ROOT SECONDARY GROWTH IN THORN APPLE AND HOT PEPPER PLANT SPECIES AT DIFFERENT GROWTH STAGES

Arafa, A.A.
Dept. Botany, Fac. Agric., Mansoura Univ.

ABSTRACT

Two annual herbaceous species of *Solanaceae* namely, *Datura stramonium* L. and *Capsicum frutescens* L. were subjected to study the incidence of the secondary growth and consequents the formation of protective tissues in their roots. The main results were as follows :

1- At an early stage of growth, the xylem tissue at the base of the root accomplished a proportion of its final amount that was decidedly bigger than that needed for the shoot system already achieved by the plant. It appeared therefore that certain amount of water conducting elements should be developed prior to the successful start and continuation active vegetative growth.

2- The amount of secondary growth varied in roots of the two investigated species; it was clear that the bigger the amount the more readiness of the root to form a periderm-like structure until a true periderm was developed with the biggest amount.

3- The periderm-like structure appeared in the exodermis or the underlying cortical layers according to the species, while the true periderm developed normally in the pericycle.

4- The endodermis and pericycle may behave alike or unlike as to their response to secondary growth.

INTRODUCTION

From the economical point of view, the numbers of the *Solanaceae* are of great importance and yield foods, drugs and ornamentals (Hutchinson, 1973 and Datta, 1988).

The fruits of *Capsicum frutescens* are used as a condiment. A number of species are used as vegetative, e.g. *Solanum melongena* and *Solanum lycopersicum*. Some species are of medicinal value. Alkaloids are derived from *Datura stramonium* (Pandey, 2003).

From the histological point of view the available information about the development of the protective tissues in the roots of the annual herbaceous dicotyledons is not sufficient. In this regard, some scientists reported that the development of protective tissues in the roots is restricted to the perennial plant species where a true periderm arises in the pericycle (Eames and Mc Daniels, 1947; Eames, 1961; Frohne and Jensen, 1998; Sieburth and Deyholos, 2006 and Barthelemy and Caraglio, 2007).

In addition, a variety of protective tissues were found in plants with a limited amount of secondary growth as shown in certain herbaceous dicotyledons : persisting thick-walled epidermis (*Ranunculaceae*); exodermis (*Primulaceae*); exogenously originating periderm (*Asteraceae*); dead and collapsed but persisting cortex (*Polygonaceae*); subdivided and suberized

endodermis (*Gentianaceae*); polydermal (*Potentilla*); periderm of deep seated origin (*Saxifragaceae*) as reported by Hayward (1951), Luhan (1955) and Bresinsky *et al.* (2008). The amount of secondary growth occurred in roots of the different herbaceous plant species varies as do the histology of tissues and periderm differentiation (Esau, 1965 and Metcalfe and Chalk, 1983).

However, Metcalfe and Chalk (1983) indicated that in roots of the solanaceous plant species, cork arises in the exodermis or may be deep-seated in origin.

Depending on the available information about the diversity in the manner by which the herbaceous dicotyledons build up the protective tissues throughout their secondary growth in addition to the scarce literature related to this parameter it was necessary to carry out the present work.

Therefore, the main aim of the present investigation was to study in some detail the response of the primary body and the histological tissue changes accompanying secondary growth of the roots of either *Datura stramonium* L. (Thorn Apple) or *Capsicum frutescens* L. (Hot Pepper).

MATERIALS AND METHODS

The current work was carried out at the experimental station, Faculty of Agriculture, Cairo University, Egypt.

Seed lots of the two investigated solanaceous species listed before i.e. *Datura stramonium* L. and *Capsicum frutescens* L were secured from the Agricultural Society, A.R.E. Seeds were sown on April 18. Forty pots 40 cm diameter filled with Nile Clay were used, 20 for each species. When plants were 22 days old, they were thinned out to 5 plants for each pot.

All plants were fertilized with a combination of NPK as 3:2:1 at a rate of 10 g/plot. This amount was equally applied at the two ages 25 and 50 days after sowing.

For the histological study at every studied age, three samples from digging out plants of the tap-root at about 2 cm below the hypocotyl in the root axis were killed and fixed in F.A.A. solution (Gerlach, 1977).

