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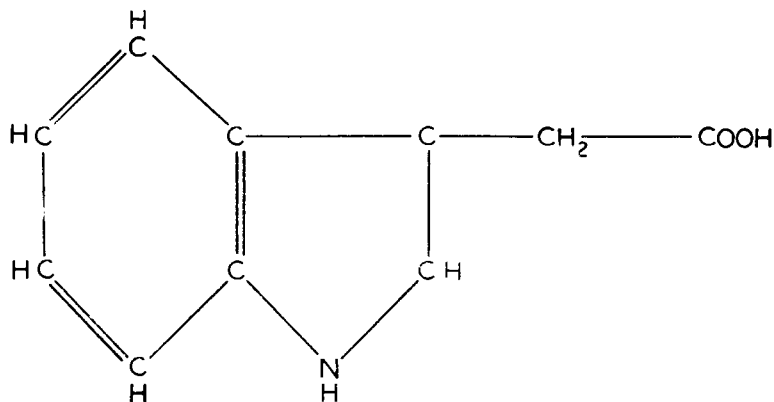
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HISTOLOGICAL RESPONSES OF STOCK (*MATTHIOLA
INCANA*) SEEDLINGS TREATED WITH B-INDOLYL
ACETIC ACID

F. M. TURRELL AND L. C. BAUGUESS

Kögl, Haagen Smit and Erxleben (1934) confirmed the finding of Salkowski and Salkowski in 1895 that urine contained B-indolyl acid, and found that this substance accounted for a portion of the activity of urine in promoting the growth of plants not accounted for by auxentriolic acid and auxenolonic acid. Majima and Hoshino (1924) had already developed a method for the synthesis of B-indolyl-acetic acid so that it became evident to the authors that sufficient amounts of B-indolyl acetic could be made readily available for plant growth hormone studies.

The junior author having worked with the synthesis of indole derivatives for the past few years (Berg and Bauguess, 1932; Bauguess and Berg 1934a, 1934b) undertook and succeeded in the synthesis of B-indolyl acetic acid. Mixtures of the acid in lanolin



paste were active in bringing about the curvature of the *Avena col-coptile*. It had a melting point of 164-166° C, uncorrected, 8.04% nitrogen and a neutral equivalent of 176.3. The corresponding theoretical values were m.p. 165° C, 7.99% nitrogen, and a neutral equivalent of 175.1.

Stock plants were grown from seed in flats in the greenhouse. When these had reached a height of from 1.3 cm to 1.4 cm (nineteen days after planting) one set of plants was watered with 15 cc of distilled water and the other with a solution of 1 mg of B-indolyl acetic acid to 15 cc of distilled water (1:15,000). Photo-

graphs of representative samples of the treated and untreated plants were made ten days later (fig. 1 and 2).

Sections of these stems were made near the tips of ten plants and portions were also cut from central portions of leaves of ten plants. The sections were killed in formalin-acetic alcohol, Rawlin's formula no. 2, dehydrated in a graduated series of butyl alcohol, embedded in paraffin, sectioned and stained in Delafield's haematoxylin and safranin, and mounted on microscope slides. All tissue measurements have been made using these permanent prepared slides.

1. *Effect on stem tissues.*

Table 1. Sizes of cells and tissues in stems of hetero-auxin treated and untreated stock seedlings. Measurements in microns.

Treatment	Stem diameter		Vascular cylinder diameter		Cortex thickness		Cortex cells	Stem
	long	short	long	short	long	short	length	length
Treated	1691.7	1357.1	799.4	632.1	381.0	248.0	230.5	29,600
Untreated	1803.2	1338.5	799.4	520.5	443.0	318.0	172.0	19,200

The treated plants grew much taller (table 1) and produced much thicker leaves which showed a tendency to roll as shown in figures 1 and 2. Microscopic measurements on the stem diameter showed that the long (major) diameter of the treated plants was much smaller than that of the untreated plants but that the short (minor) diameter was approximately the same (table 1).

