seed and seedling extracts and malic acid was identified in all seedling extracts. Inorganic acid anions were also identified.

**Literature Cited**

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**TRANSPIRATION AND THE ABSORPTION AND DISTRIBUTION OF RADIOACTIVE PHOSPHORUS IN PLANTS1,2**

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After a century of investigation the relationship of transpiration to the absorption and distribution of mineral salts in plants is still a controversial question. Evidence has been presented which supports the idea that these processes are largely independent while other researches have indicated enhanced absorption with increased transpiration. This lack of agreement among earlier investigators is brought out in the literature reviews and the experimental work of Haselbrinck (3), Mendiola (7), and Muenscher (8). More recently an exhaustive set of experiments and an extensive review of the literature has been published by Hylmo (5). His results showed that in most species of plants tested the roots were strongly dependent on water transport for their supply of ions, and in all species the ion uptake by the stem and leaves was directly proportional to the water transport. The research reported in this paper was performed to determine the effects of transpiration upon the absorption and translocation of radioactive phosphorus in the sunflower plant, using the autoradiogram technique.

Seeds of the Mammoth Russian variety of sunflower were planted in sand and at the age of two weeks the seedlings were transferred to a complete culture solution. Culture solutions were changed twice a week, and daily supplements of distilled water were added to keep the solution at the top level. At the age of one month the plants were transferred to 500-ml bottles. For four days previous to the experimentation with P32, when the plants were two months old, a record of the daily rate of transpiration was kept, and plants having approximately equal rates of water loss were selected for use. A complete culture solution containing 275 μC P32/500 ml was placed in each culture bottle, plants inserted, and allowed to remain in contact with the P32 for 90 minutes, two under ordinary greenhouse conditions with one in light and the other in darkness, and a third in a high humidity chamber in light. Another two plants under greenhouse conditions in light had sucrose added to the culture solution such that the osmotic pressures of the culture solutions were 2.24 atms and 3.83 atms as determined by the cryoscopic method. The osmotic pressure of all of the culture solutions without added sucrose was 0.47 atms. During the 90-minute period all of the plants were in air kept in constant

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motion by an electric fan. Relative humidity was computed from the readings on a wet and dry bulb thermometer.

At the end of the 90-minute run water loss was measured and then one leaf from each pair of leaves was cut from a plant, the leaves from each plant placed between blotters and dried under pressure for two hours. The leaves were then placed on a piece of herbarium paper, covered with a 20 x 25 cm sheet of Kodak No-Screen x-ray film and another sheet of herbarium paper, pressed under a weighted board, and exposed to the film for 14 hours. Films were developed with Kodak x-ray developer and fixer, and exposed to the photographic paper for 0.5 second. The distribution of radioactive phosphorus in the leaves is recorded in figure 1.

These autoradiograms indicate that there is a positive relationship between the rate of transpiration and the absorption of P\textsuperscript{32} and its translocation to the leaves of the plants, higher rates of transpiration being associated with greater amounts of P\textsuperscript{32} in the leaves. During the course of the experiment a Geiger counter was used to detect the presence of P\textsuperscript{32} in plants A and B, leaves at the top of plant A and at the bottom of plant B being the first to register on the counter. The autoradiograms of these plants show higher concentrations of P\textsuperscript{32} from top to bottom of plant A and from bottom to top of plant B. It looks as if a rapidly moving transpiration stream carried the phosphorus past the leaf traces of the lower leaves of the high transpiring plant, and a slowly moving transpiration stream allowed phosphorus to enter the lower leaf traces of the low transpiring plant. Hanson and Biddulph (2) set out to determine if there actually is a diurnal fluctuation in the amount of ions absorbed by the root or translocated to the xylem, and to determine if possible the cause of any noted variation. They found that plants in the daytime translocated four times as much radioactive rubidium to the shoots as plants in the nighttime. This increased daytime absorption and translocation of ions was attributed to some inherent capacity of the root symplast which is dependent upon an adequate supply of metabolites, a low salt-high sugar status, or possibly some unknown factor associated with illumination. They did not mention the possible effect of increased transpiration rate during the day. The difference in the amount and distribution of P\textsuperscript{32} in plants A and B could be affected by any of the factors postulated by Hanson and Biddulph since daylight and night were variables. However, it would seem foolishly to rule out the influence of the differing rates of transpiration.

