*J. Phytol. Res.* 23(1) : 9-16, 2010

# ORIGIN OF PHELLOGEN IN SOME TROPICAL TREES

J. ASOKAN
Department of Plant Biology and Plant Biotechnology, Sir Theagaraya College, Tamil Nadu, Chennai – 600 021, India.

Our scientific knowledge of bark, especially of the phloem, dates back to seventeenth century. Since then there has been a slow and steady progress of research on different aspects of the bark tissues. Initiation of the first phellogen occurs in various species at different sites. Moreover, the site of phellogen initiation may vary within the same family and even within the same plant. In a given organ phellogen may originate under various environmental conditions and at different distances from the apex. The factors that operate the onset of the first phellogen and its subsequent functioning seem to be highly variable. In this present study phenology and origin of phellogen in five plants of different families were studied and the results are interpreted.

**Keywords:** Bark; Periderm; Phellogen.

## Introduction

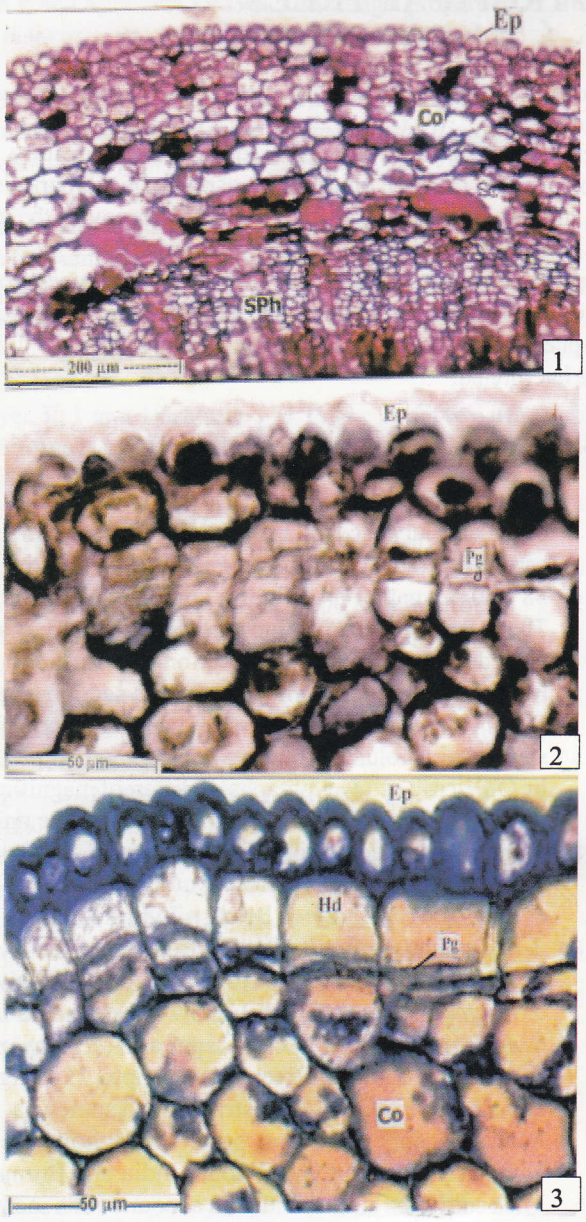
Our knowledge of Phellogenesis with respect to its place, time and its controlling factors is scanty. As van Wyk *et al.*[1] have rightly pointed out, our understanding of the different aspects of initial periderm formation is equally important as the knowledge of mature periderm / rhytidome as well as the entire bark. Arzee *et al.*[2], on the basis of their studies on *Robinia pseudacacia* contended that temperature, day length, growth substances were major factors in initiating phellogen. Ahmad *et al.*[3] found that the phellogen originated in the subepidermal layer in *Mangifera* and *Acacia*, the age of the shoots being 18 months in the former and two weeks in the latter plants. In *Mangifera* it took three years for spreading of the phellogen all round the stem. In *Psidium*, Khan and Ghouse[4] recorded deeper origin of the phellogen from the innermost cortex of 6-8 week old stems. A simple instance of phellogen origin from the cortical collenchyma was observed by Ghouse and Yunus[5] in *Dalbergia sissoo* and subsequent periderm originated from the nonconducting phloem of this plant. Perusal of literature dealing with the first phase of periderm formation showed that there is much scope for this study especially in certain tropical trees. Based upon these facts the origin of first periderm in a branch in relation to the distance from the apex of the twig in five species was studied.

