

### 3. COMPARISON OF ULTRASTRUCTURE OF THE CUTICLE IN SOME EXTINCT AND EXTANT TAXA OF GYMNOSPERMS FROM INDIA

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#### Abstract

The ultrastructure of the cuticular membrane in 3 taxa of Mesozoic "pteridosperms" (cf. *Dicroidium gouldii*, *Komlopteris indica*, *Pachypteris indica*) and 4 taxa of living cycads (*Cycas circinalis*, *C. revoluta*, *Zamia fischeri*, *Z. furfuracea*) is reviewed and discussed. It is observed that all the 4 living species show comparable ultrastructure, while the ultrastructure in two families of Mesozoic "pteridosperms", namely, *Corytospermaceae* and *Peltaspermaceae* seems to be different.

**Key words:** Ultrastructure, Cuticular membrane, Gymnosperms, India.

#### Introduction

The plant cuticular membrane (CM) is a thin extra-cellular layer that covers most of the aerial part of the plant body, such as, the leaf, herbaceous stem, flowers, seeds and fruits. It also protects the plant against environmental stress, such as drought, high concentrations of solutes within the tissue and microbial activity. The CM is attached to the outer surface of the epidermis through an intermediate pectinaceous layer (MARTIN and JUNIPER, 1970) and transcribes features of the epidermis. The main structural components of the CM are insoluble lipid polyester cutin (HOLLOWAY, 1982), or insoluble nonhydrolyzable biomacromolecule cutan (TEGELAAR et al., 1991). Investigation of the anatomy of cuticular membrane under LM, SEM and TEM provides valuable data for identification of different taxa. The identification helps in taxonomy and further in correlation of different groups of extinct and extant taxa.

Several studies have been done on the surface morphology of CM under LM and SEM, but comparatively less data is available on the ultrastructure of CM under TEM. The CM of a large number of taxa of extant plants has been investigated at ultrastructural level (MARTIN and JUNIPER, 1970; CUTLER, ALVIN and PRICE, 1982) but not many extinct taxa of gymnosperms have been studied for the ultrastructure of CM. Thus there is a great potential for the study of ultrastructure of CM of extinct and extant taxa of gymnosperms for systematic identification, taxonomic classification and correlation of compression fossils, particularly those of the gymnosperms. A comparative study of ultrastructure of CM of extinct and extant taxa of plants may perhaps lead to identification of "the nearest living relative". This is important because "morphological and systematic similarity is assumed to reflect similarity in climatic tolerance" (MOSBRUGGER, 1999). Study of ultrastructure of fossil CM may also help in taphonomy (for example, in

determining the degree of cuticle preservation), and in understanding the evolution of the cuticles (TAYLOR, 1999).

Much more attention needs to be given to the study of ultrastructure of CM of "pteridosperms", which is a large and heterogeneous group. The cuticle of late Palaeozoic "pteridosperms", including the glossopterids, is generally poorly preserved and less resistant to chemical processing as compared to the CM of Mesozoic "pteridosperms". The latter are well preserved and exhibit structural components identifiable at ultrastructural level (ARCHANGELSKY and TAYLOR, 1986; ARCHANGELSKY, TAYLOR and KURMANN, 1986; TAYLOR, TAYLOR and ARCHANGELSKY, 1989; ARTABE and ARCHANGELSKY, 1992; BARALE and BALDONI, 1993; MAHESHWARI and BAJPAI, 1996; BAJPAI, 1997; VILLAR DE SEONE, 1997).

Information about ultrastructure of the cycadalean cuticle is known from *Encephalartos lehmannii* LEHMANN (VILLAR DE SEONE, 1997), *Stangeria paradoxa* MOORE (ARTABE and ARCHANGELSKY, 1992) and *Cycas circinalis* L. (BAJPAI, 2001).

For the present study some extinct taxa of gymnosperms were chosen to examine ultrastructural features with modern analogs by comparing similar features (for example, thickness of CM, presence or absence of lamellae, nature of CM zones, amorphous, reticulate/fibrillate, etc.). The potential diagenetic influences may also be identified. The taxa investigated belong to the Corystospermaceae (cf. *Dicroidium gouldii*, *Komlopteris indica*, *Pachypteris indica*), and the Cycadaceae (*Cycas circinalis*, *C. revoluta*, *Zamia fischeri*, *Z. furfuracea*).

