ROOT INITIATION IN THE ADULT AXES OF A FEW DICOTYLEDONOUS SPECIES

(With 14 Text-Figures)

BY AMIYA DATTA AND GIRIJA P. MAJUMDAR

(Botanical Laboratory, Presidency College, Calcutta)

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Introduction

ORIGIN of shoot borne adventive roots is described in text-books as *endogenous*, exceptions being recorded in the family of Cruciferæ, *e.g.*, in *Cardamine pratensis* (Hansen, 1881) and *Nasturtium austriacum* (= *Roripa austriaca*, Wilson, 1927). In both these cases the origin is described as *exogenous* and is reported to be associated with axillary buds (Priestley and Swingle, 1929).

In assigning the position of the endogenous root initials in an organ De Bary (1884) makes the general statement that it must be " in or close to vascular bundles or masses of wood or bast " (p. 315). Lemaire (1886) made extensive studies of the " origin of naturally occurring adventive roots in the hypocotyls, stolons and rhizomes " of herbaceous dicotyledons and observed that the origin of these roots might be (1) exclusively in the pericycle, (2) partly in the pericycle and partly in the endodermis and inner cortex, (3) exclusively in the " subphloem meristem ", *i.e.*, cambium, and (4) partly in the cambium and partly in the pericycle. He, however, concluded that the pericyclic origin is by far the commonest. Van Tieghem and Douliot (1888) fix the place of origin entirely in the pericycle so far as the young hypocotyls, epicotyls, stolons and rhizomes are concerned; if, however, they state, the pericycle loses its " root-forming property ", as in old stems, the adventive roots arise in the phloem parenchyma, both primary and secondary; and still later in cambium itself. Eames and MacDaniels (1925), on the other hand, give the position as the pericycle in the cases of young stems and in older axes, where the pericycle is no longer active, in the secondary phloem (p. 238).

Priestley and Swingle (1929) distinguished the shoot borne adventive roots into two categories on the basis of their origin, namely, (1) those formed behind the apical meristem and (2) those arising upon old stems which have

ceased to extend longitudinally, but in which radial growth alone is proceeding (p. 62). They then pointed out that in young stems these initials differentiate practically always near but to the side of a vascular group, *i.e.*, on a primary ray. In old stems on the other hand the site of initiation of a lateral root moves inwards from the region of the pericycle to the living cells of the ray that lie close to the newly differentiated xylem and phloem. The root initials are formed in association with a group of cells bordering upon the vascular cambium (pp. 63, 64).

We have undertaken this investigation in order to determine in a general way the origin of naturally occurring adventive roots on adult axes of herbaceous plants, and at the same time to re-examine the validity of the general statement of Priestley and Swingle with regard to the root initiation in old stems.

By "adult" or "old" axes we would mean the hypocotyl and internodes which have ceased to elongate longitudinally, but in which the radial extension is still going on, as contemplated by Priestley and Swingle in their second category of shoots bearing adventive roots.

