

DEVELOPMENT OF STOMATA IN TWO SPECIES OF *RHAPIS* L.¹

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ABSTRACT

The development of stomata in *Rhapis excelsa* Ait. and *Rhapis humilis* Blume. has been studied in detail. The two species show the same pattern of developmental stages of stomata. However, the mature structures show differences in the length of the polar cells. The ontogeny of stomata is perigenous and the mature stomata are paracytic and tetracytic.

INTRODUCTION

A good deal of work has been done by various workers like Stebbins and Jain (1960) Stebbins and Kush (1961), Tomlinson (1961, 1974), Ghose and Davis (1974), Ghose (1979) on the morphology, anatomy and stomata in palms. Excepting the attempts made by Ghose and Davis (1973), Ghose (1979) and Trivedi and Upadhyay (1979) the developmental aspects of stomata and trichomes have been practically neglected. Previous literature on this particular aspect of palms is rather confusing as has already been discussed by Tomlinson (1974). The present paper deals with the development of stomata in two species of *Rhapis*, *R. excelsa* and *R. humilis*.

MATERIALS AND METHODS

Young developing leaves were fixed in Acetic alcohol (Glacial acetic acid 1 : Ethyl alcohol 3) for 24 to 48 hours and were then transferred to 70% alcohol. The material was hydrolysed in 1 N HCl

for 3 to 5 minutes at 60°C and then thoroughly washed with water to make it acid free.

The hydrolysed material was then transferred to 70% alcohol which helped in hardening as well as separating of the epidermal tissue. The epidermal peel was stained with 1% acetocarmine.

RESULTS

The stomata in both the species of *Rhapis* arise in longitudinal rows. The formation of new stomatal initials continue for a considerable period during the growth of the leaf. Stomata occur on the lower epidermis, in between the veins and on the leaf margins in the upper epidermis.

Development:—The epidermal cells in a very young leaf divide in various planes—oblique, longitudinal and transverse. The result of these divisions is expansion of epidermis and formation of trichomes and stomata. A further stage shows cell files which can be distinguished from one another. The cell files with

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alternate small and large cells can be seen. These cells are more or less isodiametric. The smaller cell has dense cytoplasm and deeply stained nucleus which almost completely occupies the cell. On either side of these distinctive cell file or files there are cell files with elongated cells (Figs. 1, 7 ; 13, 14).

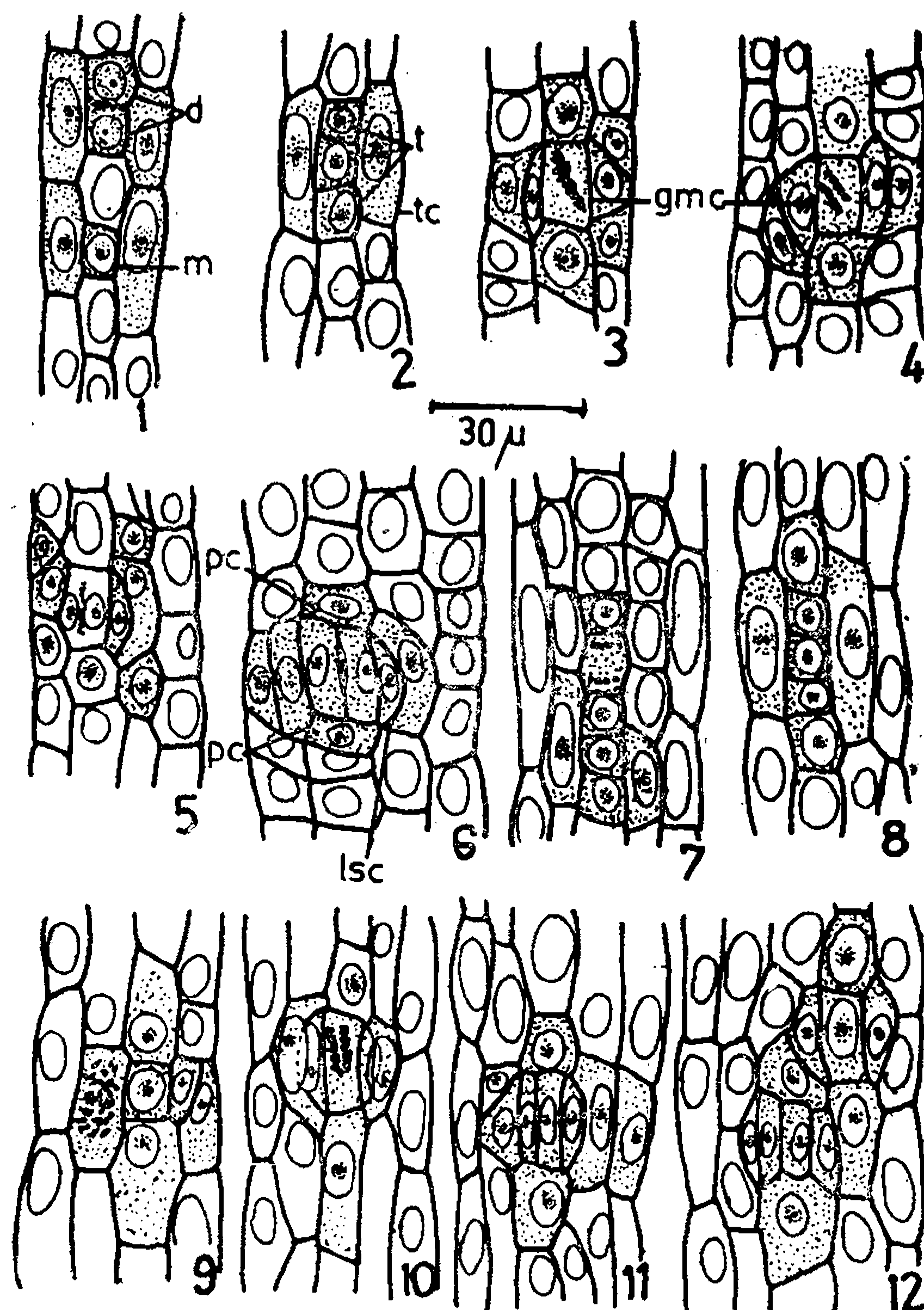
The smaller cell further divides to form a row of two cells—a diad or three cells—a triad or a many-celled file of cells which stands out because of the deeply stained nuclei. (Fig. 15). Some of these cells enlarge and become more or less rectangular having a large nucleus. This is the guard cell mother cell (Figs. 1-3, 7-9 ; 16).

