

PHYSIOLOGY OF ROOTS

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Abstract. General functions of roots are examined and it is concluded the main function of healthy roots of growing plants is a propensity to grow and form branch roots. External and internal factors affecting root growth are briefly reviewed. An important aspect of root growth is a marked sensitivity to the essential micronutrient element boron. A relationship is drawn between the behavior of boron-deficient tomato and squash roots and the initiation and growth of roots on stem cuttings.

Roots are first. When seeds germinate the radicle is the first organ to emerge from the seed, and when cuttings are made roots must be initiated and grow before growth of the cuttings progresses to any extent. Even in the newer methods of micro-propagation by tissue culture a successful result is obtained only when roots develop and, in most of these instances, root organization and formation is an early event and often the first organized growth from callus tissue in a medium containing the correct balance of nutrients and growth substances. It is commonly observed in transplanting seedlings from flats to nutrient solution culture that the shoot of the seedling doesn't begin vigorous growth until new roots are formed and a certain critical mass of roots is present. Overall, a plant is an autotrophic organism (able to synthesize its own food), but only the shoot is autotrophic; the root system is actually heterotrophic (not able to synthesize its own food). For successful growth, roots must supply water and mineral nutrients to the shoot to sustain its autotrophy. In return the root system depends on carbohydrates and other growth substances to sustain its heterotrophy. Thus there is a reciprocal relationship between shoot and root growth but, in the beginning, roots develop first.

ROOT FUNCTIONS

Perhaps the most essential function of healthy roots of growing plants is growth because the root system "mines" the soil to obtain the necessary water and mineral nutrients for the plants. As roots exhaust the water and nutrients in their vicinity they obtain new supplies by growing into new soil masses. By continued growth they invade new areas of soil to supply the steady needs of water and nutrients to sustain shoot growth and, in turn, their own growth. Roots grow in a branching pattern which makes the root system very efficient in extracting available water and nutrients from a given soil volume and also provides a firm anchorage for the plant. Thus, as a result of root growth we can account for the three common external functions ascribed to roots: anchorage, plus water and nutrient absorption.

Internal activities of roots are also keyed to growth relationships. Sites of high metabolic and growth activity are often referred to as "sinks." Growing roots are regions of high sink activity and enhance growth of shoots by using the products of photosynthesis. Roots assimilate nitrate and ammonium nitrogen into organic nitrogen compounds, principally amino acids which, in addition to being available to the root, are translocated to the shoot. Roots also synthesize hormones and other substances for export to other parts of the plant.

From the preceding we may conclude that if a root system doesn't grow, all other aspects of its functions become of minor consequence since the growth of the whole plant is checked. Thus the most essential function of roots of growing plants is growth.

FACTORS AFFECTING GROWTH

When main or primary roots stop growing due to unfavorable conditions maturation progresses down the root axis to the tip. Branch or lateral roots may emerge close to the tips of non-growing primary roots, but these laterals cease growth and mature if conditions remain unfavorable. Cessation of growth also results in the tendency for suberization to occur up to the tips of all roots. Suberization reduces root absorbing capacity and thereby greatly reduces root activity, resulting in what may be called "dormant" roots. When viewed in this way roots may be characterized as exhibiting a spectrum of growth activity, with vigorous growth at one extreme and complete dormancy at the other. By briefly examining factors affecting root growth we can visualize their position and responsiveness on this growth activity spectrum.

Important external factors affecting root growth are moisture, aeration, carbon dioxide, pH, minerals, salt concentration, and temperature. Although water in itself is not injurious to roots, excess water in soil displaces air from the pore spaces and results in oxygen deficiency and reduced growth (8). If roots are deprived of oxygen for too long a period of time they die. Good aeration assuring sufficient oxygen for metabolism is essential for healthy growing roots. If water becomes deficient growth ceases and the roots tend to mature and suberize up to their tips, resulting in a reduced capacity for absorption of nutrients and water (8). Due to root respiration, high levels of carbon dioxide may occur in the root zone. However, excess carbon dioxide is not thought to inhibit respiration as much as oxygen deficiency. Root growth is inhibited by extremes of pH (less than 3 and greater than 9), but is only slightly affected in the range of 4 to 8, providing sufficient calcium is present and toxic ions such as aluminum and manganese are not present in excess. Excesses of these elements are recognized as major factors in poor plant growth on acid soils (10). It is generally recognized that a complete and balanced mixture of essential elements is necessary for plant growth.

However, information is meager about the effects of specific ions on root growth. It is recognized that phosphorus stimulates root growth, while deficiencies of boron and calcium yield short stubby branch roots and eventually may cause the tips to die (8). Overall high salt concentrations tend to slow down and eventually stop elongation with a resultant hastening in maturation. This yields roots which are suberized to the tips and appear dormant. Species may vary widely in their tolerance to different levels of salt (8).

