Movement of 2,3,6-Trichlorobenzoic Acid from One Plant to Another through Their Root Systems

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Two very effective growth-regulating compounds were found to move from one plant to another through the root systems. 2,3,6-Trichlorobenzoic acid when placed on stems or leaves of bean plants was absorbed and translocated throughout the plants, causing growth modification. The compound moved downward through the treated plants into their roots and out into the surrounding soil. Roots of nearby untreated plants absorbed the compound and translocated it upward into their above-ground parts, where it induced modification of growth. Transfer of the compound also occurred between the root systems of various other genera. 2,3,5,6-Tetrachlorobenzoic acid also was transferred in a similar manner from one plant to another.

Movement of α-methoxyphenyl acetic acid out of the roots of plants to which it was applied into nearby roots of untreated plants was recently demonstrated (*3*). Since then, numerous compounds have been tested to learn whether they too exhibit this characteristic. The general purpose of this research is to develop a means of protecting plants from organisms that attack their roots, or of inducing useful responses through transfer of a systemically effective chemical from one root system to another. In the course of experiments on flower development, Marth (*1*) observed some responses which indicated that some chlorinated benzoic acids might, like the methoxy acid, move out of roots. Experiments were undertaken to learn whether any of these chlorinated benzoic acids move out of roots, accumulate in soil, and/or are absorbed by the roots of nearby plants.

Experimental

Some chlorinated benzoic acids induce growth modification by plants to which they are applied (*4*, *5*). In preliminary tests reported herein, both 2,3,6-trichlorobenzoic and 2,3,5,6-tetrachlorobenzoic acids, when placed on the stems of bean plants, suppressed growth and induced development of leaves that were modified in shape. Furthermore, treatment of these plants induced nearby untreated plants to respond by development of trifoliate leaves that were also modified in shape. These modified leaves on the nearby plants were typical of the modified ones on plants to which the acid was applied directly. Figure 1 shows this type of leaf modification due to application of 2,3,6-trichlorobenzoic acid. Other chlorinated benzoic acids, used in these tests, which induced from very slight to marked modification of growth of bean plants—depending upon the molecular configuration of these substances—while failing to affect the growth of nearby untreated plants were: 2,5-dichloro-, 2,3,4-trichloro-, 2,3,5-trichloro-, 2,4,5-trichloro, 2,3,4,5-tetrachloro-, and pentachlorobenzoic acids. As 2,3,6-trichlorobenzoic acid induced a very marked response in nearby untreated plants, it was selected for further experimentation.

Procedures

To learn whether 2,3,6-trichlorobenzoic acid moved from roots, young bean plants were treated with the acid in lanolin and grown in beakers containing aerated tap water (*3*). (The stimulus that was translocated within plants and moved out of roots is referred to here as 2,3,6-trichlorobenzoic acid. However, the acid may have been structurally altered by the plant to form another compound that possessed regulating activity.) Seventy-five to 100 γ of the acid was applied to each primary leaf. After 3 days, the treated plants were removed from the water and replaced with untreated ones. Two days later these untreated plants had developed typically modified leaves, thus demonstrating movement of the acid from the roots of the treated plants.

As the chlorinated acid is known to move from one plant to another, the following experiment was carried out to learn whether the acid moved from roots into soil and, if so, how long detectable amounts remained there. Forty young bean plants were selected. Each plant was grown in composted soil in a 3-inch pot. The plants were divided into two groups of equal numbers and approximately 50 γ of the acid in lanolin was applied to the upper surface of one primary leaf of each plant in one group. The circular area covered was approximately 0.5 sq. cm. and was bisected by the mid-rib about 5 mm. from the petiole attachment. Plants in the other group were not treated and were designated controls.

One week after application of the acid, roots of plants in each group were

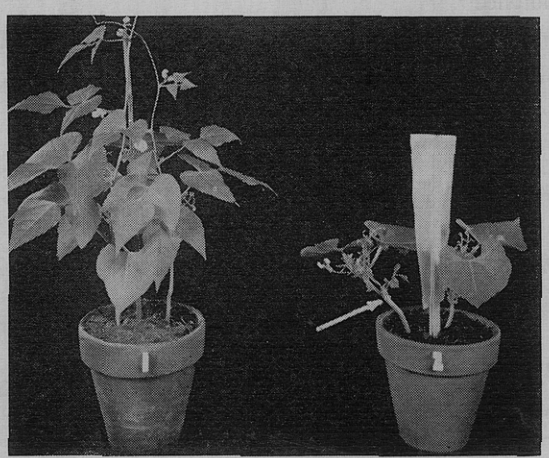

Figure 1. Growth comparison of treated and untreated bean plants

Fifty micrograms of 2,3,6-trichlorobenzoic acid applied to the stem of one plant in pot 2 (arrow) greatly reduced growth of plant. Growth of untreated plant in pot 2 was also greatly reduced by movement of compound from treated plant to untreated through the roots. A polyethylene sheet prevented contact between treated and untreated plants in pot 2

separately shaken free of soil and discarded. The soil was sifted through a 2-mm. mesh screen to remove most of the remaining roots and repotted. Four bean seeds were planted in each pot. Ten days later all plants in soil in which treated plants had grown previously, showed marked modification of the trifoliate leaves. These plants and the controls were then removed, the soil was screened again as described, and a second group of bean plants was grown in the soil. Only 19% of those plants grown in soil which had previously contained the acid showed any effect, and this was slight and temporary. During these experiments the controls showed no effect.