## Materials and Methods

Leaves of *Cycas circinalis*, *C. revoluta*, *Zamia fischeri* and *Z. furfuracea* were obtained from the herbarium of Birbal Sahni Institute of Palaeobotany, Lucknow. The carbonified crust of cf. *Dicroidium gouldii* was recovered from a Late Triassic grey argillaceous shale exposed in Janar rivulet, about one kilometer SSW of Harai Village in South Rewa Basin, that of *Komlopteris indica* from an Early Cretaceous grey shale in a shallow well in the village Naicolam, Cauvery Basin and of *Pachypteris indica* from an Early Cretaceous grey shale exposed near Sehora-on-Sher, Satpura Basin.

The carbonised pinnules removed from the compression fossils were treated with 40% hydrofluoric acid to digest silica. After thorough washing in triple-distilled water, the pinnules were cut into small pieces suitable for processing. Leaves of extant taxa were not given any chemical treatment at this stage. They were, however, also cut into pieces small enough for further processing. The material of both extinct and extant taxa was processed as per method given by MAHESHWARI and BAJPAI (1996). Final 600-700Å thin sections were cut with a diamond knife and mounted on copper grid and stained with uranyl acetate and lead citrate.

## Results

cf. *Dicroidium gouldii* RETALLACK 1977 (Plate 3.1., figs. 1-3)

The CM configuration shows a three-layer configuration. The outermost polylamellate region is made up of parallel running, alternating, 4-6 electron dense, and 4-7 electron lucent lamellae, thickness of which is not uniform. Some of the lamellae are continuous while others run only for a short while. On the leaf-air interface irregular osmiophilic deposits are present, which are possibly the remnants of the epicuticular waxes left after diagenesis. Inner to polylamellate region is a comparatively thick amorphous region that shows incipient fibrillar components. The innermost zone is the thickest of the three zones and is made up of distinct fibrillae which have a "herring bone" appearance and are oriented mainly parallel to the membrane surface. At regular intervals the innermost zone forms wedge-shaped outgrowths which are taken to represent the cuticular pegs or anticlinal flanges that penetrate in inter-wall spaces between adjacent epidermal cells.

*Komlopteris indica* (FEISTMANTEL) BARBACKA (Plate 3.1., fig. 4)

The CM is covered with remnants of epicuticular wax at the leaf-air interface. The CM, which is not uniformly thick throughout, shows two zones, a narrow electron dense outer zone, and a comparatively much wide electron lucent homogeneous inner zone. At the innermost limit of the sub-cuticular level, where the cuticle comes in contact with the epidermis, the sub-cuticular zone forms cuticular pegs (anticlinal flanges) in which extends a narrow strip of electron dense zone. The CM conforms to Type-6 of HOLLOWAY'S (1982) cuticle types.

*Pachypteris indica* (OLDHAM and MORRIS) BOSE and ROY (Plate 3.1., fig. 5, Plate 3.2., fig. 1)

Randomly arranged osmiophilic bodies are seen at the leaf-air interface of the CM. The CM exhibits two distinct zones, an outer electron dense amorphous zone with homogeneous matrix, and an inner electron lucent, irregularly reticulate-fibrillate zone. The basic framework at the sub-cuticular level comprises very fine and distinctly anastomosing fibrillae. The latter branch frequently and are more numerous near the epidermal cell wall. At places several lipophilic vesicles seem to permeate the sub-cuticular level.

