्मदर्शी द्वारा अध्ययन किया गया है। *ए. अल्पाइनम* पूर्वी हिमालय में वितरित है तथा इसमें 3-4 कोल्पोरेट परागकण विद्यमान हैं। जबकि *ए. चाइनेन्से* में बाह्यचोल रुगुलेट-जालिकावत संरचना है तथा यह मैदानी भागों और सम्पूर्ण हिमालय में वितरित है। *ए. बार्बेटम* असम में पाया जाता है तथा इसमें 3-कोल्पोरेट परागकण विद्यमान हैं। *ए. साल्वीफोलियम* हिमालय के गिरि-पादों से सुदूर तक बहुतायत में पाया जाता है तथा इसमें 3-6 पोरेट-पारोरेट-ब्रेविकॉल्पोरेट छिद्र विद्यमान हैं।

ALANGIACEAE is a monotypic dicot family with its genus *Alangium* comprising 17 species (Matthew, 1988) and has a wide distribution confined to the Old World namely India, Sri Lanka, Nepal, Bhutan, Myanmar, eastern China, Vietnam, Japan, Malaya, Java, Sumatra, Borneo, Philippines, New Guinea, Eastern Australia, Madagascar and some regions of tropical Africa, etc. From the phytogeographic point of view, its present and past range of distribution is quite interesting—right from Late Palaeocene. The fossil

pollen attributable to *Alangium* besides being frequently reported from the Indian subcontinent have also been recorded from Africa, Russia, Japan, Malesia, Europe, Pacific Islands, etc. and even from North America, when the genus is totally unrepresented today throughout the New World (Traverse, 1955; Cookson, 1957; Krutzsch, 1962; Muller, 1981; Morley, 1982).

From the Indian subcontinent, pollen of *Alangium* have mainly been reported from various Ter-

tiary deposits, the earliest fossil pollen record is from Late-Palaeocene, Assam (Biswas, 1962) and is seen well documented in Eocene, Oligocene, Miocene and Pliocene (Phadtare & Thakur, 1990; Sharma & Gupta, 1991).

In India, *Alangium* is distributed almost throughout the subcontinent including Himalaya, represented by five species, viz., *Alangium alpinum*, *A. barbatum*, *A. chinense*, *A. salvifolium* and *A. javanicum*. The last one occurs in Andamans. *A. alpinum* is confined to eastern Himalaya and also occurs in East Nepal, Myanmar and China. *A. barbatum* is restricted to Assam. *A. chinense* extending to Bhutan also occurs in Myanmar and Vietnam and besides China it is widely distributed throughout Himalaya as well as the Indian plains namely Assam, West Bengal and Andhra Pradesh. *A. salvifolium* has a much wider distribution and is found in most of the drier regions of the country. It is distributed from the foot-hills of Himalaya to extreme south of the Indian subcontinent.

Many workers have studied the pollen grains of Alangiaceae like Erdtman (1952, 1969), Chao (1954), Yeramyan (1958, 1967), Ikuse (1956), Nair (1965) Straka and Simon (1967), Reitsma (1970), Van Campo (1978), Kedves (1979), Muller (1981), Gupta and Sharma (1986), etc. Both Nair (1965) as well as Gupta and Sharma (1986) in their contributions on the pollen grains of western Himalayan flora have provided the pollen morphology of one species, i.e., *A. chinense*. Nair (1965) describes the pollen as "3-zonocolporate, suboblate (63 x 77 µm). Endocolpium lalongate (7 x 9.8 µm); exine 4.2 µm thick, punctate-reticulate, lumina being irregularly shaped (fossulate)". Gupta and Sharma (1986) described the pollen as "tricolporate, suboblate (55 x 58 µm). Colpi long, broad, with thickened and irregular margin. Ora circular-subcircular (7 x 8 µm). Exine 4.5 µm, sexine considerably thicker than nexine, pattern obscure. Tegillate with uneven contours". However, Reitsma (1970) in his monographic studies on world Alangiaceae has broadly grouped the species under different pollen types. In case of *A. chinense* complex, he has recognised four different types and even subtypes within a type to establish distributional segregation.

