TABLE I

No.	Compound Name	m.p., °C. corr.	Activity as per cent. of oestrone activity
І. II.	<ol> <li>1-Keto-1, 2, 3, 4-tetra- hydrophenanthrene</li> <li>3, 4-Dihydrophenan-</li> </ol>	94–95	0.56
III.	threne-1, 2-dicar- boxylic anhydride <sup>1</sup> Phenanthrene-1, 2-	263.5-264.5	weak*
IV.	dicarboxylic anhy- dride <sup>1</sup> Phenanthrene-3, 4-	311–312	weak*
v.	dicarboxylic anhy- dride <sup>2</sup>	253.5-254	Inactive in $100 \gamma$ doses
۷.	1 <sup>1</sup> , 3 <sup>1</sup> -Diketo-1, 2-cy- clopenteno-phenan- threne <sup>2</sup>	Dec. 240-245	0.22
VI. VII.	<ol> <li>3<sup>1</sup>-Diketo-3, 4-cy- clopenteno-phenan- threne<sup>2</sup></li> <li>3-Dimethyl-1, 4, 9, 10, 11, 12-hexahy-</li> </ol>	201.4-202	Inactive in 50γdoses
VIII.	drophénanthrene- 11, 12-dicarboxylie acid <sup>3</sup>	176–177 95–96	Inactive in 100 γ doses Inactive in 100 γ doses
IX.	Monomethyl ester of VII <sup>3</sup>	157 - 158	Inactive in 100γdoses
X.	Dimethyl ester of VII <sup>3</sup>	93.5-94	Inactive in 1000 y doses
XI.	2, 3-Dimethyl-1, 4, 11, 12, 13, 14-hexahy- drochrysene-13, 14- dicarboxylic anhy- dride <sup>3</sup>	196–196.5	Inactive in $100 \dot{\gamma}$ doses
XII. XIII.	<ul> <li>9-Methoxyphenan- threne-1, 2-dicar- boxylic anhydride</li> <li>6-Hydroxy-1, 2, 3, 4, 9, 10, 11, 12-octa-</li> </ul>	249-250	0.11
XIV. XV.	hydrophenanthrene- 11, 12-dicarboxylic anhydride <sup>4</sup> Methyl ether of XIII <sup>4</sup>	160–160.5 159–159.5	0.56 0.48
XVI. XVII.	<ul> <li>6, 7-Dihydroxy-1, 2, 3, 4, 9, 10, 11, 12- octahydrophenan- threne-11, 12-dicar- boxylic anhydride<sup>4</sup> Dimethyl ether of XV</li> <li>6-Methyl-7-hydroxy-1, 2, 3, 4, 9, 10, 11,</li> </ul>	147.5–148.5 146.5–147	1.58 1.47†
	12-octahydrophe- nanthrene-11, 12- dicarboxylic anhy- dride <sup>4</sup>	134.5–135.5	0.55

\* See text.

† Estimated from data on effects of a single injection.

methods is indicated by our data on substance I. Butenandt<sup>6</sup> obtained positive results when 70 mgms of this substance were injected in a single subcutaneous dose into spayed mice. Cook, Dodds et al.<sup>7</sup> observed XIII, which showed definite activity in 25  $\gamma$  dosages, gave completely negative results upon subcutaneous injection in 100  $\gamma$  doses.

And substance XV was inactive in 1 mgm subcutaneous dosages (3 injections in 12 hours), in spayed rats on the basis of vaginal cornification, whereas a 14 mgm dosage in a single subcutaneous injection gave a positive result in rats. It must be emphasized that the method of administration seems to be the all-important factor in determining activity (cf. Cook, Dodds et al.).

In addition to their oestrogenic effects, certain of these compounds have shown definite activity in the immature rat test, in the augmentation of ovarian growth in rats following FSH stimulation and in the sterilization of rabbits. The details of these experiments will be published elsewhere.

> GREGORY PINCUS N. T. WERTHESSEN

BIOLOGICAL LABORATORIES, HARVARD UNIVERSITY

## PLANT GROWTH EFFECTS OF HETERO-AUXIN APPLIED TO SOIL AND PLANTS

 $\beta$ -INDOLYL acetic acid (heteroauxin) in extremely dilute concentrations is known to function as a growthpromoting hormone in plants. The crystalline material was synthesized in this laboratory according to the method of Majima and Hoshino.<sup>1</sup> The indolyl acetic acid used had a melting point of 164-166° C., 8.04 per cent. nitrogen and a neutral equivalent of 176.3; the corresponding theoretical values being 165° C., 7.99 per cent. and 175.1, respectively. An aqueous solution, 1/15,000, was used to determine its effect on growth, rooting, proliferation and bending. Actively growing seedlings were used to detect the effect of heteroauxin during the grand period of growth. One hundred and eighteen day old potted stock seedlings having approximately equal stem lengths were divided into two series. To each pot of the first series 15 cc. of the indole acid solution was added. Each pot of the second series was treated only with an equal volume of water. All the plants were kept under greenhouse conditions and were watered daily throughout the experimental period.

The effect of the indole acid on stem elongation was manifest at the end of the first experimental day, and reached its maximum on the sixth day following its administration (Table I). Curling of the cotyledons on treated plants appeared at the end of the second day, but by the end of the sixth day this effect began to disappear and by the tenth day had disappeared

<sup>1</sup> Majima and Hoshino, Berichte, 58: 2042, 1924.

