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EPIDERMAL STUDIES OF LEAF IN SOME MEMBERS OF FAMILY TAXODIACEAE

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Leaves of family Taxodiaceae viz. *Sequoia sempervirens*, Endl., *Sequoiadendron giganteum*, Buch., *Taiwania cryptomerioides* Hayata and *Metasequoia glyptostroboides* Miki have been investigated using light and scanning electron microscopes, in order to evaluate their taxonomic significance. Haplocheilic stomata of various sizes occur on the under surface of leaves in *Sequoia sempervirens* Endl. and *Metasequoia glyptostroboides*, Miki whereas in *Taiwania cryptomerioides* Hayata and *Sequoiadendron giganteum* Buch., they occur on both the surfaces. In surface view anticlinal walls of epidermal cells appear straight, curved or undulated with raised ridges or irregular grooves. Periclinial walls are flat, slightly convex or concave. Other features of the epidermis that show variations are size of epidermal cells, guard cells, subsidiary cells, stomatal pore, stomatal frequency and stomatal index. The utility of these characters in the identification of each taxon is discussed in detail.

Key words : *Sequoia, Sequoiadendron, Taiwania, Metasequoia* epidermis, SEM.

The significance of epidermal characters of leaves in gymnosperms is well known. The family Taxodiaceae consists of 10 genera of which 7 are monotypic. *Taxodium* and *Cryptomeria* are the only two genera which are grown as garden plants in India. The world's tallest and largest trees the *Sequoias* viz. *Sequoia sempervirens* Endl. and *Sequoiadendron giganteum.* Buch., belong to this family. Except for the account on the foliar epidermis of some members of this family by Florin (1931) no other literature is available on most of the genera. Accordingly the present paper reports for the first time the leaf epidermal characters in four taxa. viz. *Sequoia sempervirens* Endl., *Sequoiadendron giganteum* Buch., *Taiwania cryptomerioides* Hayata and *Metasequoia glyptostroboides* Miki as observed under light and scanning electron microscopes. It describes the significance of variable epidermal features and the extent of their application in the taxonomic identification of these genera.

MATERIALS AND METHODS

Material for the present investigation was obtained from Royal Botanical Garden, Edinburgh and Arnold Arboretum, Massachusettes, U.S.A.

Light Microscopy : Cuticles were isolated in Schulze's fluid (conc. HNO_3 + $KClO_3$). The acid treated substance was washed in water and subjected to alkali treatment (10% ammonium hydroxide), Cuticles were separated with the help of needles and mounted in safranin glycerine jelly after washing in water. Slides were made permanent and sealed with colorless Kopal varnish. For each taxa a minimum of 15 leaf samples were studied. Measurements were based on 25 readings in each case. Wild Leitz Biomed microscope had been used for photomicrography.

Scanning Electron Microscopy : It was done at National Botanical Research Institute, Lucknow. Fragments of suitable size of the mature leaf samples were dehydrated serially in 50-100% alcohol dried and then coated with gold and examined with the JSM 35 C JEOL, Japan Scanning Electron Microscope. All electron micrographs were made at the Electron Microscopy Unit, National Botanical Research Institute, Lucknow, India.

RESULTS AND DISCUSSION

. In the following account both light and electron microscopic structure of cuticle and epidermis is described in a chronical manner of each genera studied individually.

Sequoia sempervirens Endl Californian Redwood : Cuticle thin, hypostomatic epidermal cells arranged in longitudinal rows, end walls transverse or oblique. Cuticle of leaf showkng differentiation of stomatiferous and non-stomatiferous bands. In the stomatiferous zone stomata arranged in a number of longitudinal rows. Epidermal cells over non stomatiferous region are rectangular longitudinally elongated 20-40 µm long x 25-50 µm wide. Anticlinal walls straight end walls oblique. Epidermal cells in stomatiferous region

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Table 1: Epidermal characteristics of four genera of Taxodiaceae.