Since, the detailed pollen morphology of extant *Alangium* taxa has not so far been attempted, the present LM and SEM pollen studies of four Indian species have been carried out which revealed considerable inter and intraspecific differences in their pollen. Among five Indian species, four occur in the mainland and one in the Andamans. The LM and SEM pollen studies of only mainland species, viz., *A. alpinum*, *A. barbatum*, *A. chinense* and *A. salvifolium* have been carried out and discussed in the present paper.

MATERIAL AND METHODS

Polleniferous material for the present studies was taken from the Herbarium sheets of Birbal Sahni Institute of Palaeobotany, Lucknow and Botanical Survey of India, Gangtok and Shillong. The Herbarium sheet particulars are as under :

1. *Alangium alpinum*
Eastern Himalaya : Sikkim, Indo-Japanese Collection, BSIP Reg. no. 5324.
2. *A. barbatum*
Shillong Herbarium.
3. *A. chinense*
(i). Eastern Himalaya : Sikkim, Indo-Japanese Collection, BSIP Reg. no. 9313.
(ii). Western Himalaya : Garhwal, Stewart's Collection, BSIP Reg. no. 1436; Kumaon, BSIP Reg. no. 6705; Himachal Pradesh, N.C. Nair's Collection, BSIP Reg. no. 9846.
4. *A. salvifolium* subsp. *sundanum*
Western Himalaya : Kumaon, BSIP Reg. no. 20355.
5. *A. salvifolium* subsp. *salvifolium*
(i). Uttar Pradesh, BSIP Reg. no. 21018.
(ii). West Bengal, Mrs Robert's Collection, BSIP Reg. no. 9846.

First of all, the polleniferous material was soaked in glacial acetic acid and then acetolysed by following the standard acetolysis technique (Erdtman, 1943, 1960; Faegri & Iversen, 1975). These acetolysed pollen were used for both LM as well as SEM studies. Light-microscopic observations were made using Leitz Diaplan microscope and the photography by Lietz Automatic photo equipment by 10 x and 100 x objectives.