The first root samples were taken after emergence of dicotyledons above soil (one week). Then daily samples were taken to detect the start of cambial activity.

From three weeks onwards, the root samples were secured every fortnight until the age of 9 weeks and after every three weeks until end of the vegetative growth.

Fixed materials were dehydrated by normal butyl alcohol method and embedded in paraffin wax of 56-58 °C. m.p Sections 15 µ thick were cut by a rotary microtome, stained with crystal violet/erythrosine before mounting in Canada balsam (Nassar and EL-Sahhar, 1998). The stained sections were examined by light microscopy and photographed.

For the external measurements or counts, ten plants for every species from 10 plots were labelled and assigned for measuring the length of main stem or the plant height and counting the number of leaves per plant at weekly intervals, starting 3 weeks after sowing.

From the rest of plants 10 were pulled out one plant from each pot, to estimate the total leaf area by means of a planimeter and the dry weight of the shoot system starting 5 weeks after sowing in *Datura stramonium* L., since it showed an early rapid growth and after 7 weeks in *Capsicum frutescens* L.

The estimation was repeated for every species every 2 weeks until 9 weeks after sowing then every 3 weeks until end of vegetative growth. The plants were dried at 105 ^{o}C for 12 h.

RESULTS AND DISCUSSION

Datura stramonium, L.

Examining the histological structure of the root at the different successive ages proved that the behaviour of the primary body due to secondary growth i.e. the formation of a protective tissue and the dilatation growth of the outer tissues agrees in many respects with that described for both eggplant and tomato plant.

Secondary growth started early, 12 days after sowing (Fig. 1) resembling that in the tomato plant but earlier by two days than in the eggplant. This early start of secondary growth was followed in the Jimson weed (Datura) by a faster rate of cambial activity, resulting in a bigger xylem core at same or even younger age.
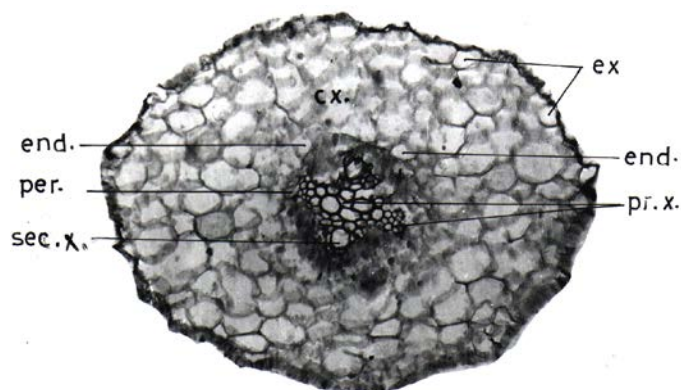

Fig. (1): Transection at the base of main root of *Datura stramonium*, 12-day old plant (X150).

ex.: Exodermis, end.: Endodermis, pr.x.: Primary xylem, Per.: Pericycle, sec.x.: Secondary xylem.

It was interesting to notice that the vascular cylinder of the root achieved about 62 % of its final xylem tissue by the end of the seventh week after sowing when only about 33 % of growth of the shoot system estimated as dry weight was accomplished. It appeared that by the end of 7 weeks, the amount of vascular tissues developed in the root was not proportional to the amount of serial vegetative growth. It appeared that by this early construction of a big

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percentage of the total amount of xylem tissue, the root became histologically ready to share in building up the still unformed grater portion of the vegetative shoot system.

In general, the topographical structure of the root at 3 weeks age was similar to both eggplant and tomato plant. The xylem diameter was 412 μ on the average.