S. No.	Name of the genera	Epidermal cells (size in µm)		Stomata				
		S.R. (µm)	N.S.R. (µm)	Size of G.C. (µm)	Size of S.C. (µm)	Size of S.P. (µm)	S.F. (µm)	S.I. (mm^2)
1.	*Sequoia sempervirens*	20(48) 60 x 12(18)25	20(32)40 x 25(42)50	20(32)38 x 7(8)10	18(24)30 x 6(7)9	14(16)20 x 0(0.5)1	15.2	17.34
2.	*Taiwania cryptomerioides*	20(64)80 x 17(20)23	18(26)37 x 24(32)40	20(32)39 x 7(8)12	16(24)30 x 6(7)10	14(18)25 x 0(0.5)1	25.32	27.27
3.	*Sequoiadendron giganteum*	30(82)100 x 20(31)35	20(26)37 x 28(34)42	24(36)42 x 7(8)10	18(26)32 x 6(7)9	14(17)22 x 0(0.5)1	21.22	23.81
4.	*Metasequoia glyptostroboides*	20(48)56 x 12(18)24	20(26)38 x 26(32)40	20(32)38 x 7(8)9	15(24)30 x 6(6)8	14(20)24 x 0(0.5)1	13.42	15.63

Figures given in the parenthesis are mean values
S.R. = Stomatiferous region NSR = Non Stomatiferous region
G.C. = Guard cells. S.C = Subsidiary cells S.P. = Stomatal pore
S.F. = Stomatal frequency S.I. = Stomatal index

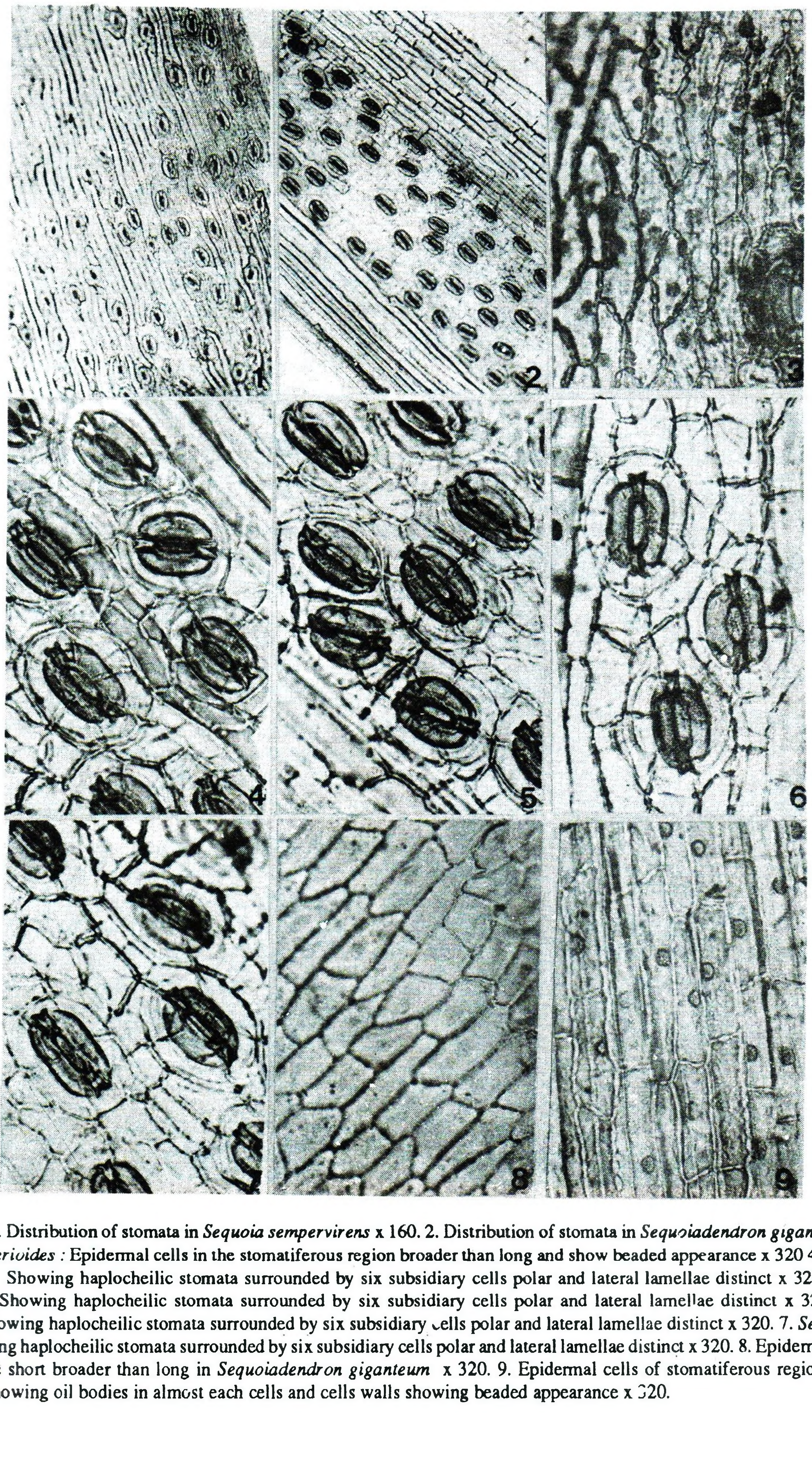
thin walled and arranged in regular longitudinal rows, 20-60 µm long x 12-25 µm wide. Stomata haplocheilic transversely or obliquely placed, guard cells surrounded by 6 subsidiary cells, lateral subsidiary cells radially elongated. Size of cells 18-30 µm long x 6-9 µm wide. Guard cells deeply sunken 20-38 µm long x 7-10 µm wide, size of stomatal pore 14-20 µm long x 0-1 µm wide. Polar and lateral lamellae thick and distinct. Stomatal index is 17.34 and stomatal frequency is 15.2 st/mm^2. Epidermal cells along margins narrower and arranged in longitudinal rows transverse walls oblique (Pl. 1, figs 1 & 6).