Root growth is often limited or stopped by low and also by high temperatures. Optimum temperature varies in relationship to species, stage of development and oxygen level, but is generally in the range of 20 to 25°C. Unfavorable temperatures inhibit elongation and, as elongation is limited, roots become differentiated up to the apex (6). However branching often continues in these roots, and laterals occur almost to the root apex. At cool temperatures roots are usually whiter, thicker in diameter, and less branched than at warmer temperatures. At high temperatures roots become filamentous and "weak" in appearance in contrast to the "robust" appearing roots seen at low temperatures (11). In pot experiments with corn it was found that for each degree increase in soil temperature in the range of 12 to 26°C, total seedling dry weights were 20% greater than at each preceding temperature, and 12% smaller with each degree increase from 26 to 35°C (15). Nutritional status, leaf numbers and size, stem lengths, and root numbers were also very dependent on root temperatures. Although corn was chosen for this study because of its known sensitivity to soil temperature, it seems clear that small changes in pot soil temperature can have significant effects on plant behavior. It would seem very likely that similar, if not as pronounced effects, could also occur in container-grown ornamental plants. In another experiment with corn, root growth was found to occur in a series of stages or pulses which were associated with stages of top growth (7). In view of the various influences reviewed, it is evident that root growth and condition is of prime importance to obtain good quality container-grown plants.

Important internal factors influencing root growth are translocation, sink relationships, nitrogen assimilation and the synthesis of hormones. Over the past decade more attention has been given to the synthetic activities of roots and their ability to act as metabolic sinks. An aspect of low temperature effects on roots is a decrease in root growth and a concomitant reduction in their capacity to act as sinks for carbohydrate translocated from shoots. In sugar cane such a slow-down of carbohydrate translocation from shoots resulted in an accumulation of carbohydrates in leaves which depressed photosynthesis and decreased yields (11). Also, in cotton it was found that low root temperature caused carbohydrate content of the tops to increase rapidly, but carbohydrate was very low in the roots (11). Thus, an actively growing root system appears essential for rapid

translocation of carbohydrate from the shoot and a continued rapid growth of the shoot. In many plants most of the nitrogen passing from root to shoot is in the organic form. From these observations it is concluded that the root system as a whole is an active site of amino acids synthesis for transport to the shoot (11). Factors associated with reduced photosynthesis and reduced metabolic activity in roots leads to an accumulation of nitrate nitrogen in plants. Reduced photosynthesis limits carbon compounds able to accept amino (reduced) nitrogen; since nitrate isn't reduced it accumulates (11). In the last 12 years abundant evidence has indicated that normally growing roots synthesize and export gibberellins, cytokinins and abscisic acid to the shoot (3, 12). Although auxin or indoleacetic acid (IAA) is the oldest of the known hormones its role in root growth has been unclear. In a critical review of auxins and roots Scott (12) concluded that auxin moves from the morphological top to the bottom of the plant in accordance with response to gravity. The evidence of xylem regeneration, lateral root initiation, and root maturation all point to development in the root from the basal part to the apex, from the top downward, and auxin regulation of root growth and development appear to be a consequence of this pattern of auxin movement.

BORON, AUXIN AND ROOTING

Of the three nutrients, phosphorus, calcium, and boron, which were previously mentioned as having direct effects on root growth, boron is unique with respect to the small amount required for activity, i.e., 0.1 ppm or less. When tomato and squash plants are grown in complete nutrient solutions with 0.1 ppm boron and then transferred to nutrient solutions with no boron, root elongation ceases as early as 6 hours and routinely is completed by 24 hours. Typical deficiency symptoms are a browning of the root tips and the appearance of lateral roots close to the primary tips. The terminal 2 to 3 mm of the tip is brown and lateral roots, which are normally present 5 to 7 cm behind the tip of the primary root, appear within 5 mm of the tip. Roots deprived of boron will recover and resume elongation if boron is resupplied within 24 hours, but if the deficiency extends beyond this critical time, recovery will not occur. Although elongation of the primary or main root stops in the absence of boron, maturation and differentiation, as evidenced by the initiation of new lateral roots, progresses down the root. If the laterals are not supplied with boron within their critical time they too stop growing. A boron deficient root thus looks stubby and bumpy. The stubbiness from lack of elongation of the primary and lateral tips and the bumpiness from lateral initials that are arrested in various stages of development (1, 2, 4, 6, 9). In this sense, stem cuttings that have many root initials that haven't elongated to produce new roots are similar to boron deficient roots.

If auxin movement is down the root axis as suggested by Scott (12) and as is indicated by the maturation which progresses down the root under boron deficiency, when the roots have stopped elongating, the amount of auxin should be greater in boron-deficient than in boron-sufficient tips. In a study conducted by Bohnsack (4) in which he measured the level of IAA oxidase (an auxin destroying enzyme) activity in squash roots, he found a higher level of IAA oxidase activity in the tips of boron-deficient roots than in the plus boron controls (Table 1.) Assuming a higher oxidase activity is indicative of a higher auxin content, it is clear that boron-deficient roots have higher auxin levels than plus boron controls. Further, in recovery experiments he found that boron added to boron-deficient roots decreased the oxidase activity from higher to lower levels as recovery progressed, until finally it was the same as normal plus-boron plants not subjected to the deficiency treatment (Table 2.).

Table 1. IAA oxidase activity of the apical 5 mm of root tips of squash plants grown with 0 and 0.1 ppm boron added to the nutrient solution.