Age of 5 weeks :

As stated before secondary growth produced a somewhat big amount of conducting tissues (Fig. 2), the xylem core reached an average diameter of 820.9 μ.
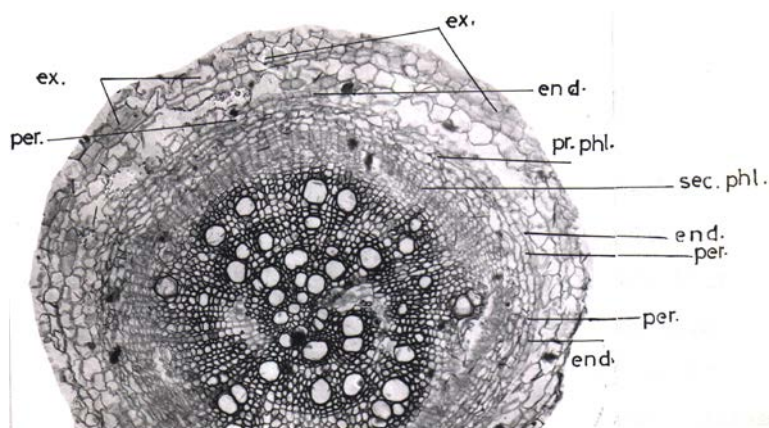

Fig. (2): Transection at the base of main root of *Datura stramonium*, 5-weeks old plant (X150).

ex.: Exodermis, end.: Endodermis, pr.phl.: Primary phloem, sec.phl.: Secondary phloem, Per.: Pericycle.

The exodermis lying next to the collapsed epidermis showed division of many of its cells especially in the periclinal plane. The underlying cortical layers had either enlarged cells or dilated ones to accommodate the increase in root girth. Thus, the cells were of varied size and shape. Both the endodermis and the pericycle had rather uniformly shaped and sized cells, in contrast with those of the cortical ones. It was difficult to distinguish between the component cells of these two adjacent tissues except by their orderly orientation and the presence of casparian strips in the endodermis.

Short duration of tangential elongation of cells followed by rapid cell division might be responsible for this cell uniformity which was noticed also for the endodermis in the tomato plant.

Age of 7 weeks :

Fig. (3) revealed that during the next two weeks, secondary growth proceeded actively bringing the xylem to an average diameter of 1417.9 μ which was about double that at the 5 weeks age. As a result of the continuous outward pressure, the exodermis showed also continuous division of its cells mainly by

tangential walls resulting at many places in the development of tiers of radially arranged cells assuming, therefore, a discontinuous cylinder of a periderm-like tissue. In this respect, the tendency of the exodermis to build-up a periderm-like tissue for protection of the inner tissues was more pronounced in *Datura stramonium* than in *Solanum melongena* while it was unnoticed in tomato plant. Dilatation of the cortical cells was still detected. The endodermis lost the uniformity of its cells due probably to unequal rate of tangential elongation or cell division at the different sites. The pericycle also was much altered since at many locations the cells were largely pressed and their lumina became too narrowed. Crystal sand was occasionally presented in the endodermal, pericyclic or phloem parenchyma cells, as well as in those of the cortex.
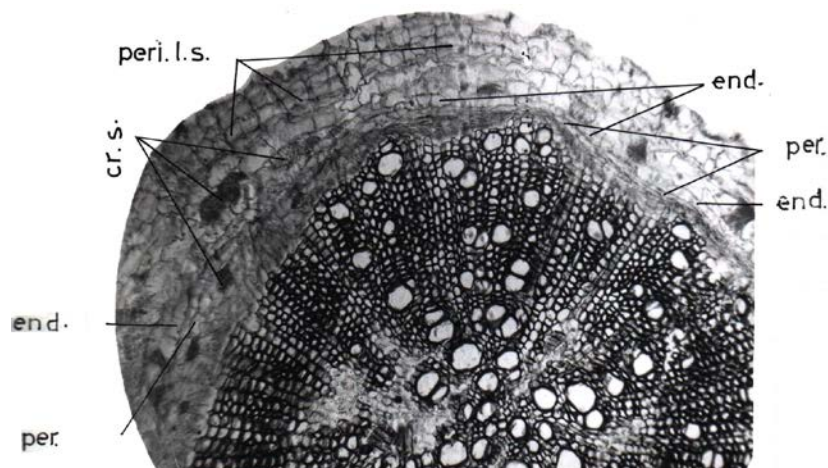

Fig. (3): Transection at the base of main root of *Datura stramonium*, 7-weeks old plant (X150).

end.: Endodermis, Per.: Pericycle, cr.s.: Crystal sand, peri.L.S.: Periderm-like structure.

