Polarity of Thiamine Movement through Tomato Petioles

STEFAN P. KRUSZEWSKI AND WILLIAM P. JACOBS

Department of Biology, Princeton University, Princeton, New Jersey 08540

ABSTRACT

Thiamine-\( ^{14}\)C moved through petiolar sections of \textit{Lycopersicon esculentum} Mill. var. Michigan State Forcing with striking similarity in kinetics to auxins and gibberellic acid moving through similar sections of other green plants. Thiamine moved with strong basipetal polarity, at a velocity of 3 to 5 mm per hour, and emerged unchanged into the basal receiver agar block, judging by chromatography. This lends support to the hypothesis that polar movement is a property of several classes of plant hormones, rather than being restricted to the auxins (as previously believed).

Multicellular plants show many polar phenomena during development and regeneration (e.g., regeneration of roots at the basal or "root" end of an excised stem section). Some of these phenomena have been explained as being manifestations of the movement of the plant hormones known as auxins, which have been known to move polarly since 1928 (10, 19). After it was shown (7, 8, 11, 16) that even the synthetic auxin-type herbicide 2,4-D moved through excised shoot tissues with a polarity like that of the endogenous auxin IAA, other classes of hormones were tested. Just recently, \( \text{GA}_3 \) was found to move with a polarity very similar to IAA through \textit{Coleus} petioles and with opposite polarity to IAA through corn root sections (12–14). Although early reports claimed polar movement of abscisic acid and a cytokinin (1, 4), the more quantitative later studies found sizeable movement in both directions, with no clear sign of polarity of these two classes of hormone (5, 9, 18). The use of different plant species and of different cytokinins might explain some of the differences in results.

The polar movement of plant hormones should have strong selective value and should consequently be characteristic of several classes of hormones. The current situation (in which \( \text{GA}_3 \) is the only hormone, in addition to the auxins, that shows statistically significant polar movement) is not a fully satisfactory confirmation of this hypothesis, because many physiological effects of gibberellin are closely allied to those of auxins. We now report that the very different hormone thiamine also moves with a very strong basipetal polarity through petioles.

Thiamine has been considered to fulfill the requirements for a hormone in higher plants on the basis of research done several decades ago (2, 3). In order to grow indefinitely in aseptic organ culture, excised roots of many species require thiamine in minute amounts. Thiamine was shown to be present in green leaves of intact plants and to accumulate above a zone of killed tissue that was interposed between the leaf and the root system. Such observations were considered as confirmation of the hypothesis that in the intact plant thiamine was synthesized in the leaves and moved from there to the roots, which were unable to synthesize it but needed it for development. We could find no literature on the movement of thiamine through classical "transport sections."

So that the results would be more useful for comparisons, we used the same general methods used earlier in this laboratory for studies of the movement of auxins, cytokinins, and \( \text{GA}_3 \) (10–14, 16, 18). Sections 5.1 mm long were excised with a double-bladed cutter from the middle of petioles of leaves of tomato (\textit{Lycopersicon esculentum} Mill. var. Michigan State Forcing). Tomato was used because of the numerous reports that its excised roots need thiamine for continued growth in culture. The excised petiolar sections were placed horizontally between donor and receiver blocks of 1.5\% agar (see inset diagram of Fig. 1). Donor blocks contained thiamine, with stated specific radioactivity of 18.9 mc/mmole labeled with \( ^{14}\)C in the thiazole 2 position, at a concentration of 15 \( \mu\text{m} \). A donor block was placed against the apical ("leaf blade") end of the petiolar section when movement in the basipetal direction was being tested (as in inset diagram), and against the basal cut end (stem end) when acropetal movement was being tested. These transport systems were placed on glass slides in moist Petri dishes which were placed in a dark incubator at 26 to 27 C. After 2, 3, or 4 hr the receiver blocks were removed and each was placed in a separate glass scintillation vial containing 15 ml of standard scintillation fluid (25\% ethanol in toluene-PPO-POPOP) and counted for 10 min. Background counts from plain agar blocks that had been put on glass slides in Petri dishes held under the same conditions as the "transport" sections were subtracted from the experimental counts. Five samples were used for each data point. Assignment of petiolar sections to acropetal versus basipetal treatments was by a mathematically random method.

Thin layer chromatograms were used to check the purity of the \( ^{14}\)C-thiamine added in the donor blocks and recovered in basal receiver blocks. Silica gel with UV\(_{365}\) fluorescent indicator was used, with a solvent system of water-pyridine-acetic acid (79:19:2) (17). Thiamine without a radioactive label was used to check the \( R_f \) on the chromatograms; and an amount of \( ^{14}\)C-thiamine providing counts equivalent to those found in the receivers at 4 hr was added directly to plain agar blocks and extracted in parallel with the basal receiver blocks as a control for artifacts resulting from the extraction procedure.

The time course of accumulation in receivers of \( ^{14}\)C from thiamine added in the donor is shown in Figure 1. The kinetics are much like those previously described for the movement of IAA and \( \text{GA}_3 \) through petiolar sections: there is strong basip
The time courses in the dark showed linear relations as shown in Figure 1. Three time courses run in ordinary laboratory lighting by Kruszewski (unpublished data) showed much more basipetal movement, suggesting that light increases movement in the normal polar direction, as was recently reported for IAA (15). Statistical tests on the 4 hr values showed that the basipetal receivers were significantly different from the acropetal ones, and that acropetal values were significantly larger than background.

There are now three different types of plant hormones—auxins, GA₃, and thiamine—that have been shown in quantitative tests, supported by statistical analysis, to move with strong polarity through plant tissues. To this list must be added cyclic AMP, recently shown by Gordon et al. (6) to move with basipetal polarity through coleoptile sections.

**Acknowledgments**—We thank H. B. Suthers for assistance. The contributions of S. F. Kruszewski were incorporated in his May 1973 experimental senior thesis at Princeton University.

**LITERATURE CITED**