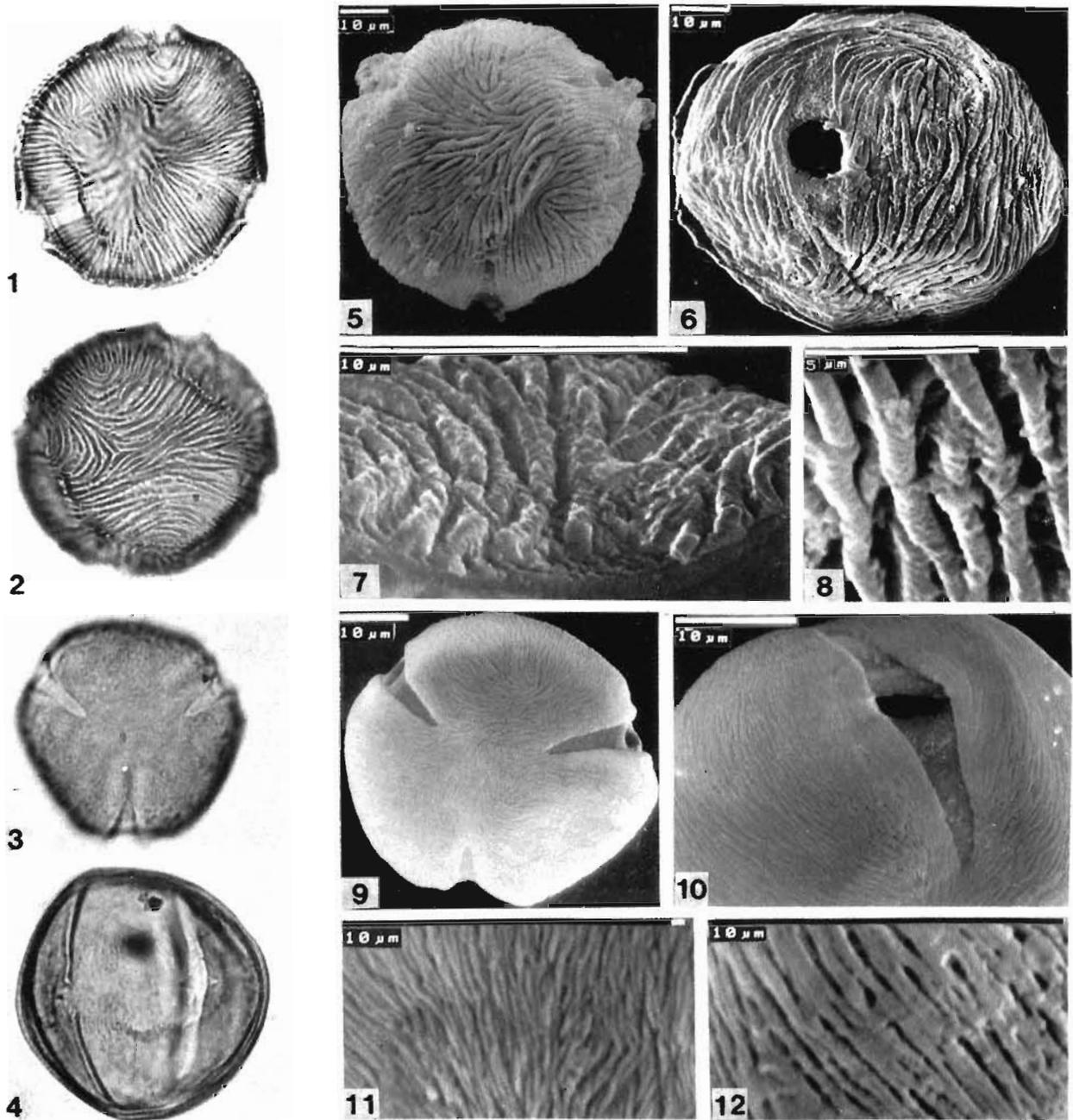


PLATE 1

- Alangium alpinum*
1. 3-Colporate pollen x 500.
 2. 4-Colporate pollen x 500.
 5. 3-Colporate pollen.
 6. Pollen in equatorial view showing circular os.
 - 7, 8. Enlarged part showing secondary ornamentation.

- Alangium barbatum*
3. 3-Colporate pollen x 500.
 4. Equatorial view x 500.
 9. 3-Colporate pollen.
 10. Magnified portion showing aperture.
 - 11, 12. Magnified portion showing striate sculpture.

For scanning electron microscopy, the pollen were subjected to sputter-coating by gold-palladium alloy. The photomicrographs are with Phillips Holland-505 microscope.

DESCRIPTION

Pollen grains are 3-4 colpi to 3-6 porate-porate- brevicolporate. Shape ranges from oblate, suboblate, oblate-spheroidal, prolate-spheroidal to subprolate and in size from 30 x 28 to 95 x 102 μm . The surface ornamentation is highly variable and reveals striate, rugulate-reticulate, gemmate-verrucate-granulate patterns. Secondary ornamentation of sculptural elements or lumina may or may not be there showing granular, rugulate to tiny reticulate patterns.

Alangium alpinum (Clarke) Smith

Light microscopic observations (Pl. 1, figs 1,2)

Aperture—3-colporate, occasionally 4-colporate.

Ectoaperture—Colpus 38-45 μm long, 6-13 μm broad, apices rounded or obtuse.

Endoaperture—Lalongate porus, subcircular (9 x 11, 12 x 15 μm), dumb-bell-shaped (15 x 10 x 11 μm , 13 x 6 x 6 μm); circular porus (10-12 μm in diameter); rarely lolongate (10 x 9 μm), broader than ectoaperture, bordered by differentiated margo, situated in the middle, rarely towards the side of ectoaperture, margin irregular.