Age of 9 weeks :

Except for the further growth of the vascular cylinder, the general topographical structure and histological features of the root remained unchanged by the end of 9 weeks after sowing (Fig. 4). The xylem diameter increased to a mean of 1904.9 μ evidently bigger than at same age in either the eggplant or the tomato plant, still indicating a higher rate of cambium division in *Datura stramonium*.
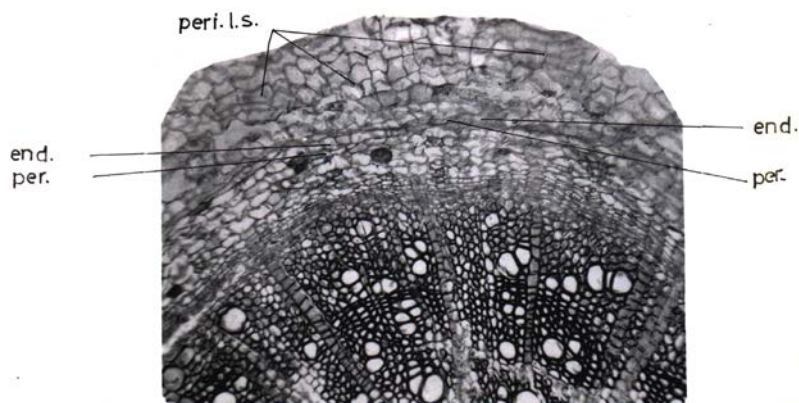
Fig. (4): **Transection at the base of main root of *Datura stramonium*, 9-weeks old plant (X150).**

end.: Endodermis, Per.: Pericycle, peri.L.S.: Periderm-like structure.

Age of 12 weeks :

By the end of vegetative growth at about 12 weeks after sowing (Fig. 5), the xylem diameter attained an average of 2303.3 µ. The superficial periderm-like structure varied in degree of distinction at the different locations. The distribution of the phloem conducting elements could be described as lying between that of *Solanum melongena* and *Solanum lycopersicum*, in other words between the scattered and regular distribution.
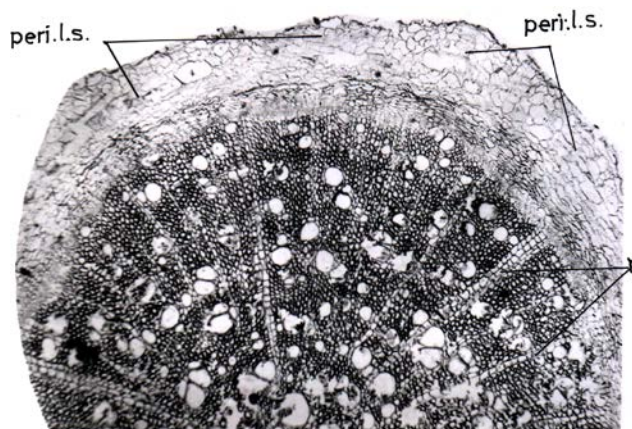

Fig. (5): Transection at the base of main root of *Datura stramonium*, 12-weeks old plant (X100).

peri.L.S.: Periderm-like structure, r.: Ray.

Xylem characteristics :

The ground tissue where the xylem vessels are embedded is mostly lignified parenchyma. The wider most vessels tend to gather around the root centre, indicating their early development as in the eggplant, while the more recent ones were distinctly narrower. On the whole, the big-sized vessels prevailed in number the small-sized ones. The vessels are rather round and present in clusters of 2 to more vessels in different patterns beside solitary ones. One-to three layered rays were observed.

As stated before *Datura stramonium* developed at an earlier age a bigger amount of root conducting tissues than in either eggplant or tomato plant. Also, at older ages it was obvious that secondary growth proceeded at a faster rate in comparison with the two other species. This might be attributed to the need for supplying simultaneously both vegetative and reproductive growth (Carlsbecker and Helariutta, 2005).