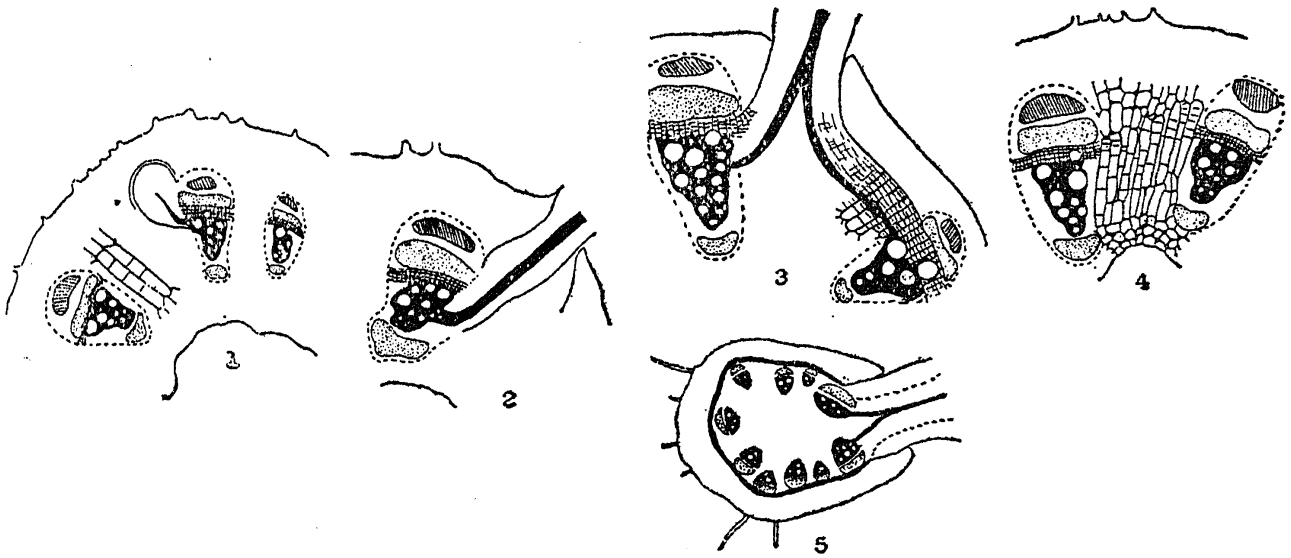
There is much controversy with regard to the use of the term 'pericycle' (Solereder, 1908; Strasburger, 1930). In this and subsequent papers we accept the definition of Van Tieghem and Douliot which runs as follows: The pericycle is "the layer or layers between the endodermis and the external phloem of a fibrovascular bundle continuous across the medullary rays but not distinguished on the inside from the medullary ray" (Carlson, p. 119, 1929).

We propose to report on the origin of naturally occurring lateral roots in the adult internodes of herbaceous dicotyledons, monocotyledons and of pteridophytes, and the present paper forms the first of the series that are expected to follow with the progress of our work. The results embodied in this paper are based on a study of free-hand sections of the hypocotyls or internodes of the species reported, and the text-figures have all been drawn under a microprojector.

Observation

Cucurbita maxima (Text-Figs. 1–4) develops a fairly elongated hypocotyl during its seedling stage of growth. The vascular bundles, ten in number, are typically bicollateral and are arranged in a ring with sclerenchymatous bundle caps characteristically associated with each of these ten bundles. These caps are very prominent in bundles near the base of the hypocotyl, but as one proceeds towards the top the cells of the cap lose their sclerenchymatous nature and do no longer give lignin reaction with aniline sulphate

or aniline chloride solution. The outer vascular cambium is much wider than the inner one, and extends laterally into a layer or two of the ray cells on the sides of each bicollateral bundle.



TEXT-FIGS. 1–5. Origin of shoot-borne lateral roots in *Cucurbita maxima* and *Oxalis corniculata*.—Figs. 1–4. *Cucurbita maxima*. T.S. of hypocotyl showing the origin of adventive roots. Fig. 4 shows the nature of the radial growth of the hypocotyl. Fig. 5. T.S. of the adult internode of *Oxalis corniculata*. ×15.

The vascular bundles in the hypocotyl thus differ from those in the stem in their arrangement round the pith, and in having isolated sclerenchymatous bundle-caps of procambial origin instead of a continuous sclerenchymatous cylinder of the adult internodes described as pericyclic in position.

Lateral roots begin to appear near the basal region of the hypocotyl which has already ceased to elongate, and proceeds to a certain distance up the organ. The root initials take their origin in the flanks of the outer cambium in the formation of which the adjoining ray cells also take part (Figs. 1 and 2). There may be two adventitious roots originating from the two sides of a vascular bundle or rarely two groups of initials from the flanks of the adjoining bundles join together, sometimes in the middle, sometimes to one side, of the intervening ray to form a single lateral root (Fig. 3). The root initials after their organization into the primordium bend through about 90° in their passage through the cortex towards the periphery of the hypocotyl without causing any or causing a very slight disturbance to the bundle-cap which retains its original position with reference to the bundle.