Abaxial anticlinal walls are marked by slightly undulate ridges of varying height and thickness. Stomata numerous slightly sunken with pronounced rim. Small wax flakes are sometimes irregularly scattered at places (Pl. 2 figs 12 & 14).

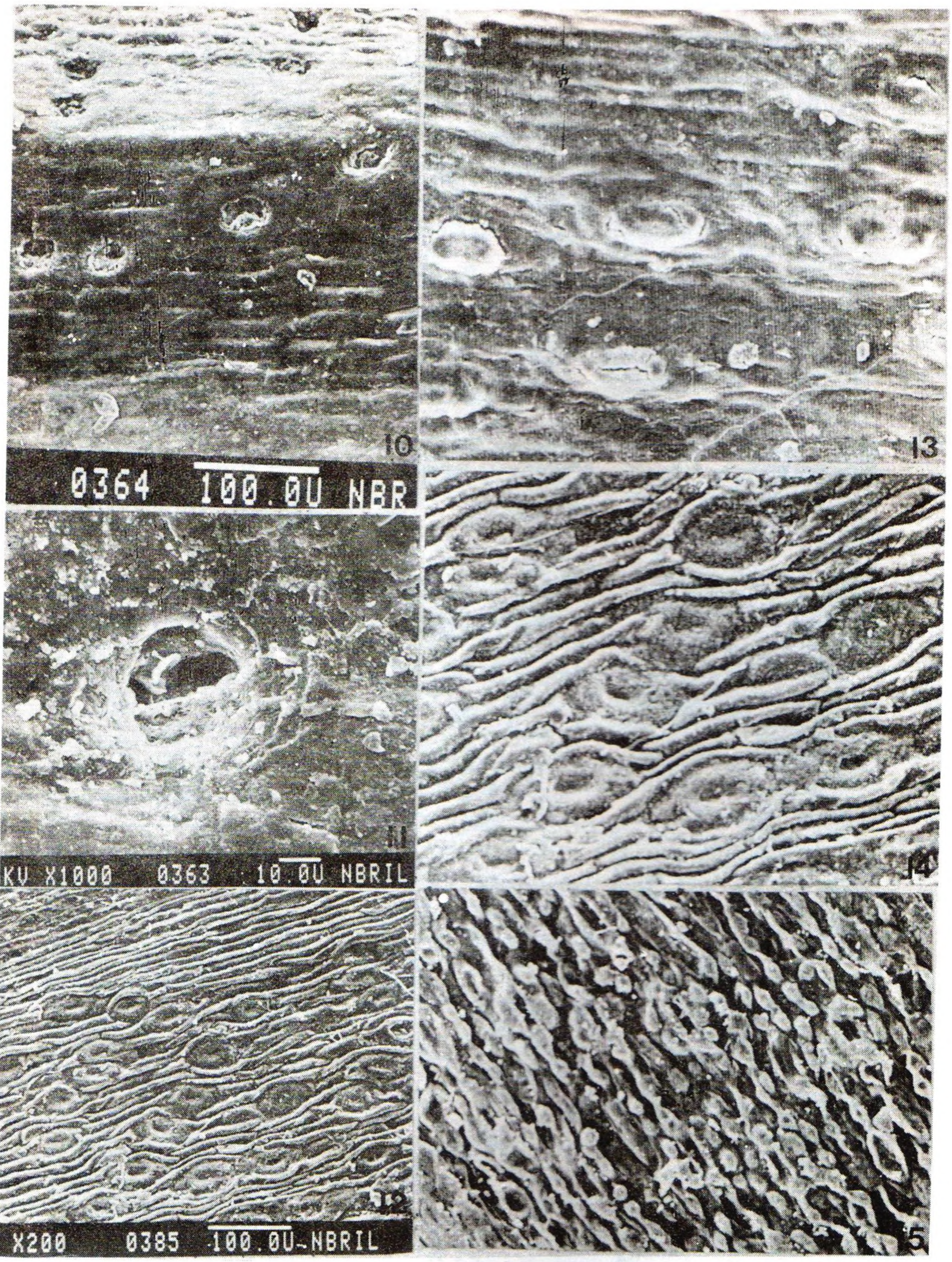
Taiwania cryptomerioides Hayata : Cuticle thin, amphistomatic, epidermal cells arranged in longitudinal rows, end walls transverse or oblique. Cuticle of leaf showing differentiation of stomatiferous and non stomatiferous bands. In the stomatiferous zone stomata arranged in two distinct white bands in longitudinal rows. Epidermal cells over the non stomatiferous region thin walled and arranged in longitudinal rows cells broader than long, 18-37 µm long x 24-40 µm wide and each cell contains oil bodies, these are found in the stomatiferous zone as well. Anticlinal walls straight end walls oblique. Epidermal cells in the stomatiferous region are thin walled and arranged in regular longitudinal rows 20-80 µm long x 17-23 µm wide their walls show beaded appearance. Stomata haplocheilic their orientation transverse but at places slightly oblique, guard cells surrounded usually by 6 subsidiary cells but sometimes their number may be 4 or 7, lateral subsidiary cells radially elongated. Size of cells 16-34 µm long x 6-10 µm wide. Guard cells deeply sunken 20-39 µm long x 7-12 µm wide. Size of stomatal pore 14-25 µm long x 0-1 µm wide. Polar and lateral lamellae thick and distinct. The stomatal index is 27.27 and stomatal frequency is 25.32 st/mm^2. Epidermal cells along margins narrow and arranged in longitudinal rows, transverse walls oblique. (Pl. 1, figs 3, 5 & 9).