However, Datura plant had to build-up conducting tissues during the period from 5-9 weeks after sowing capable of supplying an active vegetative growth which as dry weight increased from 0.970 to 4.127 g (Table 2) beside progressed reproductive growth. The xylem increased from 820.9 to 1904.9 μ during this period. The increase in height from 21.3 to 40.9 cm, in number of leaves from 7.5 to 24.5 and in total leaf area from 219.9 to 1400.2 apparently shred in the big increase in dry weight of the shoot system.

Generally, during the last 3 weeks of vegetative growth, i.e. between 9 and 12 weeks after sowing, the increase in xylem tissue was not proportional to the increase in dry weight from 4.127 to 7.378 g. The final xylem diameter averaged 2303.3 μ. The rather big increase in dry weight recorded at this period might be due for a minor part to the continued growth in plant height and for a major part to the still developed branches with their leaves since the stem is dichotomously branched. The eventual counted number of branches averaged 15.8 and the number of leaves increased from 24.5 to 37.0 during this period.

Table (1) :Mean measurements or counts for certain morphological characters and dry weight of the shoot system and the root xylem diameter at successive ages in *Datura stramonium* L.

Plant age (weeks)	Root xylem diameter (μ)	Plant height (cm)	Leaf number	Leaf area (cm^2)	Dry weight (g)
5	820.9	21.3	7.5	219.90	0.970
7	1417.9	31.7	17.8	712.77	2.427
9	1904.9	40.9	24.5	1400.20	4.127
12	2303.3	45.7	37.0	2155.70	7.378

Comparing the xylem tissue at base of the roots at end of vegetative growth, it was clear that the root possessed smaller number of vessels, but this was compensated by their bigger diameter, as tabulated below :

	Mean No. Vessels	Mean Vessel diameter (μ)
Datura stramonium	205	77.9
Capsicum frutescens	300	61.6

It was interesting to notice that the increase in amount of vegetative growth as estimated by dry weight appeared to go parallel with the increase in vessel diameter and not in number of vessels in the roots.

The two investigated species could be arranged according to amount of vegetative growth as medium category since the amount of vegetative growth represented by dry weight was 7.378 and 6.880 g/plant for *Datura stramonium* and *Capsicum frutescens*, respectively.

The arrangement could also be made with respect to the vessel diameter tabulated before.

As stated before, *Datura stramonium* developed at an earlier age a bigger amount of root conducting tissues than in either eggplant or tomato plant. Also, at older ages it was obvious that secondary growth proceeded at a faster rate in comparison with the other studied species. This might be attributed to the need for supplying simultaneously both vegetative and reproductive growth.

Capsicum frutescens L.

Secondary growth started in the diarch root 12 days after sowing. At the age of two weeks, a considerable amount of secondary vessels were developed (Fig. 6). The outermost cortical layer formed the exodermis with its thick suberized walls.
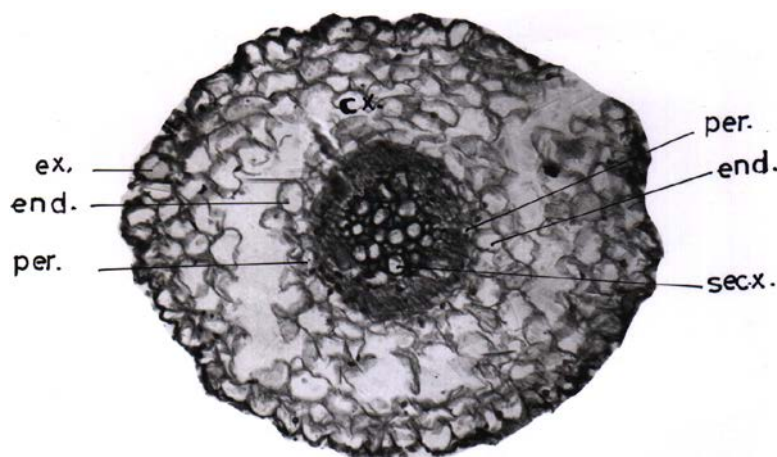

Fig. (6): Transection at the base of main root of *Capsicum frutescens*, 2-weeks old plant (X150).

Per.:Pericycle, end.: Endodermis, sec.x.: Secondary xylem, ex.: Exodermis.

The structure of the root at the age of 3 weeks differed mainly in amount of the xylem which reached an average diameter of 209 µ.