When the lateral roots begin to appear the hypocotyl in this region has already ceased to elongate, but the radial growth is still maintained. This is accomplished in two ways, namely, (1) by the radial growth of individual

bundles from the two fascicular cambia, particularly from the outer one, and (2) by the enormous radial elongation of ray cells which divide tangentially with a view, it appears, more to resist lateral pressure than for any other purpose (Fig. 4). Sometimes regular interfascicular cambia are organised, but they do not unite with the outer fascicular cambia to form a continuous cambial ring as in the normal axes of dicotyledons. These interfasicular cambia help the radial extension of the ray cells and also very frequently form isolated phloem patches in the rays.

Oxalis corniculata (Text-Fig. 5) is a creeping plant. Adventitious roots from adult internodes are rare in this plant. The few that have been found were given off from the lower side of the trailing shoots growing in moist shady places. There are ten bundles arranged in a ring and at no time a regular cambial ring has been seen to form though the growth and extension of the bundles, particularly of the bigger ones, is maintained by the fascicular cambia. The endodermis with casparian strips is found to persist, and a complete cylinder of thick-walled pericycle of varying depth is a special feature of the internodal structure of this plant.

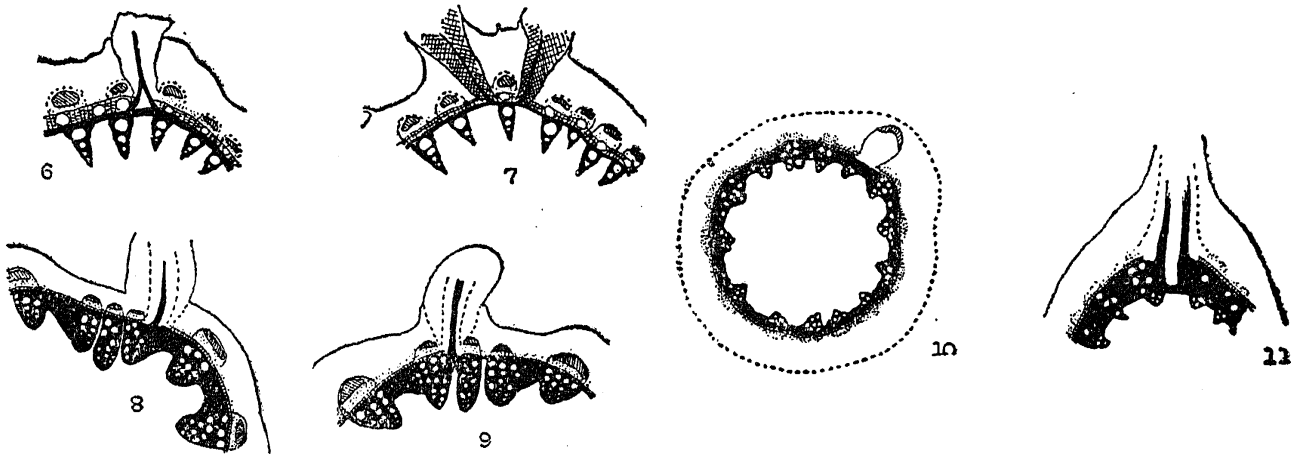
In the formation of the shoot-borne roots two groups of initials from the flanks of the adjoining vascular bundles join together in the middle of the intervening ray to form a single lateral root (Fig. 5). Vascular connection is later made with both the bundles. The root initials originate, as in *Cucurbita*, in the cambial and ray cells adjoining it.

Mikania scandens (Text-Figs. 6, 7) a twining and spreading herb which delight in moist places, particularly near the edges of a pool. Lateral roots from internodes of adult stems are very rare in this plant. The detailed developmental and adult anatomy of this plant is being worked out by Mr. I. Banerji of the Department of Botany, Calcutta University, and we propose to record here only the barest outline of the adult stem structure.

The large number of vascular bundles are surmounted each by a sclerenchymatous bundle-cap. Interfascicular cambium rarely produce any xylem vessels. In the formation of the root initials both the vascular and interfascicular cambia take part. In Fig. 6 the lateral root is median in position with reference to the medullary ray and vascular connection is made with both the bundles. In Fig. 7 two lateral roots are formed one on each side of a vascular bundle.

