

and there is no evidence that pea root growth is inhibited at concentrations below those affecting the spindle.

It is suggested that the inhibition of root growth may be the result both of an effect on the spindle and an effect on some metabolic activity connected with mitosis, similar to the 'poison' effect of various spindle-inhibiting substances reported by Levan and Östergren^{5,6}. With these compounds the poison effect was only brought about by concentrations higher than those required to affect the spindle; but the relation between the threshold concentrations for the two effects varied considerably between different compounds.

These findings indicate that the selective toxicity of phenylcarbamates is not necessarily related to differential effects on the spindle and suggest that the assessment of toxicity in terms of mitotic aberrations alone may give misleading results.

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¹ Templeman, W. G., and Sexton, W. A., *Nature*, **156**, 630 (1945).

² Ennis, W. B., *Amer. J. Bot.*, **35**, 18 (1948).

³ Ivens, G. W., and Blackman, G. E., *Symp. Soc. Exp. Biol.*, **3**, 266 (1949).

⁴ Doxey, D., *Ann. Bot.*, **13**, 329 (1949).

⁵ Levan, A., and Östergren, G., *Hereditas*, **29**, 381 (1943).

⁶ Östergren, G., *Hereditas*, **30**, 429 (1944).

Inoculation of Plants with Vascular Pathogens

Systemic infection of young plants. During the course of studies on the effect of mineral nutrition on the reaction of plants to infection with *Verticillium albo-atrum* Reinke and Berth, it became necessary to secure systemic infection of young plants without introducing the pathogen through the roots. The following method was therefore devised, using tomatoes as test plants.

Tomato seeds were sown at a spacing of 1½ in. × 1½ in. in boxes of sterilized soil. When the young plants were about 3 in. high, they were cut off at soil-level and stood with the basal ½-in. immersed in a spore suspension of *V. albo-atrum*. This spore suspension was standardized by means of a haemocytometer and contained about 50,000 spores per ml. The tomato cuttings were left in the spore suspension for 24 hr., then removed and washed for 1 hr. in running water to remove spores adhering to the stem surface. The cuttings were then rooted in small pots containing a peat-sand mixture or pure sharp sand. Tests of sample plants for the presence of *Verticillium* immediately after treatment showed that the fungus could be isolated from the vascular tissue throughout the stem.

Abundant roots were formed within ten days, and the rooted cuttings were then potted on for use in the experiments.

The first noticeable effect of the spore treatment was stunting of the treated plants which, after rooting, were usually half to two-thirds the size of the control plants grown from cuttings that had been stood in water only. Although initially stunted, the treated plants afterwards grew normally but were less forward than the control plants. Under suitable

manurial and temperature conditions for the expression of wilt, typical symptoms were first seen on the treated plants four to five weeks after inoculation.

Although particularly useful with tomatoes owing to their ready rooting, the method has proved applicable to several other plants that can grow from cuttings. I used it in 1938 to infect carnation cuttings with *Verticillium cimerescens* Woll. Plants grown from these cuttings showed similar symptoms to those grown from cuttings taken from naturally infected plants. Antirrhinum and aster plants have been similarly infected with *V. albo-atrum* and showed wilt symptoms within two to three weeks after inoculation.

No tests have been made with other vascular pathogens; but the method seems likely to be of use with any organism which invades the vascular tract. It should prove of value in investigations requiring rapid and uniform inoculation of plants with such organisms, as it enables large numbers of infected plants to be produced and obviates some of the sources of variation inherent in root-inoculation methods. Two such types of investigation are the screening of varieties for disease resistance and tests of chemicals for therapeutant effects. The value of the method for therapeutant testing is now being studied at East Malling.

Infection of single stems of mature plants. During experiments involving the inoculation of hop stems with *Verticillium albo-atrum*, difficulty was experienced in obtaining uniform infection by the usual method of insertion of fungal inoculum into a wound in the stem. A method was therefore devised making use of the pith cavity of the stem internodes as a site for a large dose of inoculum. The internodes of hop stems are hollow, but the pith cavity is sealed at the nodes. The inoculum used was a suspension of spores of *V. albo-atrum* (approximately 50,000 per ml.) made up in a 0.5 per cent solution of agar-agar in water. The spores were added to the agar when this was at a temperature of 40° C. and thus still liquid. The agar was then allowed to cool and formed a semi-solid gel. A hypodermic syringe was filled with the suspension and fitted with a fine needle.

A large needle was passed through the top of the hollow portion of the internode and withdrawn, leaving two holes leading to the pith cavity. The needle of the syringe was then carefully inserted into the pith cavity at the bottom of the internode and the agar spore suspension injected into the cavity. As the agar entered, the air escaped from the upper holes and the injection was complete when small globules of agar appeared at these holes. Owing to its semi-solid state, the agar remained in the cavity; but the holes were closed with crepe rubber bandage to exclude air. If extra large doses of inoculum were required, several internodes were inoculated on each stem in the manner described.

Many inoculations were made by this method and were invariably successful, typical wilt symptoms usually appearing above the selected internode after three to four weeks. The method could presumably be used on other hollow-stemmed plants and, since the inoculum is protected by the outer tissues of the stem, it seems likely to be more efficient than some other techniques.

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