## Material and Method

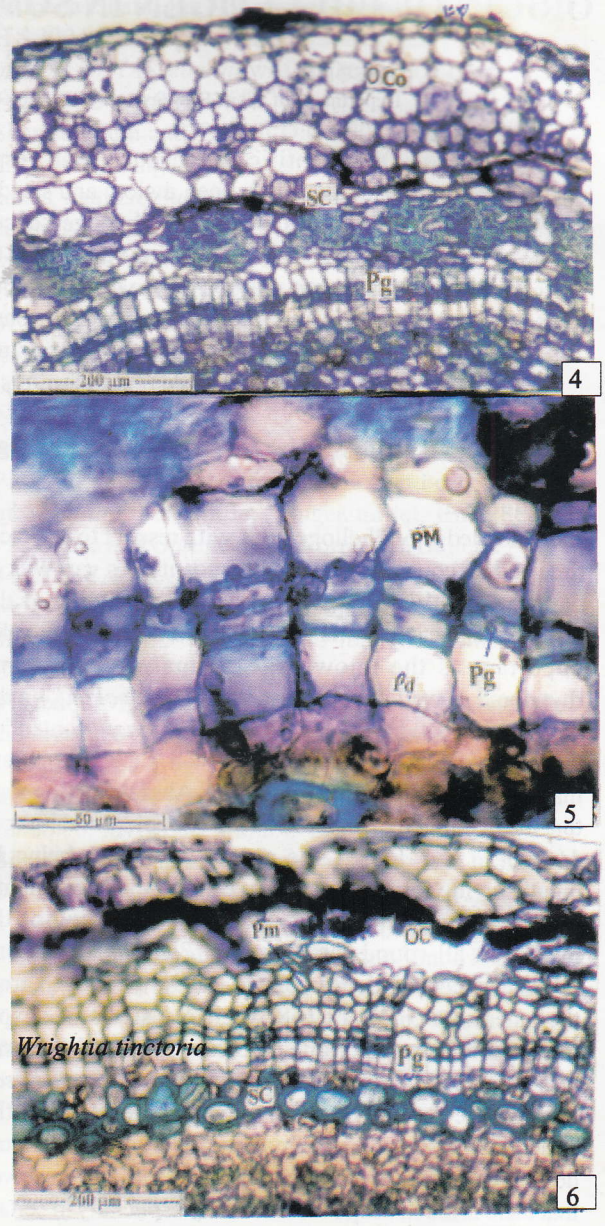
Following five tree species of different natural orders, growing in the Academy campus in Madras was selected for studies. Care was taken to select healthy plants and for normal organs. The required samples were cut and removed from the plant and fixed in FAA. After 24 hrs of fixing the specimens were dehydrated with graded series of tertiary Butyl alcohol. Infiltration of the specimens was carried by gradual addition of paraffin wax (melting point 58 - 60° C) until TBA solution attained supersaturation. The specimens were cast into paraffin blocks. The paraffin embedded specimens were sectioned with the help of rotary microtome. The thickness of the sections was 10 - 12 µm. The sections were dewaxed and the sections were stained with Toluidine blue as per the method published by O'Brien *et al.*[6]. Photographs of different magnifications were taken with Nikon Labphot 2 Microscopic unit. For normal observations bright field was used.

*1. Aegle marmelos* (L.) Corr. – Rutaceae
*2. Lagerstroemia reginae* Roxb. – Lythraceae
*3. Putranjiva roxburghii* Wall. – Euphorbiaceae
*4. Thespesia populnea* (L.) Soland ex Correa – Malvaceae
*5. Wrightia tinctoria* (Roxb.) R.Br. - Apocynaceae.

1. *Aegle marmelos* (L.) Corr. (Rutaceae) - *Aegle marmelos* (L.) Corr. is an armed tree growing upto 15m high and about a metre in girth with a straight and somewhat fluted bole. It occurs in natural state throughout the Sub-Himalayan tract, in Central South India and Burma. It is often cultivated all over India on account of its sacred and medicinal importance. Economically; the tree is very sacred among Hindus and often planted in temples. The aromatic pulp of the fruits is highly medical. The bark of the tree is considered efficacious for intermittent fever and it constitutes the main ingredient of the Ayurvedic drug "dasamula". The tree generally starts defoliation in the month of December and remains totally defoliated in January and in the beginning of February. Bud break starts in the middle of February. Flowering is initiated in March.