Shape and size—Suboblate (62 x 71 - 68 x 78 μm), oblate-spheroidal (64 x 66 - 68 x 74 μm).

Exine—2 - 2.5 μm thick, sexine considerably thicker than nexine.

Ornamentation—Striate, striae long, parallel or variously placed, unbranched or branched; lumina as broad as striae, relatively narrower or broader at places; lumina surface show very minute indistinct secondary sculpturing.

SEM observations (Pl. 1, figs 5 - 8)

Secondary sculpturing of lumina distinct, showing uni-multiseriably arranged minute net-pattern. Both striae and colpi surface have tiny projections.

Alangium barbatum (R. Br.) Baill. ex Kuntze

Light microscopic observations (Pl. 1, figs 3, 4)

Aperture—3-colporate.

Ectoaperture—Colpus 48-58 μm long, 1-5 μm broad, apices obtuse or acute.

Endoaperture—Circular porus (5-12 μm in diameter), lalongate porus subcircular (2.5 x 7-5 x 11.5 μm), rarely lolongate (12 x 5 μm), situated in middle, broader than ectoaperture and bordered by thin margo.

Shape and size—Oblate (48 x 68 μm), suboblate (49 x 59 μm), oblate spheroidal (61 x 68 μm), prolate spheroidal (66 x 61 μm), subprolate (56 x 49 μm).

Exine—2-3 μm thick, sexine nearly as thick as nexine to slightly thicker.

Ornamentation—Faintly striate, striae indistinct, irregular and parallel.

SEM observations (Pl. 1, figs. 9-12)

Sculpturing distinct; striae thin, smooth, unbranched or branched, dense and variously placed; lumina thinner than striae, smooth or with uniseriably arranged tiny elements (granule-like) dividing lumina into minute net. Aperture also with similar tiny sculptural elements at colpi surface and dissimilar orientation of striae at colpi margin.

Alangium chinense (Lour.) Harm.

Pollen from different regions of Himalaya have shown considerable variability and can be broadly identified into two pollen types and a transitional type as described below. Pollen Type-1 is from Sikkim, Pollen Type-2 from Himachal Pradesh and Transitional Pollen Type from Kumaon Himalaya :

Pollen Type-1

Light microscopic observations (Pl. 2, fig. 1)

Aperture—3-colporate, occasionally 4-colporate.

Ectoaperture—Colpus 23-28 μm long, 1-1.5 μm broad.

Endoaperture—Lolongate porus (7 x 6 - 9 x 7 μm), much broader than maximum width of ectoaperture, rarely lalongate porus (2 x 14 - 3 x 18 μm).

Shape and size—Oblate (40 x 70 μm), prolate-spheroidal (69 x 68 μm).

Exine—4 μm thick, sexine as thick as nexine.

Ornamentation—Very minutely rugulate.

SEM observations (Pl. 2, figs 5, 6, 8)

Pollen grains have rugulate (reticulate) sculpturing, rugae narrow, variously long, densely placed,