Hours of Treatment	+B	-B
	$\mu\text{g IAA destroyed} \times \text{hr}^{-1} \times \text{mg dry wt}^{-1}$	
6	0.82 ± 0.01^1	0.85 ± 0.16
9	0.95 ± 0.05	3.5 ± 0.21
12	0.77 ± 0.02	17.8 ± 1.30
18	0.83 ± 0.06	21.6 ± 0.52
24	0.78 ± 0.05	21.6 ± 0.77
36	0.92 ± 0.04	21.1 ± 0.63

¹Mean of three experiments with standard error of mean.

Table 2. IAA oxidase activity of the apical 5 mm of root tips of squash plants grown with 0.1 ppm boron until exposed to a boron deficient solution for 12 hrs followed by recovery in a solution with adequate boron.

Hours on Treatment	$\mu\text{g IAA destroyed} \times \text{hr}^{-1} \times \text{mg dry wt}^{-1}$
12 (-B)	13.9 ± 0.3^1
6 (+B)	8.2 ± 1.1
12 (+B)	3.5 ± 0.4
18 (+B)	2.1 ± 0.6
24 (+B)	0.86 ± 0.09^2

¹Mean of three experiments with standard error of mean.

²Adequate boron control: 0.85 ± 0.03 .

Since root elongation stops so rapidly when boron is withheld a role for boron in cell division was suspected. To critically investigate this possibility Cohen (5) examined meristems of boron-sufficient and deficient squash roots treated with radioactive thymidine, a chemical necessary for deoxyribonucleic acid (DNA) synthesis. By examining the pattern of radioactive labeling in cells of root tips by autoradiography one can determine which cells are synthesizing DNA and thus capable of division. His results showed that the ability of root tip cells to incorporate radioactive thymidine is correlated with the total root elongation during the boron deficiency treatment period (6). Cessation of elongation and mitosis occurred as early as 6.5 hrs after boron was withheld from the nutrient solution while DNA synthesis occurred for as long as 20 hrs after withholding boron. If boron is not supplied to boron deficient roots very shortly after root tip cells stop synthesizing DNA they will not recover and resume elongation. However, if boron is resupplied within the critical time period the cells regain first the ability to synthesize DNA, then cell division activity is restored and finally root elongation is resumed (Table 3).

Table 3. Root elongation and incorporation of radioactive thymidine in boron-sufficient, boron-deficient, and recovering root tips of squash plants grown in nutrient solutions with 0.1ppm and 0 ppm boron.

Treatment	Mean No. Labeled Nuclei ¹	Mean Root Elongation ² (mm)
+B, 20 hr	52	29.5
-B, 18 hr	58	4.3
-B, 20 hr	0	4.4
-B, 20 hr with +B recovery for:		
3 hr	0	0
6 hr	0	0.2
9 hr	7	0.5
12 hr	48	0.7

¹Means of 30 roots.

²Means of not less than 40 roots.

Root elongation is ultimately dependent on cell division of meristem cells and their subsequent enlargement. Under conditions of boron deficiency, cell division and root elongation stop, followed by the loss of DNA synthesis at the primary root tips. When cell division of the primary tip stops, auxin continues to move down the root and, concomitant with that movement, lateral root primordia are stimulated to differentiate. If those primordia receive boron before their critical time without boron is exceeded, they will start to elongate — if not, they too die. Thus we have a series of lateral meristems that go through the same sequence of events as the primary meristems.

The results obtained from the study of boron deficiency in squash and tomato roots are, I believe, strikingly similar to what one may observe in the rooting of stem cuttings using the auxin IBA. There are various reports in the literature (14) on stimulatory responses of boron with auxin on rooting. In two of these studies (16, 17) it was found that boron had a striking effect on enhancing the number of roots produced on cuttings of clematis (30% increase) and English holly (20% to 40% increase). In both of these investigations the boron and IBA treatments were given as a 12 hr soak with the cuttings stuck in sand under mist.

It is common knowledge among propagators that auxin stimulates rooting. However, the rooting response can be divided into two phases, initiation of new root initials and elongation of the initials into roots. Auxin stimulates the formation of root initials, and from the data obtained on boron and root elongation we may conclude that boron is necessary for the elongation of the initials. The appearance on cuttings of many initials that don't elongate may be due to a lack of boron essential for the cell divisions that are necessary for initial elongation and production of roots.

The physiology of boron-deficient root elongation and lateral root behavior is similar to the physiology of root initiation and growth of stem cuttings. Boron is required for the growth of laterals on boron-deficient roots and for the growth of adventitious root initials on cuttings. In both cases auxin stimulates the formation of the root initials. With roots on intact plants the source of auxin is from the top of the plant moving down the root axis with stimulation of initial formation. For stem cuttings the auxin source is the applied auxin treatment, or in non-auxin treated cuttings, the downward migration of auxin from sites of auxin synthesis such as young leaves, buds or the vascular cambium. I believe these observations and conclusions indicate it is appropriate to examine in more detail possible beneficial effects of boron and auxin on rooting responses of stem cuttings.

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