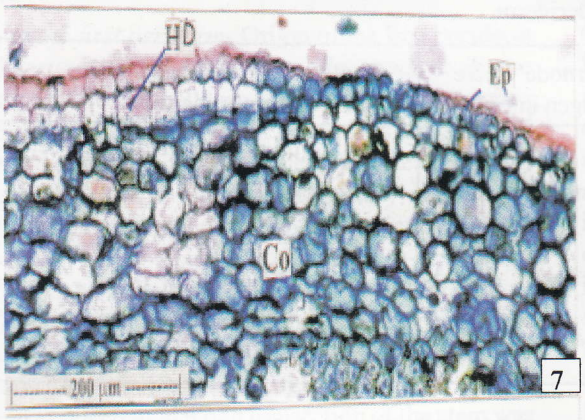

*Aegle marmelos*

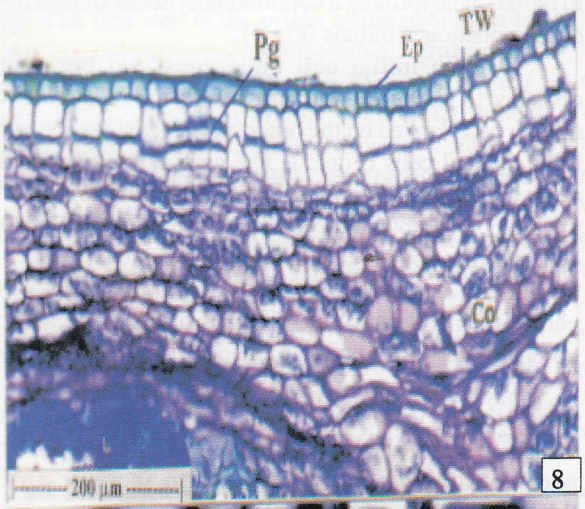


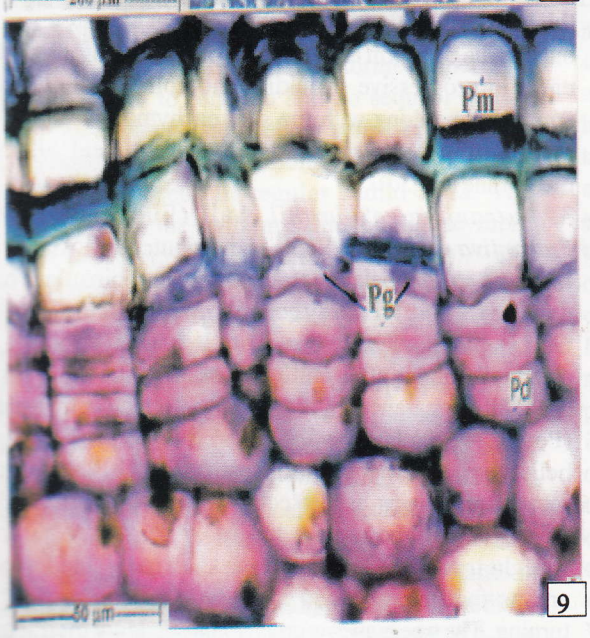

*Lagerstroemia reginae*

Mature leaves and fruits are seen upto the beginning of December.

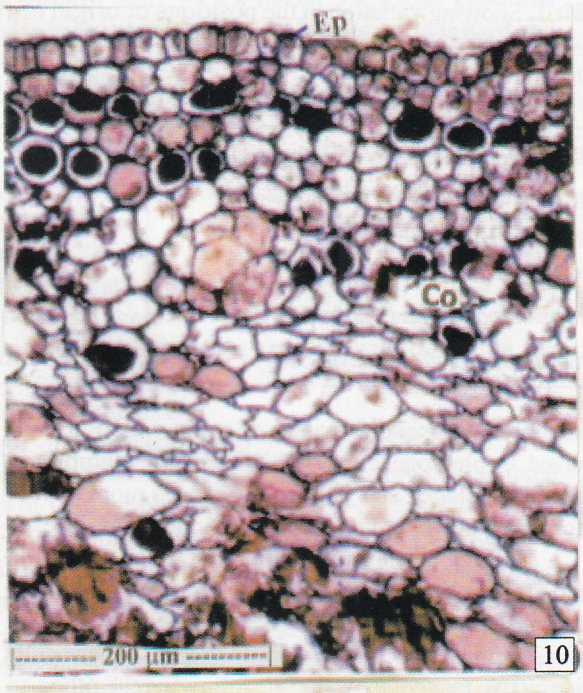
*Origin of the first phelloegen* - Numerous young branches were examined to locate the internode where the first phellogen was initiated. It was found that in all the specimens studied, the first phellogen originated in the tenth internode. In transection of the tenth internode the stem is circular and even; there is a layer of small, papillate, very thickly cuticularised epidermal cells followed by chlorenchymatous and parenchymatous cortical zones (Fig. 1). The phellogen appears in the third layer of cells inner to the epidermis (Fig. 2). It originates in isolated narrow regions with no specificity of its location. However, it was found that the phellogen originated early on the lower and later on the upper sides of the same branch. The narrow strips of phellogen initiated along different locations, function vigorously producing a hemispherical hump of cubical, thin walled parenchymatous cells. Due to the pressure