Tagetes patula (Text-Figs. 8, 9), an annual garden herb, but may be made to continue through many seasons if proper care is taken. It is towards the end of the first season that lateral roots, sometimes in profuse numbers, are given out from adult internodes. The origin of these roots are definitely

related to the trace bundles as their vertical arrangement can be followed along the course of these bundles in the internodes. Good varieties of *Tagetes* is propagated mainly by cuttings.



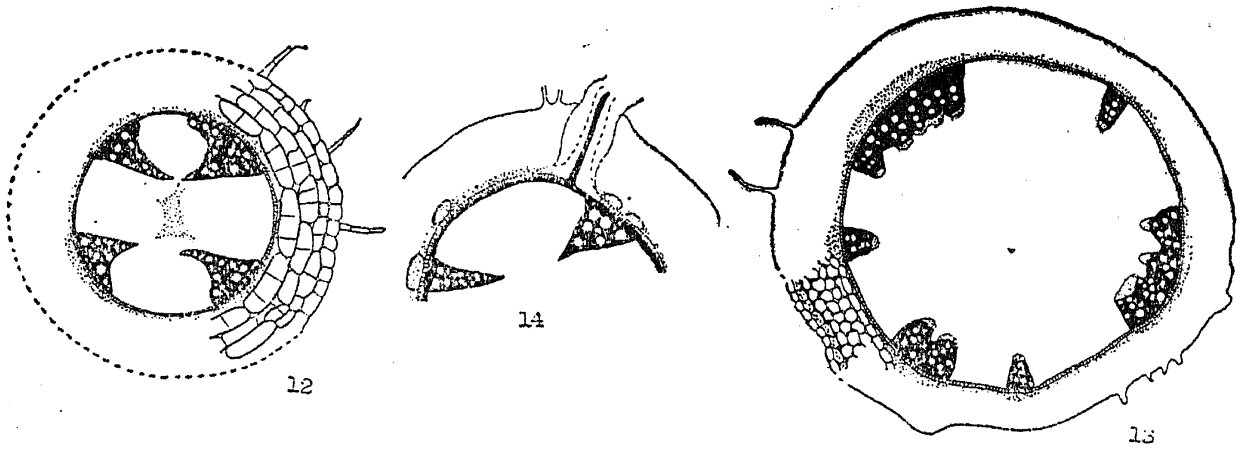
TEXT-FIGS. 6–11. Origin of shoot-borne lateral roots.—Figs. 6–7. T.S. of adult internodes of *Mikania scandens*. Figs. 8–9, that of *Tagetes patula* and Figs. 10–11, that of *Ageratum conyzoides*. ×15.

The arrangement and structure of the vascular bundles in adult internodes are typically of the *Helianthus* type. The lateral roots are given out from the flanks of the fascicular cambium of a vascular bundle. Fig. 8 shows the origin of a lateral root from the flank of a synthetic bundle in the making. The bundle-cap is not disturbed. Fig. 9 shows a root developed between two vascular bundles which has made vascular connections with both of them.

Ageratum conyzoides (Text-Figs. 10, 11).—This is also an annual herb but erect in habit. In all the specimens collected the production of lateral roots were found to be confined to the first three or four internodes growing prostrate on the ground. The anatomical structure of the axis is typically that of the woody vine type, *i.e.*, an unbroken cylinder of primary and secondary wood with no medullary rays. The lateral root arises from the flank of a vascular canbium (Fig. 10) and when it makes vascular connection with two bundles it is seen to occupy the whole of the intervening medullary ray outside the interfascicular cambium (Fig. 11).

Solanum nigrum (Text-Figs. 12–14).—This is an annual herb. Bhaduri (1933) points out that " plants identified as *S. nigrum* by Prain (1903), Hooker (1875) and others differ considerably in both morphological and cytological characters " (p. 58), and he could easily distinguish three types of *S. nigrum* from his critical studies of cytology and morphology of these plants ($n = 12$, $n = 24$, $n = 36$; pp. 60–61). We have not been able to collect, examine and compare the anatomical structure of all the three types

of Bhaduri, but Figs. 12 and 13 show that the internode and hypocotyl of this plant differ widely in their anatomical structures.



