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## EFFECT OF TRIODOBENZOIC ACID AND MALEIC HYDRAZIDE UPON THE TRANSPORT OF FOLIAR APPLIED CALCIUM AND IRON<sup>1,2</sup>

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The lack of mobility of foliar applied calcium in a downward direction has been reported (1, 2, 3, 7, 8) and it has been suggested that polarity might play a role in this movement (3). By suspending polarity by mild anesthesia (10) with di-ethyl-ether the immobility of calcium was indeed overcome and calcium was translocated from the site of application on the leaves down to the roots (3).

These reports are of considerable importance with regard to the increasing use of foliar sprays. It was, therefore, of interest to study the effect of substances, which are believed to interfere with the polar movement of auxins, upon the transport of immobile elements. Such a substance, triiodobenzoic acid (TIBA) (5, 6), was, therefore, chosen for these experiments. For purposes of comparison a second growth inhibitor, maleic hydrazide (MH), was also tested.

Tomato plants and one-year-old apple trees were sprayed with either a 100-ppm solution of TIBA or a 100-ppm solution of MH, each solution containing 0.05% of a spreader (TRITON B-1956). One terminal leaf of each plant was then treated with 0.1 ml of Ca<sup>45</sup> (0.8  $\mu$ c per ml). The Ca<sup>45</sup> was dropped on the leaf surface with a micropipette and was absorbed there quickly. The same amount of radiocalcium was applied to plants which had received no TIBA or MH pretreatment, but only a spray with water containing the same amount of the spreader as above. A few days after the application of Ca<sup>45</sup>, leaves located below the treated ones were collected from each apple tree, while from the tomato plants the lower part of the stem was sampled. These samples were ashed, and the radioactivity counted in a thin end-window Geiger-Mueller counter.

Results are presented in table I, and they confirm

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TABLE I  
EFFECT OF TIBA AND MH UPON THE BASIPETAL  
MOVEMENT OF Ca<sup>45</sup> IN APPLE TREE SEEDLINGS  
AND TOMATO PLANTS

SPECIES	PART OF PLANT TESTED	TREATMENTS (COUNTS/MIN × G FRESH MATTER)		
		Ca <sup>45</sup>	MH + Ca <sup>45</sup>	TIBA + Ca <sup>45</sup>
	<i>leaf no. below treated leaf</i>			
Apple (5 days after treatment)	3	30	136	21,930
	5	0	64	21,273
	7	0	52	500
	9	0	40	460
	20	0	0	117
Tomato (2 days after treatment)	lower part of stem	0	...	2,000

the limited transport of calcium in a basipetal direction. They further demonstrated the considerable promotive effect of TIBA on the downward transport of this element. MH on the other hand was much less effective in this respect.

A similar effect of TIBA was found upon the transport of iron, which is also frequently immobile. No radioactive material was employed in the experiments with iron. The effect of TIBA and MH upon the transport of iron was followed by the synthesis of chlorophyll in chlorotic leaves of peach and plum trees growing in high-lime soils. Iron sulphate, when sprayed on such chlorotic leaves, produces only localized green spots of chlorophyll (fig 1) as compared to a solid green surface when pretreated with TIBA, i.e., TIBA caused the iron to spread inside the leaf from the point of penetration, thus becoming available to all leaf parts for chlorophyll synthesis.

Quantitative data on the effect of TIBA upon the transport of iron and its availability for chlorophyll formation were obtained by analysis of the chlorophyll content of the leaves. Chlorotic peach and plum trees were sprayed with 100-ppm solutions of TIBA and MH respectively. The pretreated plum trees were then sprayed with a 0.5% solution of iron sulphate. In case of the pretreated peach trees the additional iron spray was not applied to the whole trees (as with plum trees), but only to the basal part of branches (5 branches per treatment). For analysis leaves from the terminal part of these branches were sampled, at a distance of about 30 to 40 cm from the treated leaves. For comparison leaves were collected from branches which received TIBA and MH pretreatments but no iron treatments, as well as leaves which received an iron spray with no TIBA and MH pretreatments. For chlorophyll determinations the leaves were extracted with ethanol, and the extracts compared with a bichromate-copper standard (4). The results presented in table II give indirect evidence of increased transport of iron under the influence of TIBA.

It is seen from table II that TIBA alone has a certain promoting effect upon the formation of chlorophyll. This promoting effect may be brought about by a freeing of immobilized iron by TIBA. A similar mechanism was indicated already on the basis of results obtained by Weinstein et al with chelates (9).

The introduction of iron into TIBA pretreated

TABLE II  
EFFECT OF TIBA AND MH UPON THE TRANSPORT OF IRON IN CHLOROTIC PEACH AND PLUM LEAVES AS MEASURED BY THE SYNTHESIS OF CHLOROPHYLL 35 DAYS AFTER TREATMENT

TREATMENTS	PEACH *	PLUM **
	VAR. C. O. SMITH	VAR. KELSEY
Control	0.129	0.078
MH	0.136	....
TIBA	0.151	0.193
Fe	0.289	0.132
Fe + MH	0.254	....
Fe + TIBA	0.463	0.351
Standard error	0.008	0.019

\* Chlorophyll percent of fresh matter of leaves distal to the leaves which received an iron spray.

\*\* Chlorophyll percent of fresh matter of leaves which received an iron spray.

leaves caused a considerable increase in chlorophyll formation, as compared with the effect of TIBA alone. Based on the above results (fig 1) it can be suggested that TIBA enables the penetrating iron to spread inside the leaves, keeping the iron from becoming immobilized and thus remaining available for chlorophyll synthesis. The experiment with peach trees shows further that TIBA promotes also, similarly to calcium, the transport of iron over appreciable distances, from the site of application at the basal leaves to the more remote apical leaves.