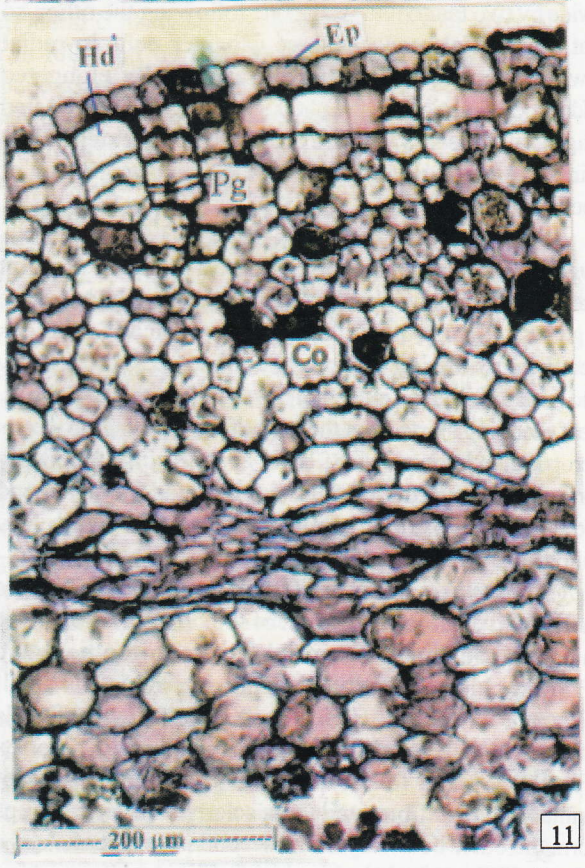



*Putranjiva roxburghii*




*Thespesia populnea*

**Table 1.** Showing variation in the positions of origin of first periderm.

| Name of the plant | No. of internode where first phellogen initiated | Depth of the phellogen initials from epidermis |
|---|---|---|
| *Aegle marmelos* (Rutaceae) | Tenth | First to third layer of cortical cells |
| *Lagerstroemia reginae* (Lythraceae) | Third/Fourth | Pericycle |
| *Putranjiva roxburghii* (Euphorbiaceae) | Tenth | Subepidermal layer |
| *Thespesia populnea* (Malvaceae) | Third | Subepidermal layer |
| *Wrightia tinctoria* (Apocynaceae) | Fourth | Subepidermal layer |

exerted by these isolated masses of cells, which are not involved in phellogenesis, peel off as thin membrane. Later, the phellogen appears in other places more or less uniformly round the stem in similar manner as before.

During the origin of the first phellogen, the phellogen initials, usually the first to third layer of cortical cells beneath the epidermis, elongate considerably in radial plane assuming a broad layer of oblong cells. These cells divide by two tangential walls, one following the other, so that each initial produces three daughter cells in radial row. Of these cells, the narrow tangentially elongated middle cell functions as the phellogen (Fig. 3). By repeated tangential divisions the phellogen produces externally a thin zone of phellem cells and one or two layers of phelloderm internally.

2. *Lagerstroemia reginae* Roxb. Lythraceae - *Lagerstoremia reginae* Roxb. is a medium sized tree, but it attains different heights depending upon the region where it grows. *Lagerstroemia reginae* is a typical deciduous tree. Leaf withering starts in the third week of January and total defoliation is seen in the second week of March. Leaf shedding starts from the base of the tree and proceeds acropetally. However leaf buds appear simultaneously throughout the tree. Leaf buds are initiated in the second week of January. Flowering is during June and July followed by fruit setting.

*Origin of the first periderm*- The first periderm originates in the third or fourth internode of the branch. The site of initiation of the phellogen is the inner to the pericylic (Primary phloem) fibres. The young internode has small elliptical, thickly cuticularised epidermal layer of cells followed by five or six layers of parenchymatous cortex (Fig. 4). This is followed by small isolated groups of primary phloem fibres and these sclerenchymatic groups form the outer boundary of the secondary phloem. A row of cells beneath the primary phloem fibres and outermost part of the secondary phloem elongates quite considerably in radial plane forming a continuous cylinder of narrowly oblong phellogen initials (Fig.4).

Each initial cell undergoes two periclinal divisions forming a central, transversely oriented narrowly oblong cell flanked on the outer and inner sides by fairly large cubical cells (Fig. 5). The central narrow oblong cells with dense cytoplasmic contents function as the phellogen. The outer cubical cells are the first phellem cell and inner one is the first phelloderm cell. By this time, more sclerenchymatic cells differentiate on the outer part of the periderm filling the gaps of the original fibre groups. Inner to the periderm also a discontinuous layer of sclereids differentiates. The sclereids are circular in transectional view and wide lumened (Fig. 6). During the further course of activity of the phellogen, it produces only phellem and no phelloderm. Due to increase in the thickness of the periderm, the sclerenchymatous cells outside the periderm as well as the parenchymatous cortex undergo progressive shrinking and collapse. Later, the entire cortex along with the sclerenchyma cylinder gets severed from the periderm and begins to peel off as a membrane (Fig. 6).

