

Zeitschrift: Berichte der Schweizerischen Botanischen Gesellschaft = Bulletin de la Société Botanique Suisse

Herausgeber: Schweizerische Botanische Gesellschaft

Band: 80 (1970)

Artikel: Studies on the metabolic interaction of triiodobenzoic acid during the transport of externally applied indole acetic acid

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DOI: <https://doi.org/10.5169/seals-56315>

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Studies on the Metabolic Interaction of Triiodobenzoic Acid During the Transport of Externally Applied Indole Acetic Acid

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Manuscript received March 26, 1970

According to Kuse (1953) 2,3,5-triiodobenzoic acid (TIBA) has a blocking effect on the transport of indole acetic acid (IAA). From earlier experiments (Vardar, 1959) we concluded that TIBA causes a metabolic change of IAA. Several workers (Goldsmith and Thimann, 1962; Christie and Leopold, 1965; Winter, 1967) observed a considerable accumulation of IAA in TIBA treated plant tissues and attributed it to the immobilization effect of TIBA on IAA transport. In the present paper the effect of TIBA during transportation of IAA-C¹⁴ applied externally to young seedlings is studied.

We used *Helianthus annuus* hypocotyls grown and treated with IAA-C¹⁴ according to the procedures in our previous work (Vardar, 1959). The radioactive auxin was provided by the Waltham Tracer Laboratory; its specific activity was 1.1 mc/mM and the concentration 0.11 mg/ml; the amount of auxin applied was 2.2 mg and the activity applied 100 × 13 CpM. The extractions obtained from hypocotyl sections according to Nitsch and Nitsch (1955) were subjected to paper and thin layer chromatography and the radioactive counting of these was done with a Nuclear Chicago M-5 Gas Flow Counter. Paper chromatograms were run on Whatman No. 2 paper in isopropanol-ammonia-water. For thin layer chromatography the Desaga-apparatus and Kaldey's technique (1964) were used. Rf values, UV absorption traces, colour spots obtained with Salkowski reagent and autoradiographic results were compared and from them the amounts of radioactivity in various spots determined.

The results from the Salkowski colour evaluation test and the autoradiographic determinations on the chromatograms (fig. 1) show that the externally applied IAA-C¹⁴ is at least partially transported as IAA in hypocotyls. It passes even under the TIBA paste ring proving thereby that TIBA does not block its transport. These results support our earlier view (Vardar, 1959) and show that IAA-C¹⁴ is at least partly responsible for the radioactivity observed in the hypocotyl tissue under the TIBA paste ring. At the same time a comparison of the radioactivity evaluations in thin layer chromatograms (table 1) shows that in the various spots the activity was generally higher in TIBA treated hypocotyl tissues than in the controls. This supports the view that the TIBA treatment enhances the rate of accumulation of externally applied IAA-C¹⁴ within the hypocotyls as pointed out by various workers (Goldsmith and Thimann, 1962; Christie and Leopold, 1965; Vardar, 1959; Winter, 1967).

To explain the physiological phenomena occurring during growth in length (induced by IAA) it is necessary to identify the substances derived from IAA. Several investigations (Andreae and Good, 1955; Klambt, 1960; Rausendorff-Bargen, 1962) show that IAA applied externally to the coleoptile is transformed into various substances depending on the concentration and the length of time of application. Klambt (1960) found 3-indole aspartic acid, 3-indole carboxylic acid and

3-indole acetic acid as the characteristic substances. Our studies confirmed these results, three spots on the chromatograms corresponding to 3-indole aspartic acid (0.07), 3-indole carboxylic acid (0.25) and 3-indole acetic acid (0.35); in addition to these we found a fourth radioactive derivate, the Rf value of which (0.45) corresponds roughly to 3-indole propionic acid or 3-indole butyric acid (Audus, 1959). With UV we noticed a derivate which approximately corresponds to 3-indole acetamide (Rf 0.80) which was also mentioned by Klambt; it did not show any radioactivity in our experiments.

In conclusion we may say that IAA-C¹⁴, applied externally to young green seedlings of *Helianthus annuus*, is transformed into different metabolic derivatives during the transport and that TIBA accelerates these changes. This might explain the action of TIBA on plant growth but additional biochemical experiments are necessary before drawing definite conclusions.

Table 1
Proportional radioactivity in different zones of chromatograms

Spot Rf		TIBA-treated		Control	
		upper part	lower part	upper part	lower part
0.07	3-indole aspartic acid	295	82	180	30
0.25	3-indole carboxylic acid	45	18	28	15
0.35	3-indole acetic acid	54	21	50	12
0.45	3-indole propionic or 3-indole butyric acid	85	15	64	7

Zusammenfassung

C¹⁴-markierte Indolylessigsäure (IAA, Keimlingen von *Helianthus annuus* äusserlich verabreicht) wird während des Transports in verschiedene verwandte Verbindungen (Tab. 1) umgewandelt. Trijodbenzoesäure (TIBA) bewirkt eine Anreicherung im Hypokotyl und intensiviert die Umwandlungsreaktionen.

This work was supported by the Turkish Atomic Energy Commission.

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