By the time these cells are differentiated as guard cell mother cells, the cells lateral to them become trapezoidal because of oblique divisions in them. (Figs. 2, 8 ; 15, 16). This results in the formation of two or three cells. The oblique walls are generally non-intersecting. The trapezoidal cell thus formed adjacent to the guard mother cell again divides by a wall parallel to the wall of the guard mother cell in a diagonally oblique fashion thus forming two partly superposed cells, the outer cell occupying upper position (Figs. 4, 5, 9, 11, 16, 17). This is how the stoma appears sunken. The final structure thus has a pair of partly superposed parallel cells on either side and one polar cell at each end of the guard cell mother cell.

The cells terminal to the gmc divide transversely or obliquely depending on their position in relation to lateral cells. Normally they divide transversely but in case they lie lateral to gmc of the adjacent cell file, they divide obliquely to form a pair of subsidiary cells (Figs. 12, 16, 17).

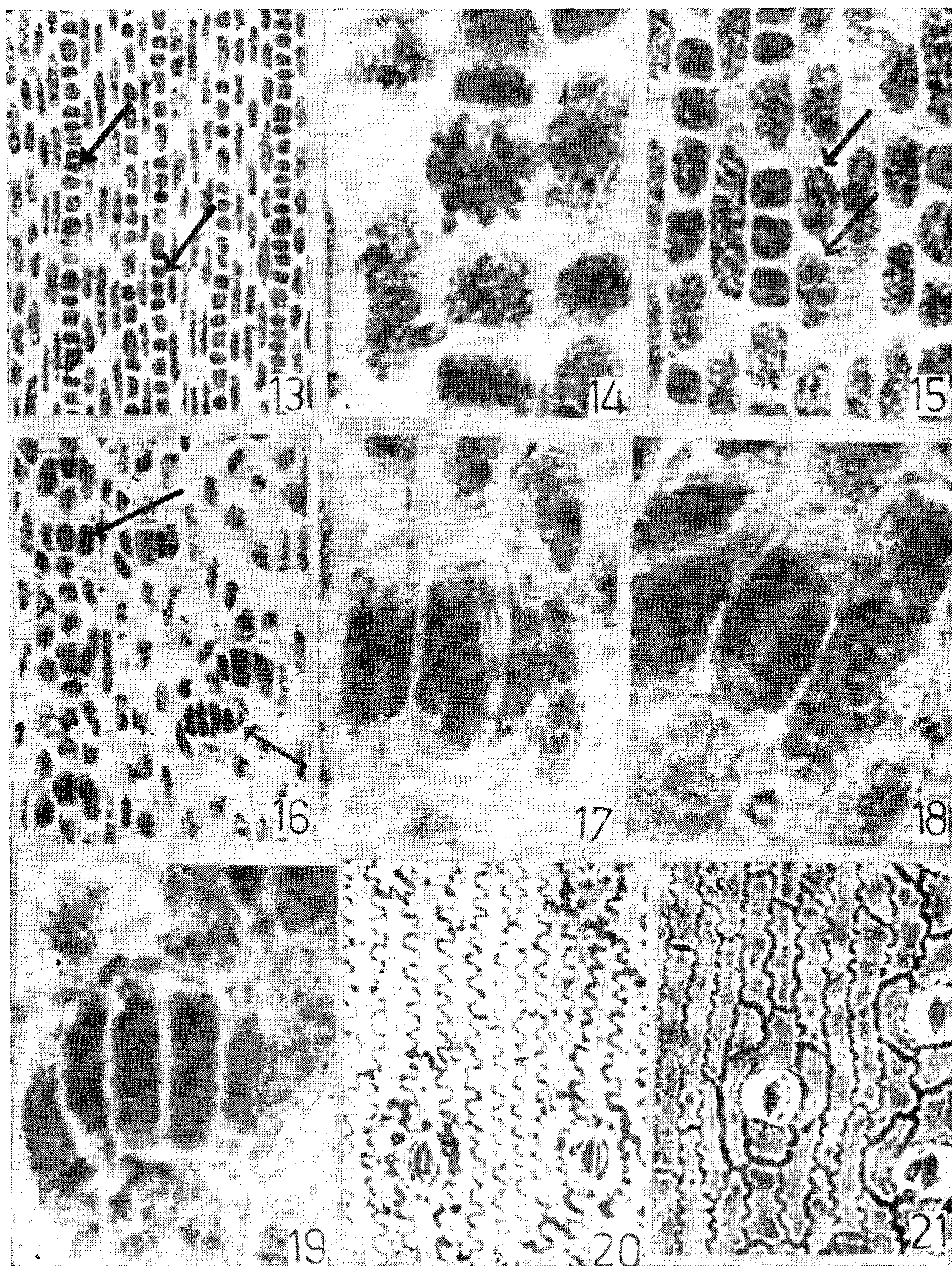
The cells in contact with the gmc are usually in an active state of division. The gmc is the last cell to divide in this

configuration. The orientation of the metaphase spindle in the gmc is, to begin with, oblique to the long axis. Later on, however, it rotates to lie parallel to the