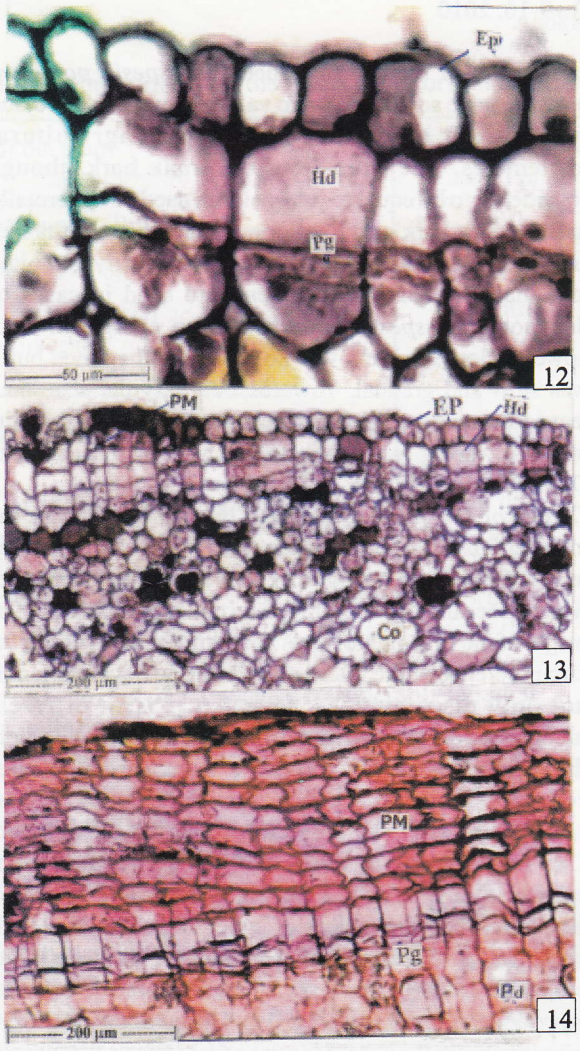
3. *Putranjiva roxburghii* Wall (Euphorbiaceae)- *Putranjiva roxburghii* Wall. is distributed all over India from the sub-Himalayan tracts through peninsular India down to Ceylon. It is fairly common in parts of the Western Ghats, but nowhere abundant. It usually occurs on riverbanks and is often cultivated as an avenue tree for its graceful appearance. "Pootranjeeva" is a Sanskrit name (Pootra = son; Jeeva = life), which signifies a popular belief that the children, who wear the nuts of the plant as rosaries and chains, are protected from evil eye. *Putranjiva roxburghii* reaches upto a height of 25m with an erect straight trunk. The leaves are used as a medicine for cold and as fodder. The wood is fairly hard and is used for turning. The tree is an evergreen plant shedding their leaves only on aging. The peak flowering season is in March and

April.