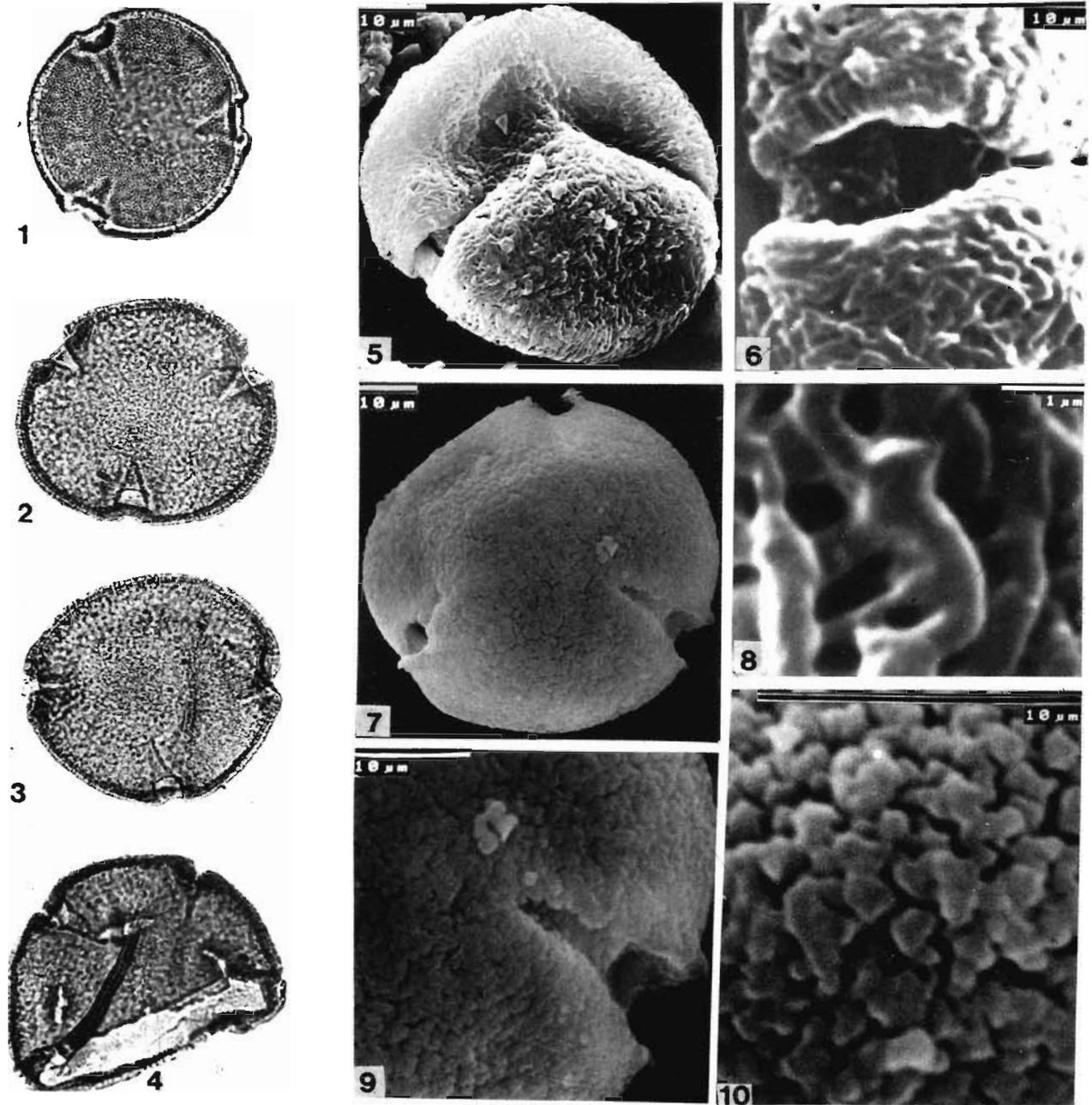


PLATE 2

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| 1. | <i>Alangium chinense</i> , Type-1
3 ² -Colporate pollen x 500. | 2. | 3-Colporate pollen x 500. |
| 5. | 3-Colporate pollen. | 3. | Pollen with transition between 3 and 4 colporate condition x 500. |
| 6. | Enlarged portion through aperture. | 4. | Pollen with more than 4 apertures x 500. |
| 8. | Magnified ornamentation . | 7. | 3-Colporate pollen. |
| | <i>Alangium chinense</i> , Type-2 | 9. | Enlarged portion showing aperture. |
| | | 10. | Magnified ornamentation . |

branched or unbranched, branches often less high than main muri, united variously with neighbouring ones forming reticulations. Colpi border have identical sculpturing with dissimilar muri orientation; rugae along colpi margin placed either transversely or obliquely. Colpi surface provided with tiny verrucate projections.

Pollen Type-2

Light microscopic observations (Pl. 2, figs 2 - 4)

Aperture—3-colporate, occasionally 4-colporate, rarely diorate. Also rarely transition between 3 and 4 colporate and even more than 4-colporate.