Age of 5 weeks :

As shown in Fig. (7) the exodermis was collapsed or crushed at several places and periclinal division of its still intact cells or of the underlying cortical cells occasionally took place. Dilatation growth of the cortex including the endodermis was detected.

The pericyclic cells were much elongate in the tangential direction, and to the extent that the tissue at several places assumed a too narrow strip, while at others it was nearly invisible. Such behaviour of the pericycle at such an early stage of growth was not recorded in the previous studied species. The xylem tissue reached 477.6 µ on the average.
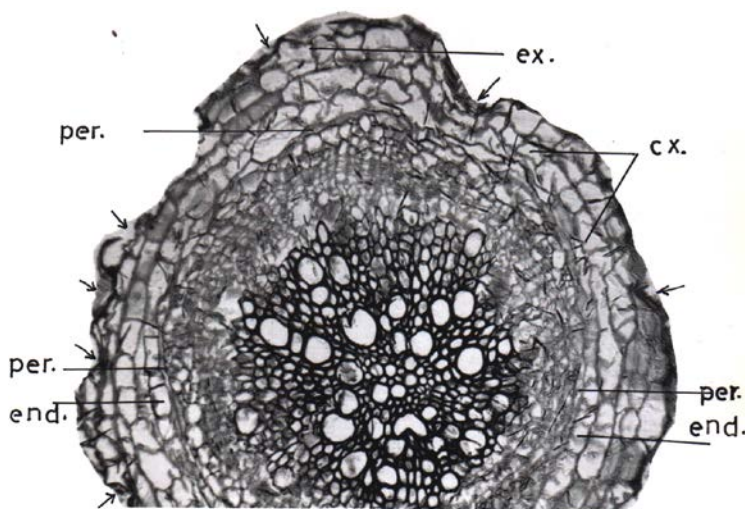

Fig. (7): Transection at the base of main root of *Capsicum frutescens*, 5-weeks old plant. At places pointed to by arrows the exodermis was crushed or collapsed (X150).

ex.: Exodermis, cx.: Cortex, Per.: Pericycle, end.: Endodermis.

Age of 7 weeks :

At the age of 7 weeks (Fig. 8) the mean diameter of the xylem tissue increases to 701.5 µ or about 30 % of its final diameter. The behaviour of tissues outside the vascular cylinder was in general, like that in the previous stage.

The cortical cells underlying the ruptured or collapsed exodermis were seen divided in one or more cells by tangential walls and thus the appearance of a periderm-like tissue was initiated.

The endodermis resembled the pericycle in appearing as much compressed tissue at many sites.

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The phloem parenchyma enlarged in size at several places, thus, separating more apart the groups of conducting elements i.e. the sieve tubes with their comparison cells.
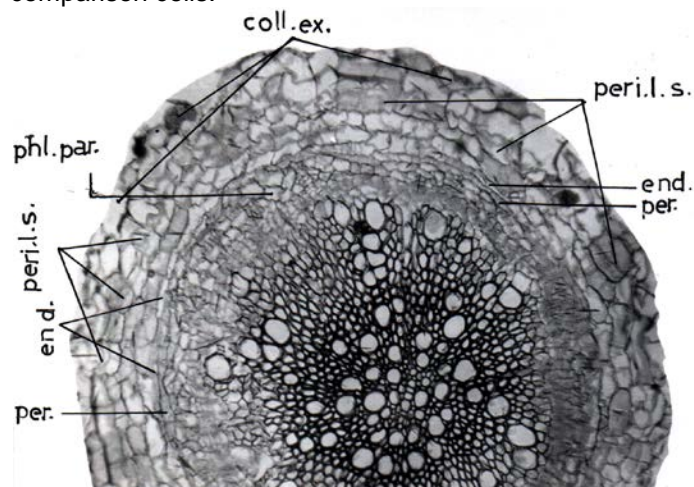

Fig. (8): **Transection at the base of main root of *Capsicum frutescens*, 7-weeks old plant (X125).**

Per.L.S.: Periderm-like structure, end.: Endodermis, Per.: Pericycle, phl.par.: Phloem parenchyma.