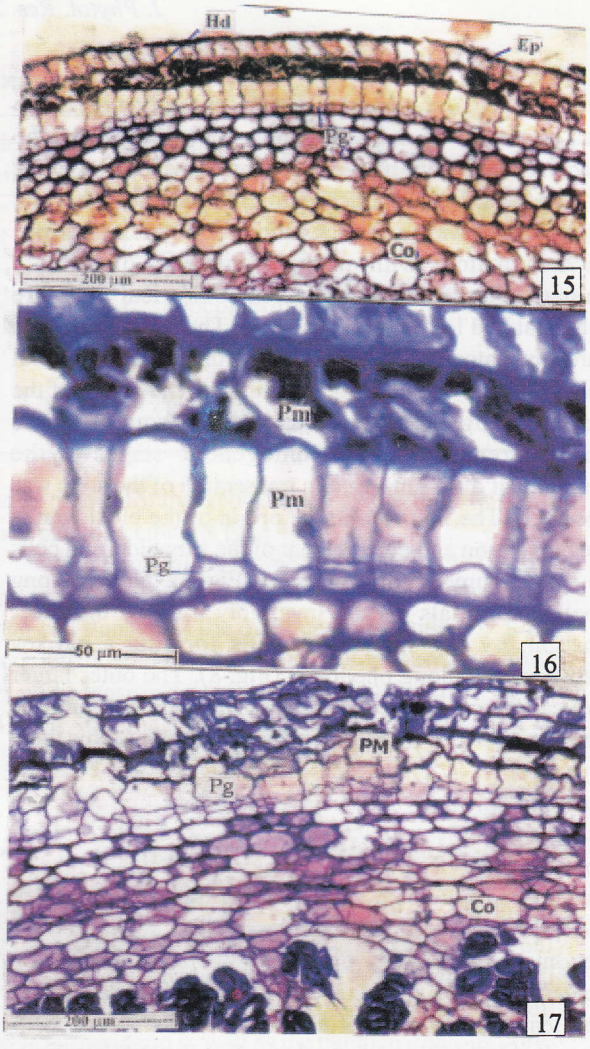
*Origin of the first periderm*- Origin of the first periderm was observed in the 10th internode onwards in the branches. Before the phellogen becomes evident the young internode has hemispherical epidermal cells with thin and straight radial walls and thick cuticle. The cortex is narrow, parenchymatous with an inner boundary of primary phloem sclerenchymatic elements. Cortical cells are spherical and less compact (Fig. 7). The sub - epidermal parenchymatous cells, not different from adjacent cortical cells, function as the phellogen initials. The origin of the first phellogen is not continuous throughout the circumference. Different ontogenetic stages of the periderm at different loci of a transection of the stem was observed. The first step in the origin of the phellogen is the elongation and enlargement of the subepidermal cells so as to assume an oblong shape (Fig. 8). These oblong phellogen initials divide, invariably towards the inner portion, by two tangential walls forming a narrow transversely oblong phellogen (Fig. 8). The outer larger cell can be considered as the first phellem cell and the inner smaller cell as the first phelloderm cell. At this three layered stage of the periderm, the spherical, less compact cortical parenchyma cells stretch tangentially, not accompanied by radial divisions (Fig.9). During further course of activity of the phellogen, it produces four or five layers of phellem whose cells still retain the nuclei; the cell walls are uniformly thick and the tangential walls become concave, the concavity facing inward. The epidermis breaks at several places and even small fissures and lenticels are also formed. On examination of thick branches, it was found that only the first formed superficial phellogen continues to function and no sequent deeper phellogen was evident. The phellem cells of the mature branch are transversely oblong, homogeneous in cellular composition and occur in regular radial files. The tangential walls of the cells are thicker due to more deposition of suberin on their outer facets (Fig.9).

4. *Thespesia populnea* (L). Soland ex Correa (Malvaceae)- *Thespesia populnea* (L.) Soland, ex Correa is one of the indigenous elements of South India, growing both wild as well as planted for ornamental and avenue purposes. It is more common along the seacoast, distributed Konkan Southwards, Burma and Westwards of Asia. In South India, especially in Tamil Nadu, it is well flourishing under cultivation and represents a typical tropical element. The Greek generic epithet 'Thespesia' means 'divine' (Since it was first noticed in temples) and the species name is derived from 'Poplar' like leaves. The plant is a medium sized tree of 8-12 m in height with a clear bole of 2-3 m and a girth of more than a meter. *Thespesia populnea* seems to have some economic value. Its wood is hard and fine-grained used for domestic and agricultural implements. The fibre from the trunk bark, though considered to be equal to jute, seldom used commercially in India. The bark contains appreciable amount of tannin and fine red matter, which are not commercially exploited in India. The bark is astringent and used in cases of diarrhoea. *Thespesia populnea* is an evergreen tree; however, leaf shedding was noticed not all at once but it was slow and gradual. Towards February the senescent leaves become yellowish which gradually fall from the tree. Flowering is throughout the year with a peak in December and January.

*Origin of the first periderm*- On examination of the transections of the young internodes it was found that the onset of the first phellogen and its subsequent activities were discernible in the third internode downwards. The origin of the first phellogen is not continuous all along the circumference of the internode; Periderm development is not only discontinuous, but also it is in different ontogenetic stages seen round axis. In the internode where the periderm is likely to originate, an epidermal layer of cells is followed by homogeneous, thin walled, less compact parenchymatous cortex can be recognized. Druses, tannin and mucilage cavities are frequent in the cortical tissue. Tanniniferous cells are more abundant especially in a few layers of subepidermal cells of the cortex (Fig.10). Some of the inner cortical cells get collapsed forming thin dark streaks before the onset of any periderm is evident (Fig. 11). Origin of the phellogen is invariably in the subepidermal cells. These cells initially assume radially oblong shape (Fig.11, 12). The hypertrophied subepidermal cells divide unequally by a tangential wall resulting in the formation of larger outer and smaller inner cells (Fig.12). In most of the cases examined, the larger outer cell divides by two successive tangential walls forming a central transverse, narrowly oblong cell, flanked externally and internally by comparatively larger cells (Fig.12, 13). The central cell functions as the phellogen while the outer cell functions as the phellem cell and the inner cell as the one and only phellodermal cell. Further development of the periderm is as usual as in other cases. The phellogen produces series of radially filed phellem cells (Fig.13, 14). On examining the internode through its circumference, different developmental stages of periderm from 2 to several cells thick zone can be seen in a single section. The epidermis remains intact till the periderm reaches 7-10 cells layers. When the total thickness of the periderm is about 200mm