Plant C with a transpiration rate reduced by higher humidity shows a relatively low concentration of P\textsuperscript{32} when compared with plant A. In this case both plants were in light and humidity was the variable, thus leading to the conclusion that differences in P\textsuperscript{32} content could be attributed to difference in transpiration rates. That light might, under certain conditions, have some influence on translocation may be inferred from a comparison of plants B and C where the experimental variables were light and relative humidity. Although the rates of transpiration in these plants were about the same, the pattern of distribution of P\textsuperscript{32} was quite different.

Plant D with a transpiration rate slightly reduced by a culture solution of 3.83 atm osmotic pressure (O.P.) showed a decrease in P\textsuperscript{32} content, a decrease greater than might have been anticipated with the degree of water loss reduction involved. The plant wilted considerably before the termination of the run, indicating that a portion of the water loss came from the plant tissues and not from the culture solution, thus largely cancelling the effects of a transpiration stream which could carry P\textsuperscript{32} from the roots to the leaves. The plant placed in a culture solution of 22.24 atm osmotic pressure is not recorded in figure 1. In this case the water loss was the same as from plant A in a culture solution of 0.47 atm osmotic pressure, and it was difficult to distinguish any differences in the autoradiograms.

The use of sucrose in the culture solution may be open to some criticism. That higher osmotic pres-
sures of culture solutions obtained by the addition of sucrose bring about a marked reduction in the entrance of water into a plant has been demonstrated by Tagawa (11), Roseone (9), and Hayward and Spurr (4). According to Long (6) the addition of sucrose to the culture solution greatly reduced water entrance into the roots of tomato plants but did not greatly disturb mineral entrance. Speoehr (10) and Went and Carter (12) found that sucrose was not absorbed through roots and its effect on growth was negligible. In direct contrast Dormer and Street (1) studying carbohydrate nutrition of tomato roots postulated a phosphorylation of sucrose with consequent entrance into plant cells. Hanson and Bidulph (2) studied the effect of added sucrose on the translocation of absorbed ions. They found that in darkness the addition of sugar to the culture solution had no significant effect on the translocation of phosphate to the shoots, but that in the daytime the translocation of phosphate did increase. This latter phenomenon was attributed to the increase in photosynthetic sugar and not to the sugar in the culture solution. The autoradiograms obtained from the plants whose roots were in contact with sucrose in the experiment here recorded show no enhanced absorption of phosphorus. Quite to the contrary, there is a reduced amount of P32 in the leaves of these plants, a condition attributed to a lack of transport of P32 to the tops of the plants because of a reduction in water flow through the xylem.

After two months of growth in a complete culture solution sunflower plants were transferred to culture solutions containing 275 μe P32/500 ml and allowed to remain in contact with the P32 for 90 minutes. Transpiration was varied by light vs darkness, different relative humidities, and different osmotic pressures of the culture solution obtained by the addition of sucrose. The distribution of P32 in the leaves was recorded by means of autoradiograms. It was noted that whatever means was used to vary the amount of transpiration, and, therefore, the amount of water absorbed and transported upward through the plants, there was a positive relationship between water transport and the amount of P32 accumulation in the leaves, higher rates of water loss being associated with greater amounts of P32 in the leaves.

LITERATURE CITED


STUDIES ON NITRITE METABOLISM IN HIGHER PLANTS

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The theory that nitrate reduction proceeds in a stepwise manner as follows

\[
\text{NO}_3^- \rightarrow \text{NO}_2^- \rightarrow \text{HNO} \rightarrow \text{NH}_2\text{OH} \rightarrow \text{NH}_3
\]

has received much support recently with the isolation of nitrite reductase by Evans and Nason (2), the isolation of hydroxylamine reductase by Zucker and Nason (7) and the demonstration of a nitrite reductase system which reduces nitrite to ammonia (4).

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In this report manometric experiments with nitrite and experiments with N15O2+ are presented to show further evidence that nitrite is an intermediate in nitrate reduction.

During manometric studies to determine the effect of nitrite on the processes of respiration and photosynthesis it was observed that leaves infiltrated with nitrite and exposed to light rapidly evolved a gas and caused a simultaneous disappearance of nitrite. Investigation showed that the gas was almost entirely oxygen and that the amount of oxygen evolved was