Age of 9 weeks :

During the next two weeks, the xylem reached about 50 % of its final diameter being 1119.4 µ on the average.

Tangential division inside many of the cortical cells underlying the original exodermis was actively continued and consequently the appearance of a periderm-like tissue became more easily manifested (Fig. 9). The rest of the cortical cells were dilated to varied extent and this caused the cells at some places to overlap during their tangential elongation. It was difficult to demonstrate the presence of both the endodermis and the pericycle due to the much narrowing of their extremely elongated cells at the tangential plane.

The phloem parenchyma cells showed tangential elongation followed in a few cells by radial division.
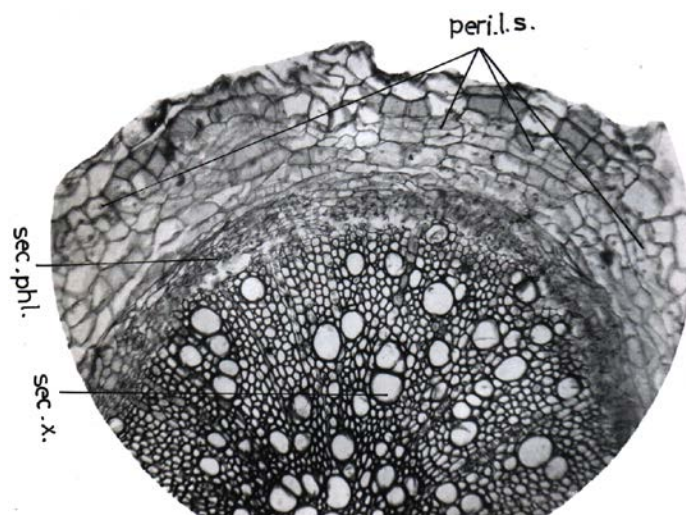
Fig. (9): **Transection at the base of main root of *Capsicum frutescens*, 9-weeks old plant (X150).**

Per.L.S.: Periderm-like structure, sec.phl.: Secondary phloem, sec.x.: Secondary xylem.

Age of 12 weeks :

At this age, the diameter of the vascular cylinder was nearly doubled than at the previous age (Fig. 10).

The xylem reached an average of 2000 µ in diameter. It appeared that the extraxylory tissues i.e. cortex and phloem, respond to this big increase root girth by active dilatation growth, that brought about various shapes and sizes of the resultant cells.

The periderm-like tissue originating as stated before in the cortical layer underneath the exodermis was obvious at many places but this tissue never formed a continuous layer characterizing a true periderm.

The phloem conducting elements were present in a rather scattered pattern and it was clear that the phloem was rich in parenchyma cells.
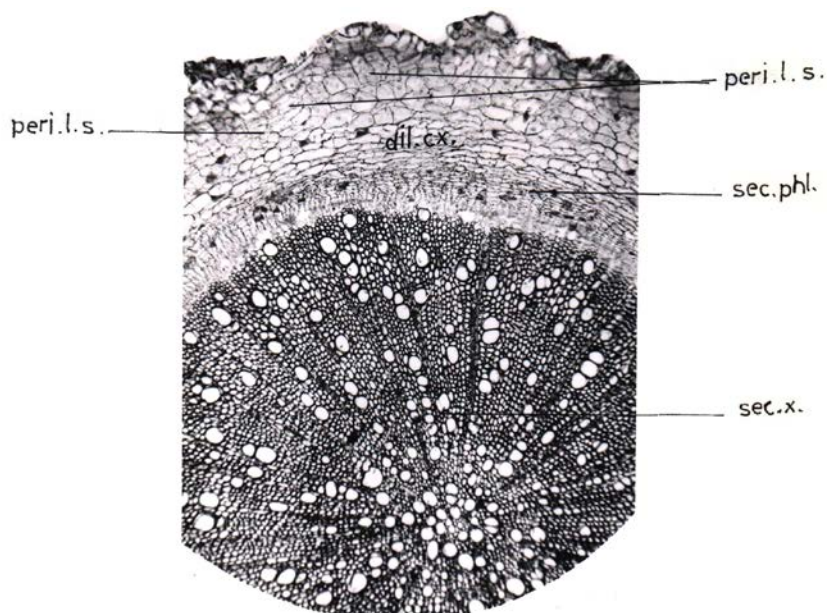
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Fig. (10): Transection at the base of main root of *Capsicum frutescens*, 12-weeks old plant (X77).