*Thespesia populnea*




*Wrightia tinctoria*

When the total thickness of the periderm is about 200mm with more than 20 layers of phellem cells, the epidermis breaks uniformly all round the axis and gets crushed along with some of the underlying old phellem cells. The phellem cells of the branch periderm have uniform wall thickness with respect to radial and tangential walls which may be straight or sinuous (Fig.14) and suberised.

5. *Wrightia tinctoria* (Roxb.) R.Br. (Apocynaceae)- *Wrightia tinctoria* (Roxb.) R.Br. is a small tree growing 9-15m high with a bole girth of about 1.5m. It is found in scrub jungles and hill slopes. The tree is both wild and planted in gardens. The economic importance of the plant is not much known except that its wood is used for making agricultural implements. *Wrightia tinctoria* is a deciduous tree. In Madras climates leaf withering starts in February, and in March the trees are totally defoliated. New leaves appear in April followed by development of flower buds. The trees are in full foliage and flowering for several months. *Origin of the first periderm*- The phellogen is initiated very early in young branches even in the fourth internode. Phellogen originates simultaneously throughout the perimeter of the axis, but with some difference in the rate of activity of the meristem. The origin is from the cortical cells and the consistency in the pattern of the phellogen activity is strikingly uniform. The epidermal cells are oblong with fairly thick cuticle and wavy radial walls. The epidermis is followed by a few layers of collenchymatous

epidermis can be distinguished as the hypodermis. This layer elongates in radial plane considerably assuming oblong shape. The cells divide by periclinal walls forming smaller outer cubical cell layers and an inner layer of rectangular cells. The cells of the outer layer accumulate darker contents, presumably tannin (Fig.15). Now the outer zone of the cortex may have three cell layers barring the epidermis.

Of these, the innermost radially oblong cell layer divides by periclinal walls forming inner narrow transversely oblong cell layer which function as the phellogen. The slightly larger outer layer can be considered as the first phellem layer (Fig.16), which does not under go any further ontogenetic changes. The phellogen remains persistent and produces several layers of Phellem outside. The cells of the phellem are radially elongated and oblong in shape, with straight tangential walls and wavy radial walls. Shallow fissures and lenticels are frequently developed. The single phellogen persists with no evident development of sequent periderms and rhytidome.

**Results and Discussion**

Two primary components of bark are the periderm/ rhytidome and Secondary phloem. The former is often designated as outer bark and the latter as inner bark[7]. The outer bark is distinct from the inner one not only by virtue of its independent origin from a separate meristem, the phellogen, but also by its dead tissues with specific structural and chemical compositions. The inner bark is a composite tissue system consisting of diverse cell types differing from each other in their mode of origin, organization and general histology, best suited for the functions they are to perform. The outer and inner barks though stand apart from each other structurally, ontogenetically and even functionally, interact with each other to impart characteristic surface configuration and fissuring patterns to the bark[8].

Albeit variations in external and internal characteristics of mature barks are of considerable significance in discussion of systematic and phylogenetic problems, the position in which the first periderm originates is also taxonomically significant, because of its consistency even at species level[1]. This proposition is further strengthened by observations made during the present investigations. In *Putranjiva roxburghii*, *Thespesia populnea* and *Wrightia tinctoria* the first phellogen originates from the subepidermal layer of the cortex (Fig. 8, 12, 15). In *Aegle marmelos* it was from the third/fourth layer of the cortex (Fig. 2). In *Lagerstroemia reginae* the phellogen is initiated from the pericycle (Fig. 4).

Notwithstanding the consistency in the place of origin of the initial phellogen at species level, much variation was observed among different species of a family in the vertical distance from the apex to the locus of origin of the phellogen in an internode. No correlation was evident between the vertical distance and depth of cells layers acting as phellogen initials. The internodal region, where the first phellogen initiation was spotted varied from third to tenth in all the species.