In the vascular cylinder the long (major) diameters were about the same in treated and untreated plants. However, the treated plants had a significantly larger vascular cylinder when the short (minor) diameter was measured (table 1). However, the cortex thickness of the treated plants was smaller than the untreated when the short (minor) diameter was measured, thus compensating for the larger short (minor) diameter in the vascular cylinder of the treated plant stems (table 1). The long diameter measurement of the cortex indicated that this tissue was narrower in the treated plants than in the untreated (table 1).

As shown in table 1, the cortex cells measured from longitudinal sections of the stem were much longer in the treated than in the untreated. This fact accounts for the greater height of the treated plants. Compare figures 7 and 8.

2. *Effect on leaf tissues.*

The seedlings treated with B-indolyl acetic acid developed leaves having the same upper epidermal thickness as the controls (un-

Table 2. Sizes of cells and tissues in leaves of hetero-auxin treated and untreated stock seedlings. Measurements in microns.

Treated Untreated	Tissue Thickness					Stomatal length	Stomatal width	Palisade cell diameter
	Upper epidermis	Lower epidermis	Palisade	Sponge	Leaf			
	29.2	23.9	45.1	125.2	207.0	30.1	28.9	32.2
Treatment	29.4	21.7	38.3	87.5	177.0	32.4	22.9	25.8

treated), table 2. The lower epidermal thickness was slightly greater (10.1%) in the treated than in the controls (table 2). The thickening of the lower epidermis in the absence of thickening in the upper, in part explains the rolling of the leaves of the treated plants. Palisade, sponge, and leaf thickness were all greater in the treated plants than in the controls (table 2). The greater percentage thickening of the sponge over the palisade may also be responsible for leaf curl. Compare figures 5 and 6. The greater leaf thickness of treated plants is due to greater thickness of the palisade and sponge tissues. While the leaf dimensions were not measured it appears from the measurement of the stomatal width (guard cell + stomatal pore) and from measurement of the palisade cell diameters that the leaf of the treated plants expanded more than the leaves of the untreated plants for the palisade cells of the treated plants were larger in diameter, and the short diameters of the stomata were greater than in the untreated plants (table 2.) The comparison of the palisade cell diameters may be made from the photomicrographs in figures 3 and 4.

DISCUSSION AND CONCLUSIONS

B-indolyl acetic acid (heteroauxin) stimulates the growth of young stock seedlings. The effect of this substance seems to be on the cells of the cortex of the stem where cell length is increased bringing about a taller plant.

The hormone also effected a thickening of the palisade, sponge and lower epidermis of the leaf, though the upper epidermis was unaffected. The sponge of the treated plants increased 43.1% in thickness while the palisade increased but 17.7% in thickness. The greater thickening of the sponge probably is the chief cause of the upward rolling of the leaves of the treated plants. While the thickening of the lower epidermis no doubt contributes some force, it is relatively small, however, as compared with that exerted by the thickening sponge tissue.

Growth hormones such as hetero-auxin are known to diffuse and concentrate in plant organs on the side away from the light and to cause growth in this localized region. As the leaves of the stock

seedlings were exposed to the most intense light on their upper surfaces the hetero-auxin would be expected to concentrate in the lower tissues of the leaves. This condition may account for the observation that no thickening occurred in the upper epidermis while the palisade and sponge showed progressively higher percentages of thickening, and the lower epidermis some thickening. It suggests that the mechanism of mesophyll and epidermal thickening which results in xeromorphic leaf structure when leaves are exposed to intense light may be due to a hormone in low concentration which tends to diffuse away from the most intensely lighted side of the leaf to the less intensely lighted side, thus accelerating the vertical growth of these tissues.