Ectoaperture—Colpus 23 - 40 μm long, 3 - 10 μm broad, margin uneven.

Endoaperture—Lalongate porus, subcircular (9 x 10-7 x 14 μm); circular porus (5-11 μm in diameter); occasionally lolongate porus (5 x 4 - 8 x 6 μm).

Shape and size—Oblate (52 x 71- 62 x 86 μm), suboblate (40 x 50 - 64 x 84 μm).

Exine—2 - 3 μm thick, sexine as thick as nexine or slightly thicker.

Ornamentation—Minutely imperfectly rugulate-granulate, rugae appear as linearly adherent granules.

SEM observations (Pl. 2, figs 7, 9, 10)

Pollen grains have imperfect rugulate (-reticulate) sculpturing; rugae broad, stumpy, appearing as fused gemmae and may unite with neighbouring ones to form reticulations. Pollen from Himachal Pradesh specimens show lesser reticulations than from Garhwal. Colpi border bear identical sculpturing, surface is dominated by gemmae.

Transitional Pollen Type

Pollen grains from Kumaon specimens are intermediate between Pollen Type-1 and Pollen Type-2. Sculpturing shows mixture of regular and irregular rugae (with reticulations) and exine 4 μm in thickness. Shape varies from oblate (46 x 66 - 56 x 79 μm), suboblate (59 x 75 - 62 x 71 μm) to oblate-spheroidal (66 x 72.5 μm). Regular muri and thick exine resemble Pollen Type-1 whereas in having irregular muri and suboblate shape with Pollen Type-2.

Differentiation in Pollen Type-1 and Pollen Type-2—Pollen Type-2 differs from Pollen Type-1 by its

imperfect orientation of rugae and thin exine. The former type also shows diorate condition and the aperture transition between 3-colporate and 4-colporate and even sometimes more than 4 apertures.

Alangium salvifolium (L.) Wangerin

Two subspecies, i.e., *A. salvifolium* subsp. *sundanum* and *A. salvifolium* subsp. *salvifolium* show dissimilar pollen morphology and are being described separately.

Alangium salvifolium subsp. *sundanum* (Miq.) Bloembergen

Light microscopic observations (Pl. 3, figs 1, 2)

Aperture—4-occasionally 3, 5, 6 brevicolporate-pororate to porate.

Ectoaperture—Colpus to porus, colpi short, up to 20 μm long, 9 μm broad, apices acute to rounded; porus circular-subcircular.

Endoaperture—Circular porus (6-10 μm in diameter); lalongate porus (6 x 9 - 8 x 12 μm); lolongate (7 x 6 μm), broader than maximum width of ectoaperture.

Shape and size—Suboblate (65 x 79 - 69 x 83 μm), oblate spheroidal (64 x 70 - 74 x 75 μm).

Exine—1 μm thick (without sculptural elements), sexine slightly thicker than nexine.

Ornamentation—Gemmate with or without inter-mixed granules.

SEM observations (Pl. 3, figs 5 - 8)

Sculptural elements gemmate, gemmae unequal in size, densely placed, with only visible upper portion, exposed gemmae may reveal short columellae at the bases, and the underlying surface ornamentation showing granular appearance. Apertural region beset with identical sculptural elements.

Alangium salvifolium subsp. *salvifolium* (L.) Wangerin

Light microscopic observations (Pl. 3, figs 3, 4)

Aperture—4, 5 brevicolporate-pororate-porate, sometimes 6, rarely only 3.

Ectoaperture—Colpi small, up to 20 μm long, 4.5 - 8 μm broad, rounded apices; pori circular-subcircular.