MH, on the other hand, had no effect upon the transport of iron (as indicated by the lack of chlorophyll synthesis), although it had, however only limited, an effect on the transport of calcium.

Under conditions of similar concentrations of TIBA and MH it is as yet difficult to say whether the effects of these substances differ only quantitatively at the level used in these experiments, or whether different mechanisms are involved through which these substances affect the plant.

The above results are of special interest with regard to TIBA as a growth inhibitor. The mobility and downward transport of nutrients might well be related to the suggested suspending effect of TIBA upon polarity, its interference with the polar movement of auxin, and its destroying effect upon apical dominance.

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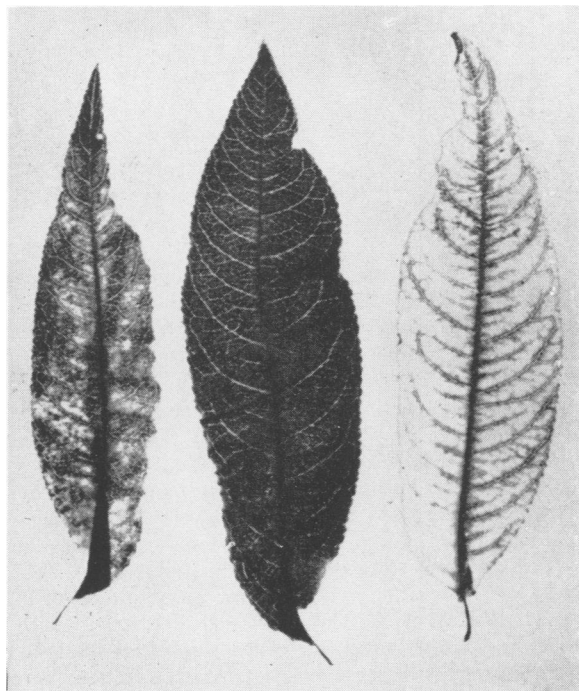


FIG. 1. The effect of TIBA upon the distribution of externally applied iron in chlorotic peach leaves, as measured by the formation of chlorophyll. Left: Fe without pretreatment with TIBA; center: Fe, pretreated with 100 ppm of TIBA; right: control (no treatment).

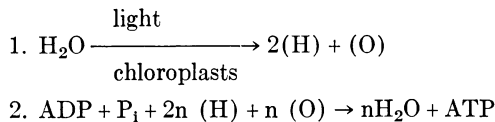
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## THE ACTION SPECTRUM FOR PHOTOSYNTHETIC PHOSPHORYLATION BY SPINACH CHLOROPLASTS<sup>1,2,3</sup>

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Photosynthetic phosphorylation by chloroplasts was first described by Arnon et al (1). It is believed to involve the following reactions:



Reaction 1 is presumably the only reaction where light is involved in photosynthesis, in the Hill reaction, and in photosynthetic phosphorylation. Under this hypothesis the action spectra of the three processes should be identical. It has previously been shown that the action spectra for photosynthesis and for the Hill reaction do indeed correspond to each other and to the absorption spectrum of chlorophyll (2). It seemed of interest to determine whether the action spectrum for photosynthetic phosphorylation would similarly relate to the absorption spectrum of chlorophyll.

The experiments were performed with the high irradiance spectrograph of the U. S. Department of Agriculture at Beltsville, Maryland. Chloroplasts and the reaction mixture were prepared as previously described (3) with phenazine methosulfate and magnesium as the only cofactors. Phenazine methosulfate has an absorption peak at 388  $m\mu$ , and only very slight absorption above 400  $m\mu$ . It has previously been found (3) that photosynthetic phosphorylation

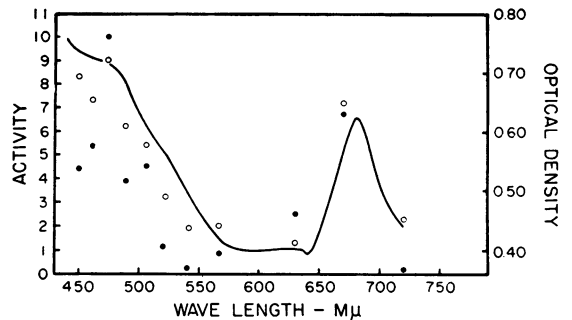


FIG. 1. Action spectrum for photosynthetic phosphorylation. The solid line represents the absorption spectrum of the chloroplast suspension (scale on right). The points represent the activity of the preparation expressed as micromoles phosphate taken up per mg chlorophyll per hour per milliwatt/cm<sup>2</sup> of light energy at the particular wave length (scale on left). The solid circles were obtained in one experiment, with .114 mg chlorophyll per 25-ml Erlenmeyer flask; the open circles from a 2nd experiment with 0.076 mg chlorophyll per flask. One and five-tenths ml of reaction mixture were spread over the bottom surface of approximately 10 cm<sup>2</sup>. The flasks were placed in a rectangular, glass bottom, Warburg bath kept at 15° C and were shaken continuously at a small amplitude. The light was directed at the flask bottoms with the aid of a mirror. Reactions were run under N<sub>2</sub> for 30 to 60 minutes.

The reactions represented in this figure were run with the spectrograph set as a single prism instrument, with the slit width equal to 1.5 times the separation of wave length positions of adjacent flasks between 450 and 570  $m\mu$ .

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