TECHNICAL PAPERS

Absorption of Radioactive Phosphorus by Mycorrhizal Roots of Pine[1]

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It has been demonstrated that under certain conditions, especially when growing in infertile soil, tree seedlings bearing mycorrhizal roots grow better than those which do not possess mycorrhizae. The improved growth apparently results from greater absorption of mineral nutrients. Routien and Dawson (*3*), for example, found that seedlings of *Pinus echinata* possessing mycorrhizae absorbed more calcium, magnesium, iron, and potassium than seedlings lacking mycorrhizae, and McComb and Griffith (*2*) found the same to be true of certain other species of conifers with respect to phosphorus. Hatch (*1*) attributed most of the beneficial effect of mycorrhizae to the increased absorbing surface, but others (*2, 3*) believed the higher metabolic activity of mycorrhizal roots to be the cause of their greater intake of minerals.

Apparently no direct measurements of mineral absorption by mycorrhizal roots have been made, the conclusions concerning their high efficiency as absorbing structures being based on differences in growth and in chemical composition of plants possessing them as compared with plants lacking them. By immersing roots in radioactive phosphorus and preparing radioautographs it is possible to obtain direct evidence concerning the relative amounts of phosphorus absorbed by various root regions. This method was used to compare the amounts of phosphorus accumulated in mycorrhizal and nonmycorrhizal roots of pine seedlings. The species used were loblolly pine (*Pinus taeda* L.) and red pine (*P. resinosa* Ait.).

Pieces of root 10 to 15 cm in length obtained from potted pine seedlings were carefully washed in distilled water, and gently freed of bits of soil and other foreign matter with a camel's hair brush. P^{32}, essentially carrier-free, was obtained in acid solution and was diluted with distilled water to the desired activity which varied from 100–500 μc 1 in various experiments. The pH of the solution was adjusted to about 5.5 with NaOH. Roots were exposed for 3 or 4 hr to the radioactive solution in shallow dishes to insure adequate aeration. They were then rinsed in distilled water for 30 sec, in 0.001N H_3PO_4 for 10 sec, and again rinsed in distilled water for 30 sec.

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The acid rinse was used to aid in freeing the surfaces of the roots from adsorbed radioactive phosphorus by exchange. The washed roots were blotted and dried between glass plates. Medium lantern slides were then placed with emulsion side toward the roots, but separated from them by sheets of aluminum foil, 0.01 mm thick, for periods ranging from a few hours to 3 days, depending on the activity of the roots.

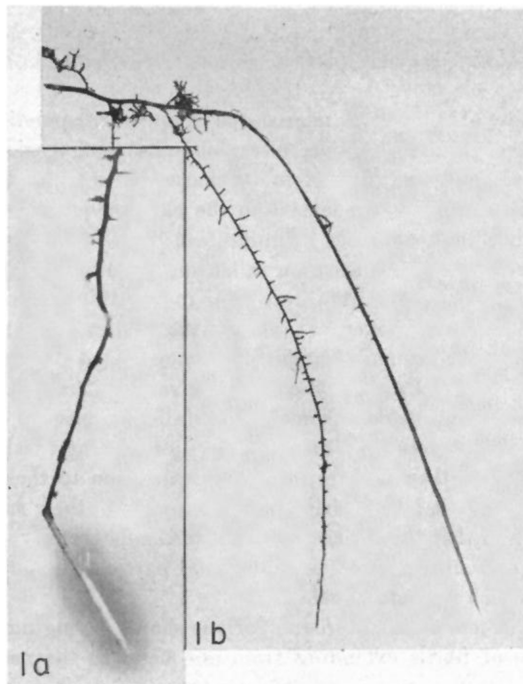

FIG. 1. Photographs (actual size) of portions of roots of red pine seedlings grown in soil, showing unsuberized (lower portion of each root) and suberized regions, mycorrhizal clusters (upper portion of 1b), and short branches. Many of the latter appear to be pseudomycorrhizal or nonmycorrhizal.

Photographs of typical roots of red pine are shown in Fig. 1, a and b, and their radioautographs in Fig. 2, a and b. These roots were cut into 1-cm segments and the activity of each segment was measured with a Geiger-Müller counter. The relative activity of each segment is indicated on the autographs.