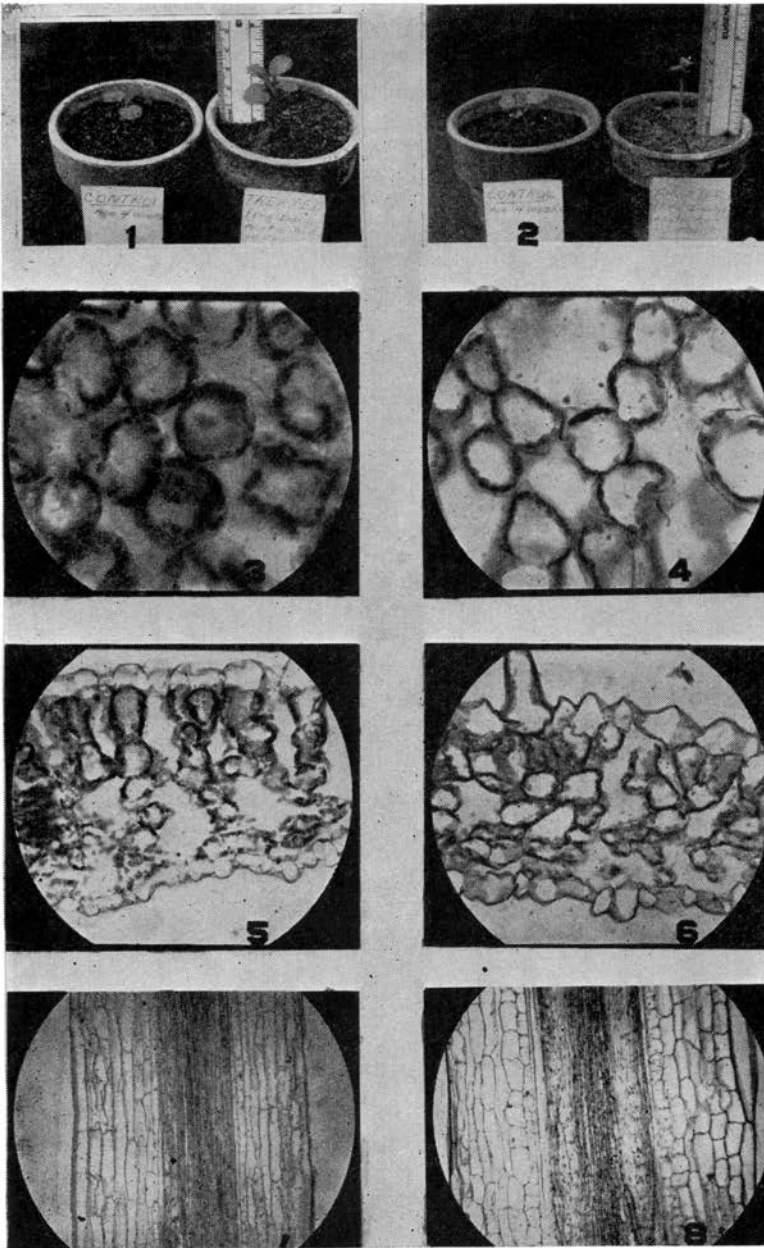
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EXPLANATION OF FIGURES

- Fig. 1 and 2. Photographs of representative B-indolyl acetic acid untreated and treated stock seedlings.
- Fig. 3. Photomicrograph of a tangential section of a leaf of a B-indolyl acetic acid treated stock seedling. Section through the palisade layer. X346.
- Fig. 4. Photomicrograph of a tangential section of the leaf of an untreated stock seedling. Section through the palisade layer. X346.
- Fig. 5. Photomicrograph of a transverse section of a leaf of a B-indolyl acetic acid treated stock seedling. X164.



- Fig. 6. Photomicrograph of a transverse section of a leaf of an untreated stock seedling. X164.
- Fig. 7. Photomicrograph of a longitudinal section of the stem of B-indolyl acetic acid treated stock seedling. X38.
- Fig. 8. Photomicrograph of a longitudinal section of the stem of an untreated stock seedling. X38.