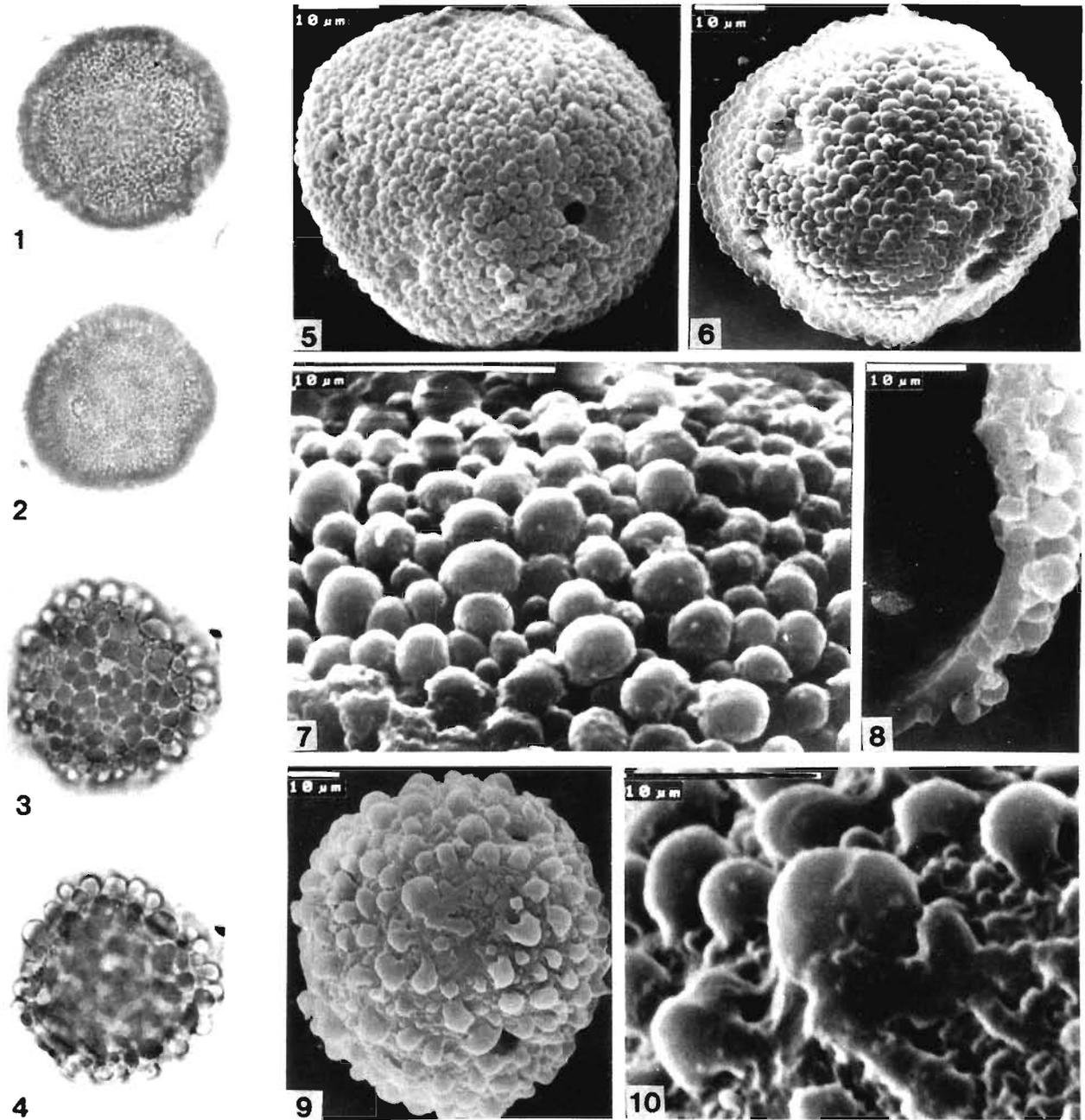


PLATE 3

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| <p><i>Alangium salvifolium</i> subsp. <i>sundanum</i></p> <p>1. 4-porate pollen x 500.</p> <p>2. 5-porate pollen x 500.</p> <p>3, 4. Pollen showing aperture and ornamentation x 500.</p> | <p>8. Sectional view of wall.</p> <p><i>Alangium salvifolium</i> subsp. <i>salvifolium</i>.</p> <p>9. Pollen with gemmate rugulate ornamentation.</p> <p>10. Enlarged ornamentation.</p> |
| <p>5, 6. Pollen showing aperture and gemate ornamentation.</p> <p>7. Magnified ornamentation.</p> | |