As would be expected, there is less phosphorus accumulated in the completely suberized portions of the main roots than in the unsuberized region near the root tips. This is apparent in Fig. 2a and the left hand root of Fig. 2b. The right hand root in Fig. 2b is less mature and the numerous branches are scarcely visible. The coralloid clusters of mycorrhizal roots (upper portion of Fig. 2b) show the largest accumulation, supporting the common belief that they are actively involved in nutrient intake. Perhaps even more interesting is the relatively great accumulation of radioactive phosphorus by the short branch roots, even by those which show none of the hypertrophy or dichotomous branching characteristic of typical mycorrhizae. Some of these branch roots resemble those pictured by Hatch (*1*) in his Plate III, A, B, and F, and in Plate XIII, B, and described as nonmycorrhizal or pseudomycorrhizal roots. These he believed to be of negligible importance in the absorption of nutrients. Although these roots appeared to be suberized almost to their tips and therefore had a very limited absorbing surface we found that they accumulated surprisingly large amounts of phosphorus. Measurements of radioactivity indicated that the tips of the main roots

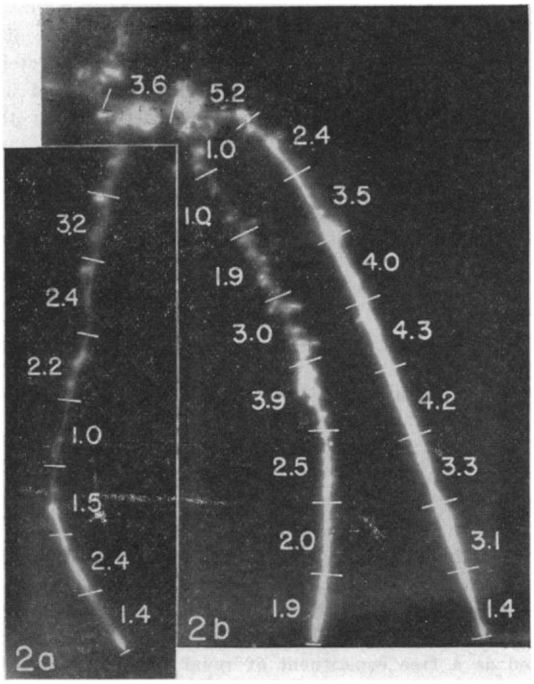

FIG. 2. Radioautographs of the roots pictured in Fig. 1. The numbers indicate relative radioactivity of 1-cm segments as measured by a Geiger counter.

did not contain as much radioactive phosphorus as the older portions bearing branches. A heavily suberized root bearing a few short branches or a small cluster of mycorrhizal roots apparently can absorb more phosphorus than an unbranched tip with a much larger unsuberized surface.

In a few instances autographs indicated that completely suberized roots absorbed considerable phosphorus, but microscopic examination of such roots showed that they were covered with a superficial layer of mycelium which apparently has a very large capacity to accumulate phosphorus. The few mycorrhizal branches which failed to show accumulation were dark and wrinkled, and apparently dead. A few unsuberized root tips showed very low accumulation of phosphorus, indicating that not all unsuberized root surfaces are equally active in absorption of solutes.

It appears possible that the increased accumulation of phosphorus by mycorrhizal roots may not result only from increased surface but perhaps may be related to a greater capacity for absorption because of high metabolic activity. Routien and Dawson found the oxygen consumption of mycorrhizal roots to be two to four times that of nonmycorrhizal roots. In preliminary experiments we have found that 10^{-3} M sodium azide, which may be expected to inhibit oxygen consumption, reduces the accumulation of P^{32} by both mycorrhizal and nonmycorrhizal roots. Study of the effect of respiration inhibitors was complicated by variations in proportion of mycorrhizal to nonmycorrhizal tissue in root samples, making quantitative comparisons difficult.

The results of these experiments show that mycorrhizal portions of roots of pine can accumulate much larger quantities of phosphorus than nonmycorrhizal portions. Root segments bearing short, unbranched, pseudomycorrhizal lateral roots appear to accumulate more phosphorus than typical unsuberized root tips. It seems possible that mycorrhizal roots have not only a greater surface, but also a greater capacity to accumulate phosphorus per unit of surface than nonmycorrhizal roots. Apparently in pine roots the region of maximum intake of minerals is not the root tips, as in roots of herbaceous plants, but the older regions where mycorrhizal or pseudomycorrhizal branch roots have developed.

References

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A Qualitative Analysis of the Amino Acids in Royal Jelly[1]

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Royal jelly, a secretion of the pharyngeal glands of the young worker of the honey bee, *Apis mellifera* L. (*6*), is the sole food of bee larvae which develop into sexually mature female adults, or queens. Larvae which develop into sexually immature female adults, or workers, are fed royal jelly for 2 days, then receive a mixture of pollen and honey during the remainder of their 5-to-6-day feeding period (*9*). Research toward an explanation of the role of royal jelly in the development of castes of *Apis mellifera* L. has been reviewed by Haydak (*6*).

Chemical analyses of royal jelly have shown that it is a complex mixture of substances having a protein content of 9 to 18% of the fresh material (*6*). The proteins were found by Abbott and French (*1*) to consist of an

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