*Cycas circinalis* L. (Plate 3.2., figs. 2,3)

At the leaf-air interface a heavy deposit of epicuticular wax is seen with the thin superficial deposit of osmiophilic bodies. The CM of mature leaf exhibits a three-layered configuration. The outer well-developed polylamellate zone is made up of compactly arranged, parallel running, 8-9 electron dense lamellae alternating with 7-8 electron lucent lamellae. The lamellae are mostly uniform in width, but some lamellae run continuous while other runs only for a short distance. The polylamellate zone is followed by an amorphous homogeneous matrix that seem to form the major part of the CM. Inner to this zone is another zone that has matrix of the same density but with fine reticulations. The orientation of the reticulum is mostly parallel to the general surface of CM. Irregular masses of lipophilic secretions are seen to permeate at the sub-cuticular surface contributing to the thickness of the CM. These secretions impart an irregular dendroid appearance to the subcuticular layer.

*Cycas revoluta* THUNB. (Plate 3.2., fig. 4)

The CM of a mature pinna exhibits an outermost electron dense layer of epicuticular wax followed by a polylamellate zone. This zone is made up of less compactly arranged parallel running 6-7 electron dense lamellae alternating with 5-6 electron translucent

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Plate 3.1.

- 1-3. *Cf. Dicrodium gouldii* RETALLACK. 1. 21.000x, 2. 46.500x, 3. 168.000x.
4. *Komlopteris indica* (FEISTMANTEL) BARBACKA. 6.600x.
5. *Pachypteris indica* (OLDHAM and MORRIS) BOSE and ROY. 10.650x.

Plate 3.2.

1. *Pachypteris indica* (OLDHAM and MORRIS) BOSE and ROY. 3.600x.
- 2,3. *Cycas circinalis* L. 2. 10.650x, 3. 25.350x.
4. *Cycas revoluta* THUNB. 15.600x.

Plate 3.3.

- 1,2. *Zamia fischeri* MIQ. 1. 4.500x, 2. 10.650x.
- 3,4. *Zamia furfuracea* AIT. 3. 4.500x, 4. 15.600x.

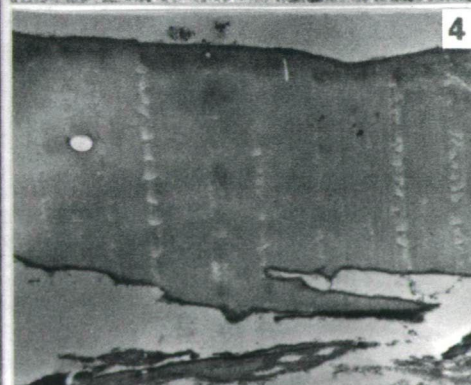
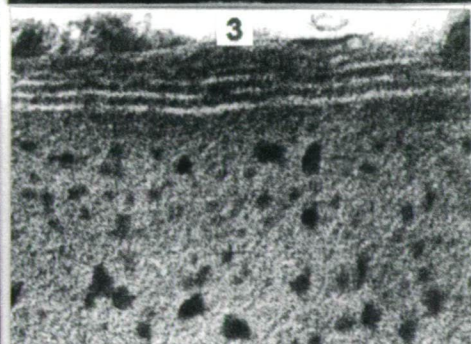
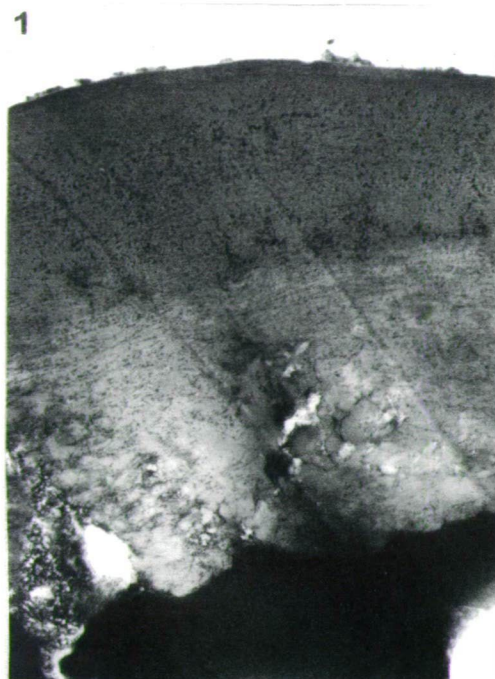


Plate 3.1.