Abaxial anticlinal walls almost straight ridges of varying height and thickness. Stomata in longitudinal rows slightly sunken. Wax thickly deposited on the stomata (Pl. 2, Fig 13).

Sequoiadendron giganteum Buch. Big Wood : Cuticle thin amphistomatic, epidermal cells arranged in longitudinal rows end walls transverse or oblique. Cuticle of leaf showing differentiation of stomatiferous and non stomatiferous bands. In the stomatiferous zone stomata arranged as two distinct bands in longitudinal rows. Epidermal cells over the non-stomatiferous region thin walled and arranged in longitudinal rows cells broader than long 20-37 µm long x 28-42 µm wide. Anticlinal walls straight end walls oblique. Epidermal cells in the stomatiferous region are thin walled and arranged in regular longitudinal rows 30-100 µm long x 20-35 µm wide. Stomata haplocheilic transversely or



Figures 1-9. Fig. 1. Distribution of stomata in *Sequoia sempervirens* x 160. 2. Distribution of stomata in *Sequoiadendron giganteum* x 160. 3. *Taiwania cryptomerioides* : Epidermal cells in the stomatiferous region broader than long and show beaded appearance x 320 4. *Metasequoia glyptostroboides* : Showing haplocheilic stomata surrounded by six subsidiary cells polar and lateral lamellae distinct x 320. 5. *Taiwania cryptomerioides* : Showing haplocheilic stomata surrounded by six subsidiary cells polar and lateral lamellae distinct x 320. 6. *Sequoia sempervirens* : Showing haplocheilic stomata surrounded by six subsidiary cells polar and lateral lamellae distinct x 320. 7. *Sequoiadendron giganteum* : Showing haplocheilic stomata surrounded by six subsidiary cells polar and lateral lamellae distinct x 320. 8. Epidermal cells of non stomatiferous zone short broader than long in *Sequoiadendron giganteum* x 320. 9. Epidermal cells of stomatiferous region in *Taiwania cryptomerioides* showing oil bodies in almost each cells and cells walls showing beaded appearance x 320.