TEXT-FIGS. 12–14. Origin of lateral roots and the structure of internodes and hypocotyl in T.S.—Figs. 12 and 13. T.S. of the hypocotyl and internode respectively of *Solanum nigrum*. Fig. 14. T.S. of hypocotyl showing origin of lateral root. Figs. 12 and 14, ×15 ; Fig. 13, ×25.

Fig. 14 shows the origin of a lateral root in the hypocotyl of *S. nigrum*. It will be seen that there are only 4 primary bundles and a cambium ring is very early differentiated. The root initials originate in the interfascicular and fascicular cambia close to a vascular bundle, and the vascular connection is made with the secondary xylem and phloem.

Discussion

Adventive roots may arise on any part of a stem, but the normal position is the node. The present study as has been mentioned in the Introduction is confined to the origin of naturally occurring roots in the nodeless segments of adult shoots of a few dicotyledonous herbs ; and in this respect our observations differ from those of others dealing mostly in the regeneration of roots on stem and leaf cuttings (Swingle, 1940).

From the two reported cases it may be assumed that exogenous adventitious roots are developed only in association with axillary buds. In *Cotoneaster Dammeri* development of lateral roots in association with axillary buds is a normal feature. Miss Wolfe (1934) noted their origin in the parenchyma at the flank of the cambium of the branch (bud) trace. It is doubtful if one can call this an exogenous origin.

Lemaire and Van Tieghem and Douliot studied the origin of naturally occurring roots on hypocotyl, rhizomes and internodes, but they do not, so far as we are aware, appear to have noted any difference in the origin of lateral roots in young and adult organs. Priestley and Swingle are perhaps the first to note this difference. They formulated their second type of origin from a study of the hypocotyl in *Helianthus* and *Ricinus*, and of the

epicotyl of *Vicia* and *Solanum*. The six cases reported here are additional examples.

Trécul (1846) suggested the possibility of the existence of preformed root initials in stems of certain species. 32 years later Vöchting (1878) reported regular occurrence of preformed root premordia in the cuttings of Willow. Goebel (Vol. II, p. 275) reports the presence of pre-existing sub-cortical "root germs" in *Salix*, and these primordia are found to develop on the cuttings used for vegetative propagation in these plants. Van der Lek (1924) noticed occurrence of such preformed root germs in young branches of *Salix*, *Populus* and *Ribes nigrum* closely associated with cambium and the end of medullary rays at the outer edge of xylem. Pre-existing "root germs" have also been noticed by Swingle (1925) in apple stems and by Sandison (1934) in *Lonicera japonica*. Though *Tagetes* is often propagated by cuttings in this country we have so far failed to discover the presence of preformed root germs in this plant or in other plants examined by us. Preformed root-primordia have not also been found in *Salix caprea*, *S. aurita*, *Populus alba* and *Vitis vinifera* (Carlson, 1938).

In all the cases studied by us the site of origin is shifted to the flanks of a vascular bundle close to *cambium*, *interfascicular cambium* or *ray cells* abutting these meristematic tissues. *Cambial origin* has been reported by Corbett (1895–96) in herbaceous stem cuttings of *Geranium*, *Coleus* and allied plants; by Smith (1925, 1928) in *Coleus* and *Clematis*, by Taylor (1926) in *Acanthus montanus*, by Sandison (1934) as a response to wounding in *Lonicera japonica*, and by Arlot Smith (1936) occasionally in *Begonia maculata* and *B. semperflorens*.

Origin in *interfascicular cambium* has previously been recorded by Regal (1876) in *Begonia maculata* (= *Begonia argyrostigma*); by Connard and Zimmerman (1931) in cuttings of *Portulaca oleracea* and by A. Smith in *Begonia maculata* and *B. semperflorens*.

Pericyclic origin has been observed in *Coleus Blumei* by Carlson (1929), in *Veronica Beccabunga* by Priestley and Swingle (1929); origin in the *secondary phloem* either within or between the bundles by Carlson (1933) in the cuttings of Dorothy Perkins rose; and in the *ray parenchyma* between a poorly defined pericyclic region and the interfascicular cambium by A. Smith (1942) in the cuttings of *Tropaeolum majus*. The case reported by Crooks (1934) is very interesting in so far that in cuttings of the upper part of the hypocotyl of Flax seedlings he noticed initiation of a root by the activity of the ray cells in the regions respectively of the pericycle, phloem and pith.