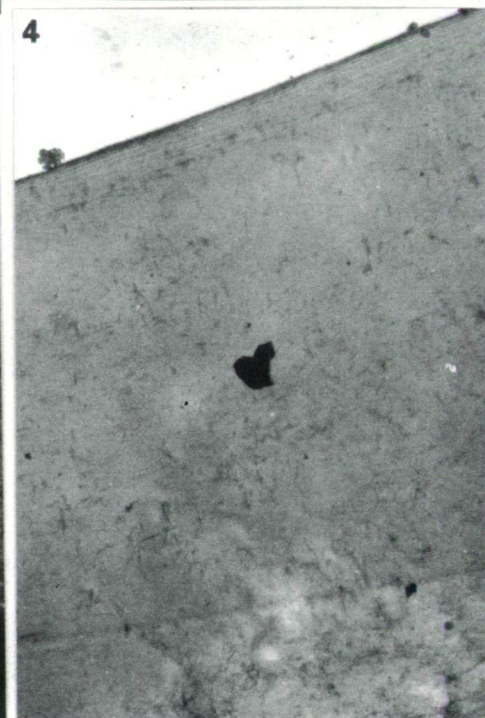


Plate 3.2.



Plate 3.3.

lamellae. A fine reticulate zone follows the polylamellate zone. At places the fine meshes in the CM are osmiophilic and have a coarse, almost channel-like appearance. At the innermost region where it adheres to the cell wall, the CM has a loose texture and appears fissured. At the base irregular masses of newly synthesized lipophilic secretions seem to permeate at sub-cuticular level.

*Zamia fischeri* MIQ. (Plate 3.3., figs. 1,2)

The CM is coated with epicuticular wax at the leaf-air interface. The structure of the wax layer varies from tubular, flat, fluted to at places fused along the edges and forming a thick highly electron dense layer. The outermost zone of the CM is polylamellate and is made up of robust and prominent 7-8 translucent and 8-9 opaque lamellae. The opaque lamellae vary in thickness at places. On the surface of lamellae, fine striations are present. An amorphous zone of constant homogeneous matrix follows this zone. The density of fibrillar network is very high at the junction with cell wall. The thickness of the cell wall of the epidermal cell also varies from place to place. The cytoplasm of the epidermal cell contains a well-defined nucleus with nucleolus, numerous ribosomes, endoplasmic reticulum, small vacuoles and numerous lipophilic granules.

*Zamia furfuracea* AIT. (Plate 3.3., figs. 3,4)

The CM is irregular in thickness, the thickness varies from one epidermal cell to other. The epicuticular wax is made up of wax flakes. At places this layer is detached from the CM. It is followed by a faintly polylamellate zone, with 4-5 lamellae. The lamellae are very loosely arranged and do not run parallel to each other. Some of the lamellae are oriented downwards towards the periclinal walls of the epidermal cells. The rest of the zone is structurally homogeneous. The basal portion of the CM attached to the surface of epidermal cell is electron dense zone because of the compactly arranged fibrillae.

### Discussion and Conclusions

The cuticular membrane in all the four species of extant cycads investigated shows a polylamellate outermost zone, though faintly so in *Zamia furfuracea*. On the other hand, of the three species of extinct "pteridosperms", only that of the *Corystospermaceae* shows a polylamellate outermost zone. This zone is absent in the two species of the *Peltaspermaceae*. Apparently the taxa of *Corystospermaceae* and *Peltaspermaceae* (both Mesozoic "pteridosperms") may not be closely related. However, more taxa need to be investigated before arriving at a definite conclusion. The "herring bone" structure seen in the CM of cf. *Dicroidium gouldii* is assumed to have developed under stress conditions as the plant was living in an arid environment.

The fibrillate nature of the inner zone in the CM of *Pachypteris indica* as compared to electron lucent homogeneous inner zone of *Komlopteris (Thinnfeldia) indica* indicates that the two taxa are different and may not belong to the same genus as believed by certain authors. The lipophilic bodies or vesicles observed in *Pachypteris indica* possibly added to the structural thickness of the CM as has been observed in extant *Cycas revoluta*.

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