Figures 10-15. Scanning Electron Micrographs

Fig. 10. *Sequoiadendron giganteum* showing abaxial anticlinical walls marked by shallow furrows, few stomata are seen either sraised or sunken x 200. 11. Portion in figure A more magnified to show a single sunken stoma with narrow over-arching cuticular rim. Wax is found as small flakes and several micropapillae are seen on athe periclinal walls x 1000. 12. *Sequoia sempervirens* showing abaxial anticlinal walls which are marked by slightly undulate ridges. Stomata numerous, slightly sunken withs pronounced rim x 200. 13. *Taiwania cryptomerioides* showing almost straight abaxial anticlinal walls, ridges of varying height and thickness. Stomata sunken and wax thickly deposited on stomata x 1000. 14. *Sequoia semperyirens* portion in fig. 12 more magnified to show numerouos stomata which are slightly sunken pronounced rim small wax flakes are sometimes regularly scattered at places x 1000. 15. *Metasequoia glypatostroboides* anticlinal walls showing pronounced papillae in the stomatiferous region. Stomata in longaitudinal rows subsidiary cells papillate over arching the guard cells x 1000.

obliquely placed, guard cells surrounded by 6 subsidiary cells, lateral subsidiary cells radially elongated size of ceils 18-32 µm long x 6-9 µm wide. Guard cells deeply sunken 24-42 µm long x 7-10 µm wide. Size of stomatal pore 14-22 µm long x 0-1 µm wide. Polar and lateral lamellae distinct. The stomatal frequency is 21-22 st/mm² and stomatal index is 23.81. Epidermal cells along margins narrow and arranged in longitudinal rows transverse walls oblique (Pl. 1, figs. 2, 7 & 8).

Abaxial anticlinal walls are marked by shallow furrows of varying width. Periclinal walls are prominently convex with a low relief pattern of small micropapillae. Stomata are either raised or sunken with narrow over-arching cuticular rim. Wax is found as very few small flakes. Adaxial anticlinal walls are indicated by distinctly raised ridges of variable thickness. The outer walls also bear a low relief pattern, several micropapillae are more pronounced on the periclinal walls (P1,2 figs 10 & 11).

Metasequoia glyptostroboides Miki Dawn Redwood : Cuticle thin hypostomatic, epidermal cells arranged in longitudinal rows end walls transverse or oblique. Cuticle of leaf showing differentiation of stomatiferous and non stomatiferous bands. In the stomatiferous zone stomata arranged in longitudinal rows. Epidermal cells over the none stomatiferous region thin walled and arranged in longitudinal rows 20-38 µm long x 26-40 µm wide, cells broader than long Anticlinal walls straight end walls oblique. Epidermal cells in the stomatiferous region are thin walled and arranged in regular longitudinal rows 20-56 µm long x 12-24 µm wide. Stomata haplocheilic transversely or obliquely placed, guard cells surrounded by 6 subsidiary cells, lateral and polar lamellae distinct. Subsidiary cells radially elongated size of cells 15-30 µm long x 6-8 µm wide. Guard cells deeply sunken 20-38 µm long x 7-9 µm wide. Size of stomatal pore 14-24 µm long x 0-1 µm wide. Stomatal frequency is 13.42 st/mm² and stomatal index is 15.63. Epidermal cells along margin narrow and arranged in longitudinal rows transverse wall oblique (Pl. 1, fig. 4).

Anticlinal walls showing pronounced papillae in the stomatiferous region and usually absent over the non stomatiferous region. Stomata in longitudinal rows, subsidiary cells papillate over-arching the guard cells. Cells in the non-stomatiferous region arranged in longitudinal rows anticlinal walls ridged (Pl. 2, fig. 15). The variations in the epidermal characters of the four taxa can be attributed to various factors like size of epidermal cells, guard cells, subsidiary cells, stomatal frequency, stomatal index, wax deposition etc. as shown in Table 1 and in the key.

Key

1. + Papillae present .....2.
   - Papillae absent .....3.
2. +Stomata on both surfaces. Anticlinal walls with shallow furrows. Stomata scattered - *Sequoiadendron giganteum*.
   - Stomata on under surface. Anticlinal walls without shallow furrows. Stomata in longitudinal rows - *Metasequoia glytostroboides* sp.
3. + Stomata on undersurface. Anticlinal walls with slightly undulate ridges. Wax in small flakes- *Sequoia sempervirens*.
   - Stomata on both the surfaces Anticlinal walls with straight ridges. Wax thickly deposited - *Taiwania cryptomerioides*.

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