<sup>&</sup>lt;sup>6</sup> A. Butenandt and G. Schramm, *Ber.*, 68 B: 2303, 1935. <sup>7</sup> J. W. Cook, E. C. Dodds, C. L. Hewett and W. Lawson, *Proc. Roy. Soc. Lond.*, B 114: 272, 1934.

entirely. The differences of stem length between control and treated groups also began to diminish after the sixth day, and by the end of the tenth day no apparent differences between the two groups of seedlings were to be found. The data are statistically significant. It is striking to note the transitory effect of heteroauxin, the action being confined chiefly to the early phase of the grand period of growth with the dosage used directly upon the soil.

TABLE I COMPARATIVE GROWTH OF STOCK SEEDLINGS (Matthiola incana) TREATED WITH  $\beta$ -INDOLYL ACETIC ACID WITH UNTREATED CONTROL SEEDLINGS. STEM LENGTH IN MM.

Day	Control seedlings	Treated seedlings
$\begin{array}{c} 1 & \dots & 2 \\ 2 & \dots & 3 \\ 4 & \dots & 5 \\ 5 & \dots & 6 \\ 6 & \dots & 6 \\ 7 & \dots & 8 \\ 9 & \dots & 9 \\ 10 & \dots & 11 \\ 11 & \dots & 1 \end{array}$	$\begin{array}{c} 16.0 \pm 0.2 \\ 17.0 \pm 0.5 \\ 19.0 \pm 0.5 \\ 19.5 \pm 0.0 \\ 25.5 \pm 0.0 \\ 29.0 \pm 0.1 \\ 30.0 \pm 0.4 \\ 35.0 \pm 0.4 \\ 43.0 \pm 0.3 \\ 46.0 \pm 0.1 \\ 50.0 \pm 0.1 \end{array}$	$\begin{array}{c} 16.0\pm0.5\\ 21.0\pm0.5\\ 26.5\pm0.0\\ 30.5\pm0.4\\ 38.0\pm0.0\\ 40.0\pm0.5\\ 41.0\pm0.2\\ 43.0\pm0.3\\ 45.0\pm0.3\\ 45.0\pm0.3\\ 48.0\pm0.1\\ 51.0\pm0.0 \end{array}$

When localized areas on young healthy plants were repeatedly treated with heteroauxin a definite symptom complex was observable, the speed and intensity of the responses varying with the concentration and amount of the heteroauxin solution used. The sequence of events was, first, a bending away from the treated area due to accelerated growth in the treated region shown in stems, petioles and leaves. Next, swelling due to cell enlargement was observable in and about the treated regions in one to two days. Finally a slight blanching accompanied by proliferation became noticeable, while the stems and petioles continued to increase in thickness. Root primordia appeared a few days later and then gradually developed into anatomically normal roots. At this stage stems had doubled in thickness, become bronzed and sometimes fissured due to excessive growth of medullary tissues. These results confirm those obtained by Hitchcock,<sup>2</sup> using indole-3n-propionic acid in somewhat greater concentrations on tomato, African marigold, tobacco, dahlia and buckwheat.

The fact that the hormone-like  $\beta$ -indolyl acetic acid occurs in a wide variety of animal tissues (lung, thyroid, thymus, pancreas and gonads) as well as plant tissues (fungi, bacteria and angiosperms) suggests that it may have a common function especially in relation to cellular metabolism. It seems especially important to meristematic and mitotic activity related to differentiation.

> W. F. LOEHWING L. C. BAUGUESS

STATE UNIVERSITY OF IOWA

## SCIENTIFIC APPARATUS AND LABORATORY METHODS

## **RECONDITIONING APHIDS FOR STUDY**

IN 1903 W. T. Clarke, of the University of California, published an article in the Canadian Ento $mologist^1$  in which he described a number of new and well-known species of aphids. His work was of the pioneering, fundamental type and was quite typical of the times. Some of his specimens were mounted in balsam on microscopic slides, and these were subsequently lost or destroyed, so that no authentic types remained. This loss has resulted in much confusion in the study of the California Aphididae in recent years. Attempts have been made to restore certain of the species named by Clarke, but these efforts have been based largely upon conjecture and circumstantial evidences only. I have had occasion to discuss the probable identity of his types with Clarke, whom I knew well for the last ten years of his life, but his long divorcement from the study of insects had erased any distinct recollections which might serve to reestablish his species. In 1930 I discovered in the attic storeroom of Agriculture Hall a small box containing some thirty small homeopathic vials of aphids bearing labels in the handwriting of Clarke. This find was

<sup>1</sup> Can. Ent., 35: 247-254, 1903.

extremely interesting and important to me, but careful examination revealed a sad state of affairs. Although the corks appeared to be tight, the liquid preservative had disappeared completely from all except two or three vials containing only the common, well-known woolly apple aphis, Eriosoma lanigerum (Hausmann). The specimens in the dry containers appeared as mere dust composed of finely shattered bits of legs, antennae, wings and other body fragments. In spite of the fact that one of these vials contained Aphis mori, marked "types," and some other material which might prove to be cotypes of Clarke's lost species, I was convinced that no real information could be salvaged from the find. Nevertheless, the collection was preserved in toto. More recently in clearing aphids in KOH for microscopical study it was found that dried specimens were not only restored to much of the original form, but were also rendered clear and partly transparent. Accordingly, all the vials containing the Clarke specimens were filled with a 10 per cent. solution of KOH and set aside for seven days. To my satisfaction I discovered that what appeared to be a mass of fragments was composed of aphids, many of which were in

<sup>2</sup> Hitchcock, Contributions Boyce Thompson Inst., 7: 87, 1935.