According to Priestley (1931) and Priestley and Swingle (1929) three conditions must be satisfied for the initiation of a root in an organ, namely, (1) presence of a meristem or potentially meristematic cells, (2) the mother tissue must be free from air spaces and (3) placed very near the xylem and phloem. The first is necessary because by the division of its elements new growing centres of roots are organised; close proximity to vascular supply ensures adequate supply of nutritive materials and absence of air space will permit a steady diffusion of solutes (pp. 65, 66). The presence of a meristematic tissue, *i.e.*, cambium, renders grafting a practical process.

In roots the pericyclic origin of lateral roots is obviously an advantage, but in adult hypocotyl and internodes the bundles are collateral with phloem and in many cases with additional sclerenchymatous bundle-caps on the outside. Therefore to effect direct xylem connection the origin must be close to xylem in the flanks of a fascicular cambium. Intercellular spaces are already formed throughout the radial course of ray cells, the only region free from such intercellular spaces being the cambium, interfascicular cambium and the ray cells recently formed in the neighbourhood of cambial ends. The ray cells are relatively smaller and are filled with active protoplasm. When all these facts are taken into consideration the ends of the cambium, the outer region of the interfascicular cambium and the living cells just cut off from the meristematic tissue and the proximity of the newly differentiated xylem and phloem are the ideal places for the origin of a lateral root upon a radially expanding nodeless axis.

The radial growth of the hypocotyl of *Cucurbita maxima* is very interesting, but this kind of growth in thickness has already been reported in the stems of *Coccinia* (= Cephalandra), *Trichosanthes*, *Wilbrandia*, *Anisosperma* and *Alsomitra* (Potter, Schenk, Herail, in Solerder I, p. 395). In all these cases, as in the hypocotyl of *C. maxima*, the interfascicular cambia help primary rays in keeping pace with the radial expansion of the bundles.

Summary

Six additional cases of naturally occurring shoot-borne adventive roots on adult axes of herbaceous dicotyledons have been described in this paper.

These support the view of Priestley and Swingle that the site of initiation of a lateral root in adult hypocotyls and internodes moves inwards from the pericycle to the flanks of the vascular cambium close to the newly differentiated xylem and phloem.

The radial growth of the hypocotyl of *Cucurbita maxima* is maintained by the independent growth of the vascular bundles, the interfascicular

cambium helping the primary rays in keeping pace with the growth in thickness of the former.