Lev-Yadun and Lipschitz[9], after studying the phellogen initiation in the needle bearing and scale bearing conifers, concluded that in needle bearing conifers the first phellogen initiation was superficial in origin, a short distance from the apex. They attributed this to the fact that the needles were the main photosynthetic organs, which were almost independent of the periderm development. In scale bearing conifers, photosynthesis took place in small sprays and early periderm development of the sprays would surely reduce the photosynthesis. Thus, the poor photosynthetic efficiency of the scale leaves was correlated to late and deeper origin of the phellogen in small sprays, which compensated the photosynthetic process of the scale leaves. In needle bearing conifers, the sprays developed early periderm, and the periderm was superficial in origin since they were not involved in the photosynthesis, because of the presence of photosynthetic needles. The concept of correlating delayed and deeper or early and superficial periderm development with scale leaves and needle leaves respectively points out the indirect relationship between periderm formation and photosynthetic process in young shoots. When a shoot has efficient and enough photosynthetic units, then the young chlorenchymatous cortical tissues of the internodes are free to develop early and superficial periderm and suberization of the phellem. This explanation appears to be valid in general and applicable to other plants. Deciduous behaviour of certain plants may perhaps play a factorial role in determining the time and position of the first periderm development. Because in deciduous plants leaf shedding is naturally compensated by the chlorenchymatous cortical tissues of young sprays[10]. So, delay in suberization of the internodes in such plants seems to be an obvious adaptive strategy as exemplified in *Aegle marmelos*. The fact that origin of first phellogen in the third/fourth internodes in the deciduous trees, such as *Lagerstroemia reginae, Thespesia populnea* and *Wrightia tinctoria* and from the tenth internode in evergreen tree of *Putranjiva roxburghii* seems to be not congruous with the afore said tenet.

Origin of phellogen is not generally synchronous over the entire circumference of the stem in all species

studied, excepting *Lagerstroemia*. Most of the previous investigators also reported similar observations. Sanio[11] in his early studies on the phellogen development in many dicotyledons noted discontinuous development of periderm round the stem. Lier[12] says that this situation arises from the fact that there are varying factors at different parts of the circumference, which regulate the frequency of cell divisions of cork cambium. The discontinuous development of the periderm round the internode can be explained by the concept that localized phellogen activity prevents the neighbouring cells to assume meristematic activity and to develop into phellogen, i.e.; developing zone of phellogen may inhibit the initiation of other phellogen regions nearby. This explanation is based upon the "field theory" of inhibitory hormones or some other factors around pre-existing growth centers.

In *Lagerstroemia reginae* and *Wrightia tinctoria*, both origin and further activity of the phellogen are strikingly simultaneous throughout the circumference of the stem. Schoute's 'Field theory' of concentration gradients of inhibitory hormones around the pre-existing meristamatic centers does not seem to be tenable in explaining the situation in *L.reginae* and *W.tinctoria*. It can be only presumed that either such 'inhibiting gradients' do not exist in these plants, or some other factors may operate to nullify such hormone gradients

**References**


1. van Wyk A E, Botha D J and Coetzee J 1980, The Genus *Eugenia* L.
2. Arzee T, Liphschitz N and Waisel Y 1968, The origin and development of the phellogen in *Robinia pseudacacia L. New Phytol* **67** 87 - 93
3. Ahmad, Z, Khan M I H and Begum H 1969, Periderm in Some Perennial Plants. In: *Recent Advances in the Anatomy of Tropical Seed Plants. (Ed.)* K.A. Choudhury, Hindustan Publishing Corp. India. PP.75-83
4. Khan M I H and Ghouse A K M 1978, The origin and development of phellogen in *Psidium guajava* L. *Geobios* **5** 224-225.
5. Ghouse A K M and Yunus M 1975, Intrusive growth in the phloem of *Dalbergia*. *Bull. Torrey Bot. Club.* **102** 14 – 17
6. O'Brien T P, Feder N and Mc Cull M E 1964, Polychromatic staining of plant cell walls by toluidine blue o. *Protoplasma* **59** 367 - 373.
7. Trockenbrodt M 1990, Survey and discussion of the terminology used in bark anatomy. *IAWA. Bull.* **11** 141 - 166.
8. Whitmore T C 1962, Studies in Systematic bark morphology.
   I. Bark morphology in Dipterocarpaceae. *New Phytol.* **61** 191 - 207.
   II General features of bark construction in Dipterocarpaceae. *ibid* 208 - 220.
   III Bark Taxonomy in Dipterocarpaceae. Gdns. Bull. **19** 321 - 371.1963
9. Lev-Yadun S and Liphschitz N 1989, Sites of first phellogen initiation in Conifers. *IAWA Bull* **10** 43 - 52.
10. Kozlowski T T 1992, Carbohydrate sources and Sinks in Woody plants. *Bot.Rev.* **58** 107-222
11. Sanio C 1860, Vergleichende Untersuchungen Über den Bau und die Entwickelung des Korkes. *Jarhb. Wiss. Bot.* **2** 39 - 108.
12. Lier F G 1955, The origin and development of cork cambium cells in the stem of *Pelargonium hortorum*. *Amer. J. Bot.* **42** 929-936.