Figures 1-6. Development of stomata in *R. excelsa* Ait. 1. figures showing a diad-*d*, meristemoid-*m*. 2. prototype stomatal configuration-triad-*t*, trapezoidal cell-*tc*. 3. guard mother cell-*gmc*, showing oblique orientation of metaphase plate. 4. *gmc* showing early anaphase (oblique orientation). 5. parallel orientation of daughter cells of *gmc* guard cells. 6. the two daughter cells of *gmc* separate to form a pore.

Figures 7-12 : Development of stomata in *Rhapis humilis* Blume. 7. a triad, one of the cells in the meristematic cell file showing anaphase. 8. triad or a prototype stomatal configuration. 9. division in one of the lateral cells of *gmc*. 10. completion of division in lateral cells and anaphase stage in *gmc* which is the last cell to divide. 11. two daughter cells of *gmc*. 12. polar cell of one of *gmc* acting as lateral cell of *gmc* of adjacent cell file.



Figures 13-21 Development of stomata in *Rhipis* L. 13. *R. humilis* Blume. Files of isodiametric meristematic cells ($\times 232$). 14. Isodiametric cell dividing ($\times 834$). 15. Cell lateral to meristematic cell files divides obliquely and shows oblique walls ($\times 696$). 16. Tetarcytic condition ($\times 179$). 17. Metaphase plate obliquely oriented ($\times 710$). 18. Early telophase in the gmc showing oblique orientation of daughter cells $\times (834)$. 19. The guard cells showing parallel orientation ($\times 1083$). 20. Stomata showing long polar cells $\times (250)$. 21. *R. excelsa* Ait.-stomata showing short polar cells ($\times 196$).

cell file before the telophase is complete (Figs. 3-5, 10 17, 18, 19).