Per.L.S.: Periderm-like structure, sec.phl.: Secondary phloem, sec.x.: Secondary xylem.

Xylem characteristics :

Lignified parenchyma cells from mainly the ground tissue of the xylem. As seen in cross sections, the rays were one cell wide.

The vessels were almost of same size being round or oval. Their presence solitary dominate their development in clusters of 2 to 3 vessels, nearly in radial pattern.

Table (2):Mean measurements or counts for certain morphological characters and dry weight of the shoot system and the root xylem diameter at successive ages in *Capsicum frutescens* L.

Plant age (weeks)	Root xylem diameter (μ)	Plant height (cm)	Leaf number	Leaf area (cm^2)	Dry weight (g)
5	477.6	6.5	8.4	----	----
7	701.5	10.3	11.7	173.2	0.722
8	1119.4	19.5	16.6	410.4	1.672
12	2000.0	31.9	53.2	970.7	4.666
15	2447.7	39.8	65.7	1021.3	6.880

From Table (2) it appeared that the doubling of the xylem tissue in amount during the period between 9 and 12 weeks after sowing, brought the tissue to more than 80 % of its final diameter. This active formation of conducting tissues seemed to be required for the active vegetative growth during this period. The average dry weight of the shoot system increased from 1.672 to 4.666 g/plant. The mean stem length increased from 19.5 to 31.9 cm; number of leaves from 16.6 to 53.2 and the leaf area from 410.4 to 970.7 cm^2. Also, it was noticed that the doubling of diameter of the xylem tissue during the 3 weeks mentioned above was to secure a supply of water and minerals, not only enough for the active vegetative growth, but also for the reproductive activities already started by the end of the mentioned weeks.

Vegetative growth continued also actively during the three weeks lying between 12 and 15 weeks after sowing. The average dry weight of the shoot system increased from 4.666 to 6.880 g/plant.

Finally, it was shown at the age of 7 weeks that the xylem at the root base achieved a portion of its final diameter bigger than that of the final dry weight of shoot system accomplished by the plant. The portion percentages were 30 % and 10.3 %, respectively.

It appeared that at younger ages vascularization in the root progressed at a rate faster than needed for the accomplished vegetative growth, thus affording early in growth a sufficient number of xylem conducting elements which with the continued development of new ones, could successfully perform the complete vegetative body of the plant.

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النمو الثانوي فى جذور كل من الفلفل والداتوره خلال مراحل نمو متتالية

عرفه أحمد عرفه

قسم النبات الزراعي – كلية الزراعة – جامعة المنصورة

كان موضوع الدراسة هو إتجاه النمو الثانوي وما يتبع ذلك من تكوين أنسجة الحماية فى جذور نوعين من الأعشاب الحولية العشبية من العائلة الباذنجانية هما الداتوره والفلفل.

ولقد كانت النتائج الرئيسية كما يلى :

1- وجد فى الجذر ولدى قاعدته أن نسيج الخشب قد تكونت نسبة من كميته النهائية هي بالتأكيد أكبر من إحتياجات المجموع الخضري المتكون فعلاً فى النبات وذلك خلال مرحلة مبكرة من النمو.

2- وجد أن كمية النمو الثانوي فى جذور كل من النوعين محل الدراسة متباينة ولقد كان واضحاً أن كبر كميته ما هو إلا إستعداد من الجذر لتكوين شبيه البريدرم ومع إزدياد الكمية يتكون البريدرم الحقيقي.

3- وضح أن شبيه البريدرم يتكون فى الإكسودرمس أو الطبقة القشرية الأسفل بينما يتكون البريدرم الحقيقي عادة فى البريسيكل وذلك تبعاً للنوع.

4- يمكن أن يسلك كلا من الإندودرمس والبريسيكل نفس السلوك أو يختلفا تبعاً لإستجابتهما للنمو الثانوي.

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