LITERATURE

1. Bhaduri, P. N. .. "Chromose numbers of some Solanaceous plants of Bengal," *Jour. Ind. Bot. Soc.*, 1933, **12**, 56–64.
2. Borthwick, A. W. .. "The production of adventitious roots and their relation to Birds-eye formation in the wood of various trees," *Notes Roy. Bot. Gard.*, 1905, Edin. **4**, 15–36.
3. Carlson, M. C. .. "Origin of adventitious roots in *Coleus* cuttings," *Bot. Gaz.*, 1929, **87**, 119–27.
4. ———— .. "Origin and development of roots in cuttings of Rose and Willow," *Contrib. Boyce Thompson Inst.*, 1933, **5**, 313–30.
5. ———— .. "The formation of nodal adventitious roots in *Salix cordata*," *Amer. Jour. Bot.*, 1938, **25**, 721–25.
6. Clark, W. A. .. "Vegetative propagation in Cotoneaster," *Trans. & Proc. Bot. Soc.*, Edin. 1932, **31** (2), 256–61.
7. Connard, Mary H., and P. W. Zimmerman "The origin of adventitious roots in cuttings of *Portuloca oleracea* L.," *Contrib., Boyce Thompson Inst.* 1931, **3**, 337–46.
8. Corbett, L. C. .. "The development of roots from cuttings," *W. Va. Agric. Expt. Stn., Ann. Rep.*, 1895–96, **9**, 196–99 (A. I. Smith, 1936).
9. Crooks, D. M. .. "Histological and Regeneration studies on the Flax seedlings," *Bot. Gaz.*, 1934, **95**, 209–39.
10. Eames and MacDaniels .. *Introduction to Plant Anatomy*, 1925.
11. Goebel, K. .. *Organography of Plants*, Part II, 1905, p. 274, Eng. Ed. (quotes Van Tieghem and Douliot).
12. Jimenez, P. G. .. "Callus and root-formation in stem cuttings of Kapok, Achuete and Santol," *Philippine Agr.*, 1937, **26**, 585–635.
13. Van der Lek, H. A. A. .. *Over die wortelvorming van houtige stekken*, H. Veeman and Zonen, Wageningen, 1924–1925 (Carlson, 1929).
14. Lemaire, A. .. "Recherches sur l'origine et le development des racines laterales Chez les Dicotyledones," *Ann. Sci. Nat. Bot.*, Ser. VII, 1886, **3**, 163–274 (Carlson, 1929).
15. Prain, D. .. *Bengal Plants*, 2 vols., 1903.
16. Priestley, J. H. .. "Vegetative propagation from the standpoint of Plant Anatomy," Reprint, *Proc. 9th Int. Hort. Congress*, 1931.
17. ———— .. "Problems of Vegetative Propagation," *Jour. Roy. Hort. Soc.*, 1926, **51**, 1–16.
18. ——— and C. F. Swingle "Vegetative propagation from the standpoint of Plant Anatomy," *Technical Bulletin U.S. Dept. of Agriculture*, 1929, No. 151.
19. Regel, F. .. "Die vermehrung der Bigoniaceen aus ihren Blättern," *Jena, Zeitschr. Med. Naturwiss* (N.F. 3), 1876, **10**, 447–92 (A. I. Smith, 1936).
20. Sandison, S. .. "Rooting of cuttings of *Lonicera japonica*, a preliminary account," *New Phyt.*, 1934, **33**, 211–17.

21. Smith, A. I. .. "Adventitious roots in stem cuttings of *Begonia maculata* and *B. semperflorens*," *Amer. Jour. Bot.*, 1936, **23**, 511–15.

22. ———— .. "Adventitious roots in stem cuttings of *Tropæolum majus* L.," *ibid.*, 1942, **29**, 193–94.

23. Smith, E. P. .. "A comparative study of the stem structure of the genus *Clematis* with special reference to anatomical changes induced by vegetative propagation," *Trans. Roy. Soc.*, Edin., 1928, **55**, 643–44.

24. ———— .. "The origin of adventitious growth in *Cotcus*," *Trans. & Proc. Bot. Soc.*, Edin., 1925 (1936), **29**, 145–50.

25. Swingle, C. F. .. "Regeneration and Vegetative Propagation," *Bot. Rev.*, 1940, **6**, 301–55.

26. ———— .. "Burrknot formations in relation to the vascular system of the apple stem," *Jour. Agri. Res.*, 1925, **34**, 533–44.

27. Solereder .. *Systematic Anatomy of the Dicotyledons*, Eng. Ed., 2 vols., 1908.

28. Strasburger, E. .. *Text-Book of Botany*, Eng. Ed., 1930.

29. Taylor, G. .. "The origin of adventitious growth in *Acanthus montanus*," *Trans. Proc. Bot. Soc.* Edn., 1926, **29**, 291–96.

30. Van Tieghem, P., and H. Douliot "Recherches comparatives sur l'origine des membres endogenes dans les plantes vasculaires," *Ann. Sci. Nat. Bot.*, 1888, Sér. VII, **8**, 1–660 (Carlson, 1929).

31. Trécul, A. .. "Recherches sur l'origine des racenes," *ibid.*, 1846, Sér. III, **5**, 340–44 (Carlson, 1938).

32. Vöchting, H. .. *Über Organbildung im Pflanzenreich*, Theil 1, Bonn. (Carlson, 1938).

33. Wilson, C. L. .. "Adventitious roots and shoots in an introduced weed," *Bull. Torr. Bot. Club*, 1927, **54**, 35–38.

34. Wolfe, F. H. .. "Origin of adventitious roots in *Cotoneaster Dammeri*," *Bot. Gaz.*, 1934, **95**, 686–94.