

that in the series of cultures on the 2 per cent glucose medium, at all concentrations of gibberellic acid there was a distinct increase in the rate of heteroblastic development, with the first adult (4-fid) leaf arising several nodes earlier than in the control cultures. Any suggestion that gibberellic acid is a specific 'maturity-hormone' is ruled out at once, however, by the fact that an even greater advance in the rate of heteroblastic development was obtained merely by raising the sugar concentration from 2 to 4 per cent, while furthermore, at the higher sugar concentration, gibberellic acid had no appreciable effect on leaf segmentation.

The results of the present work are certainly opposed to the view that gibberellic acid functions as a youth hormone. The undoubted effects of gibberellic acid on heteroblastic development in the investigations of various authors are more likely secondary consequences of the changes in growth rate, which so frequently follow a supply of this substance. An explanation along these lines has the advantage of accounting for the apparently contradictory results obtained with different species. Thus, the increase in growth vigour following on the supply of gibberellic acid might be expected to increase the rate of heteroblastic development in cases where the appearance of adult characteristics is dependent on the enlargement of the apical meristem⁷ (as in *Marsilea* and presumably in *Eucalyptus*), while an increased utilization of carbohydrate might lead to a delay in the appearance of adult characteristics when their formation is dependent on an increasing accumulation of soluble carbohydrates in the developing organs⁸ (as seems probable in ivy and *Ipomoea*).

The results of Doorenbos and of Frank and Renner can also be explained in a similar way without the necessity for postulating the existence of a youth-hormone⁸. It may, therefore, be concluded that although it is perhaps unlikely that changes in the nutrient balance will account for all types of heteroblastic development, there is no convincing evidence at present in favour of the existence of specific juvenility hormones.

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Gibberellic Acid and Nodulation of Legumes

IN 1958 Thurber, Douglas and Galston¹ reported that potassium gibberellate applied to *Phaseolus vulgaris* plants had a marked reducing effect on the number of nodules formed and in some cases almost inhibited their formation. When this communication was published we were in the course of determining the effect of gibberellic acid on the nodulation of *Trifolium repens* and ultimately reported² that, at 25 p.p.m., gibberellic acid in agar culture had no effect on nodule numbers in this plant; and further that up to 1000 p.p.m. had no effect on the growth of *Rhizobium trifolii*. It was thus obvious that not all legumes reacted in the same way regarding nodulation. Prof. Galston explained these apparently conflicting results³ by stating that 'this difference in nodulation response is probably related to the already known fact that different genotypes of plants presumably of varying endogenous gibberellin-levels react quite

differently to identical gibberellin applications'. Further experiments which we have recently carried out with small-seeded legumes and their corresponding *Rhizobia* furnish more evidence of this variability and of the fact that reduction in nodulation, where present, is not due to the effect of the gibberellic acid on the *Rhizobia*.

The legume seeds were surface-sterilized, germinated on sterile water-agar and transferred to boiling tubes containing nitrogen-free mineral salt agar⁴. Each tube contained one plant, and there were ten replicates per treatment. One ml. of 500 p.p.m. potassium gibberellate containing 6.0 million *Rhizobia* was added to each tube. Control tubes containing no gibberellate were also set up. The plants were allowed to grow for six weeks in a cool greenhouse and then harvested.

Table 1. EFFECT OF SODIUM GIBBERELLATE ON THE NODULATION OF SMALL-SEEDED LEGUMES

| Species | Number of nodules | | Dry weight of plant (mgm.) | |
|-------------------------|-------------------|---------------|-----------------------------|------------------------------|
| | No gibberellic | + gibberellic | No gibberellic (inoculated) | + gibberellic (uninoculated) |
| <i>Trifolium repens</i> | 7 | 8 | 12 | 7 |
| <i>T. hybridum</i> | 11 | 7 | 17 | 11 |
| <i>T. pratense</i> | 22 | 12 | 25 | 18 |
| <i>T. incarnatum</i> | 24 | 11 | 30 | 26 |
| <i>T. subterraneum</i> | 24 | 12 | 33 | 31 |
| <i>Medicago sativa</i> | 7 | 9 | 21 | 15 |

From Table 1 it can be seen that potassium gibberellate had no effect on the nodulation of *T. repens* and *M. sativa* whereas the nodule numbers were reduced by some 50 per cent in *T. hybridum*, *T. pratense*, *T. incarnatum* and *T. subterraneum*. In all cases the nodules formed in the presence of potassium gibberellate were effective, the dry weight of the plant being as heavy in the presence as in the absence of potassium gibberellate and heavier than the uninoculated control. In order to determine whether the reduction in nodulation, where noted, was due to the effect of potassium gibberellate on the *Rhizobia* used in the experiment or on the plant, the *Rhizobia* were grown in a liquid medium⁵ containing the chemical. Turbidimetric readings were taken daily for 14 days with an 'Eel' nephelometer, and showed little or no deviation from the control. Only the readings taken at the 14-day period are shown in Table 2.

TABLE 2

| Strain of <i>Rhizobia</i> Conc. of gibberellic (p.p.m.) | N/CLF (T. repens) | Y/SU/297 | | N/AH ₂ (M. sativa) |
|---|-----------------------|-----------------------------|-----------------------------------|-------------------------------|
| | | (T. hybridum) (T. pratense) | (T. incarnatum) (T. subterraneum) | |
| 0 | 149 × 10 ⁷ | 91 × 10 ⁷ | 154 × 10 ⁷ | 134 × 10 ⁷ |
| 500 | 145 × 10 ⁷ | 80 × 10 ⁷ | 156 × 10 ⁷ | 135 × 10 ⁷ |
| 1000 | 148 × 10 ⁷ | 88 × 10 ⁷ | 158 × 10 ⁷ | 133 × 10 ⁷ |

From these figures it is clear that the potassium gibberellate had no effect on the growth of the *Rhizobia*. The reduction in nodulation where noted in some of the species then was due to the effect of the chemical on the plant and Prof. Galston's explanation appears to be a very likely one.

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