Three additional experiments were carried out during the summer and similar results were obtained. These results show that some of the acid moved or was transferred from roots of the treated plants into soil surrounding them and that the amount in the soil decreased markedly within 2 weeks.

The following experiment indicates that 2,3,6-trichlorobenzoic acid moved from one plant to another through the roots and not by evaporation and movement through the air. Six pairs of young bean plants, each pair grown with its roots in soil in a 3-inch clay pot, were selected for size and uniformity. Stems of plants in each pair were approximately 1 inch apart. Another group of 12 comparable plants, each plant with its roots in a separate pot, was selected. Plants in this second group were arranged so that their stems also were approximately 1 inch apart. A small sheet of polyethylene was then placed between the plants of each pair to prevent contact of the above-ground parts. One plant of each pair was treated by placing about 25 γ of 2,3,6-trichlorobenzoic acid in lanolin on the inner surface of each cotyledon.

After 5 days, all untreated plants grown with their roots in the pots with treated ones, exhibited the response typical of that induced by the chlorinated benzoic acid. The untreated plant of pairs with their roots in separate pots showed no chemical effect. In a similar experiment, comparable results were obtained by placing approximately the same amount of the chemical in a band around the first internode of one plant of each pair.

Two additional experiments were carried out to test further whether vaporization rather than movement out of roots was responsible for the responses observed. In the first of these, four glass rods were placed about 1 inch from a young bean plant. Wire screen was used to enclose a portion of each rod (Figure 2) so the plant could not touch that portion. A mixture containing about 50 γ of the chlorinated benzoic

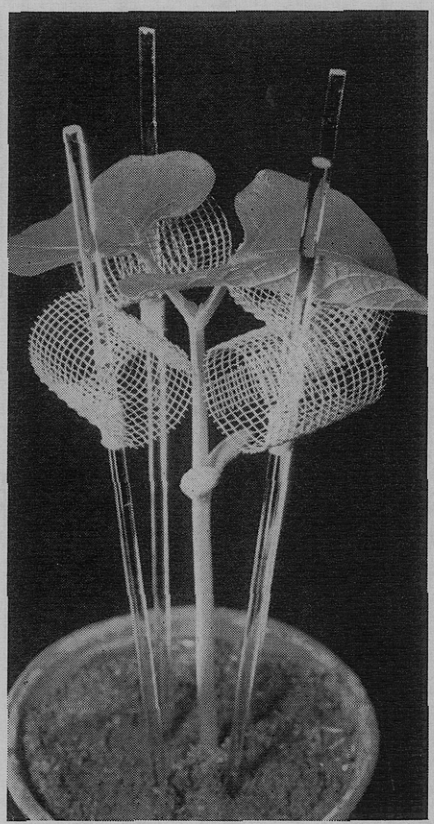

Figure 2. Setup for detecting evaporation of 2,3,6-trichlorobenzoic acid from a mixture of acid and lanolin

Bean plant surrounded by four glass rods—a portion of each rod treated with the acid mixture; the treated portions of the rods are protected with wire screens

acid in lanolin—a mixture containing 1% of the acid—was applied in a narrow band around each enclosed portion. Each plant was thus surrounded with four rods bearing a total of approximately 200 γ of the acid. None of the eight plants tested showed growth modification, indicating that a detectable amount of the compound did not evaporate from the lanolin mixture.

In the second test, filter paper disks were impregnated with a solution of the acid in 95% ethyl alcohol (*2*). Each impregnated paper was fastened near the top of a cellophane bag containing a young bean plant. The bags (four per treatment) were sealed and placed in darkness for 48 hours at 80° to 85° F. The plants were then removed from the bags, placed in a greenhouse, and checked periodically for growth responses. Plants in bags containing 200 γ of the acid on filter paper showed no visible response, whereas all those in bags with 500 and 5000 γ per filter paper developed modified leaves. Evaporation of the acid did not cause the responses observed in other experiments reported here, as only 200 γ or less of the compound was used per plant in these tests.

An experiment was performed to determine the time required for a leaf of a bean plant to absorb and translocate to its stem sufficient acid to effect a detectable root transfer of the regulator to a nearby plant. Approximately 50 γ of the chlorinated acid in lanolin was applied to one primary leaf of each plant. Each treated plant was grown in a pot together with an untreated one. The treated leaves were removed from each of four plants immediately after application of the chemical. At subsequent 2-hour intervals, an additional group of treated leaves was removed. Sufficient acid was absorbed within 4 hours to induce a typical response in 25% of the nearby untreated plants. When the treated leaves were left attached longer, all nearby plants clearly showed the response.

2,3,6-Trichlorobenzoic acid apparently moved from the roots of plants of several genera into the nearby roots of plants of other genera. Snap bean, cucumber, sunflower, barley, and cornseeds were planted together as pairs of the different genera in soil in 3-inch pots. Approximately 50 γ of the acid was applied as a lanolin mixture to the stem of one dicotyledonous plant, or near the tip of the first leaf of one monocotyledonous plant of each pair. The treated plants were thus grown in pots together with untreated ones of a different genus, but in each instance the treated plant was separated from the untreated one by polyethylene. Formative effects developed by the dicotyledonous plants indicated that the chlorinated benzoic acid moved from bean to sunflower and cucumber, from sunflower to cucumber and bean, from cucumber to bean and sunflower, from barley to bean, and from corn to bean. In similar experiments detectable amounts of α-methoxyphenylacetic acid did not transfer from corn to bean plants (*3*).

Acknowledgment

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