Endoaperture—Circular porus (7-11 μm in diameter), lalongate porus (6 x 5 - 11 x 9 μm), occasionally lalongate porus (6 x 7 - 7 x 9 μm), broader than maximum width of ectoaperture, rarely narrower, bordered by thickened costae.

Shape and size—Suboblate (66 x 77 μm), oblate-spheroidal (79 x 82 μm), subprolate (81 x 65 - 84 x 70 μm).

Exine—1.5 - 2 μm thick, sexine as thick as nexine.

Ornamentation—Rugulate-gemmate-verrucate-granulate, variously intermixed or showing predominance of rugae or gemmae-verrucae; up to 8 μm high and 10 μm broad, apertural margin often with larger, densely placed elements.

SEM observations (Pl. 3, figs 9, 10)

Sculptural elements distant, free or occasionally two or more united, rugae appear as thread like extension. Basal portion of sculptural elements show highly variable columellae often united with those of adjoining elements to form network pattern at places, underlying surface granular in appearance, secondary ornamentation at the surface of primary elements show loosely scattered tiny projections.

DISCUSSION

Pollen of extant Indian taxa of Alangiaceae show specific morphological differences of taxonomic significance which are helpful in the delimitation of various species of *Alangium*. They show dissimilarity in the sculpturing pattern and aperture. Pollen of *A. alpinum* has distinctly prominent striate sculpturing with 3-4 colporate apertures; the striae under SEM further reveal very minute secondary sculpturing, lumina often as broad as striae, striae with uni-multiserially arranged net work. Pollen of *A. barbatum* show 3-colporate aperture with fine and faint striate sculpturing, striae smooth, lumina often narrower than striae with uniserially arranged net pattern. *A. chinense* pollen is also 3-4 colporate but has extremely reduced rugulate (-reticulate) sculpturing, indistinct under LM but distinct under SEM. Unlike *A. alpinum*, *A. barbatum* and *A. chinense*, the pollen of *A. salvifolium* has (3-) 4-5 (-6) porate, pororate-brevicolporate apertures. *A. salvifolium* subsp. *sun-*

danum pollen has granulate sculpturing with mostly 4 apertures, whereas *A. salvifolium* subsp. *salvifolium* shows rugulate-gemmate-verrucate-granulate sculpturing with 4-5 apertures.

The pollen variations in *A. chinense*, a widely distributed species in the Indian subcontinent and adjoining countries, amply demonstrate evolutionary trends probably as a result of eco-adaptive isolation, i.e., the stock in eastern Himalayan vis-a-vis western Himalayan material and the intermediate form seen in the pollen grains from Kumaon specimens. Incidentally, it may be pointed out that Reitsma (1970) does divide *A. chinense* into two sub-types I and II based on the comparative thickness of sexine and nexine under the category Type A. He further states that sub-types I and II are the elements having distribution in the eastern region but our segregation of Indian material studied for the present investigation showing pollen differences in the material from western Himalayan and eastern Himalayan region. Further pollen investigations in *A. chinense* from different regions are yet to be carried out which may lead to determine infra-specific entities within the species.

KEY TO INDIAN ALANGIACEAE

1. Aperture colporate2
1. Aperture porate, pororate, brevicolporate3
2. Sculpturing pattern rugulate — *Alangium chinense*
2. Sculpturing pattern striate4
3. Sculpturing gemmate - *A. salvifolium* subsp. *sundanum*
3. Sculpturing rugulate-gemmate-verrucate-granulate—
— *A. salvifolium* subsp. *salvifolium*
4. Striations coarse, distinct *A. alpinum*
4. Striations fine, faint *A. barbatum*

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