Finally the two daughter cells of the gmc are transformed into kidney, shaped guard cells. The ledges are developed later on along the margins of the guard cells which surround the stoma (Fig. 6 ; 20, 21).

DISCUSSION

Stomatal ontogeny in monocots is reported to be perigenous by Pant and Kidwai (1966), Paliwal (1969). Tomlinson (1974) and Ghose (1979) but Williams (1974) reported it to be mesoperigenous in *Ludisia discolor* a member of Orchidaceae.

According to Payne (1970) any monocotyledonous stoma with terminal subsidiary cells is not perigenous. In *Rhapis*, the cells of stomatiferous cell file are derivatives of the same mother cells or their daughter cells. Differentiation of guard cell mother cell (gmc) and polar cells takes place in due course. The polar cells remain as such with or without distinction from other epidermal cells at the poles of the guard cells. How far they are involved in stomatal functioning is difficult to predict. However, at places and in different species they are different from other epidermal cells. In *R. humilis* the polar cells are distinct at regions where the stomatal frequency is less. In other regions they are not distinct. The inner cells from the lateral pair of cells are thin walled and contiguous with the guard cells and different from other epidermal cells in both the above cases, and hence can be designated as subsidiary cells. The mature stomata are therefore of two types viz., paracytic and tetracytic. In *R. excelsa* a condition exactly opposite to above is seen. Here the frequency of tetracytic stomata is more than the paracytic ones.

As in *Allium* (Palevitz & Hepler,

1974) here also the dividing nucleus in the guard cell mother cell is obliquely oriented in the beginning. The lateral trapezoidal cells also show obliquely oriented dividing nucleus indicating probable functional relationship between the two. The orientation of the metaphase plate in the polar cells however, is usually transverse unless it lies adjacent to the gmc in the neighbouring cell file. Simultaneous divisions in the lateral subsidiary cell and the terminal cells indicate the induction of meristematic activity by the gmc in the surrounding cells as has been suggested by Stebbins and Khush (1961).

All these observations point to the perigenous type of stomatal development and paracytic and tetracytic types of mature stomata.

REFERENCES

- GHOSE, M. AND T. A. DAVIS 1973. Stomata and trichomes in leaves of young and adult palms. *Phytomorphology* **24** : 216-229.
- GHOSE, M. AND T. A. DAVIS 1974. Ontogeny and structure of stomata in *Cocos nucifera* L. *J. Indian bot. Soc.* **53** : 19-23.
- GHOSE, M. 1979. Ontogenetic study of stomata and trichomes in some palms. *Phytomorphology* **29** : 26-33.
- PANT, D. D. AND P. F. KIDWAI 1966. Structure of leaves and stomatal ontogeny in some Pandanales and Spathiflorae. *Senckenberg. Biol.* **47** : 308-333.
- PALIWAL, G. S. 1969. Stomatal ontogeny and phylogeny I. Monocotyledons. *Acta Bot. Neerl.* **18** : 655-668.
- PAYNE, W. W. 1970. Helicocytic and allelocytic stomata : Unrecognised pattern in the dicotyledons. *Am. J. Bot.* **57** : 140-147.
- PALEVITZ, B. A. AND P. K. HEPLER 1973. Studies on cell plate orientation during stomatal differentiation in *Allium*, *J. Cell. Biol.* **59** : 258a.
- STEBBINS, G. L. AND S. K. JAIN 1960. Developmental studies of cell differentiation in the epidermis of monocots I. *Allium*, *Rhoeo* and *Commelina*. *Develop. Biol.* **2** : 409-426.
- STEBBINS, G. L. AND G. S. KHUSH 1961. Variation in the organisation of stomatal complex in the leaf epidermis of monocotyledons and

- its bearing on their phylogeny. *Am. J. Bot.* **48** : 51-59.
- TOMLINSON, P. B. 1961. *Anatomy of monocotyledons Vol-II* Oxford.
- TOMLINSON, P. B. 1974. Development of the stomatal complex as a taxonomic character in the monocotyledons. *Taxon* **23** : 109-128.
- TRIVEDI, B. S. AND NIRMALA UPADHYAY 1979. Epidermal structure in Palmae. *Biol. Mem.* **4** : 83-117.
- WILLIAMS, N. H. 1975. Stomatal development in *Ludisia discolor* (Orchidaceae) : mesoperigenous subsidiary cells in the monocotyledons. *Taxon* **24** : 281-288.