PHENYLSERINE METABOLISM IN CUCUMBER SEEDLINGS

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SUMMARY

Phenylserine, benzoic acid and salicylic acid were isolated from cucumber seedlings after feeding DL-threo-β-phenylserine to the root. Traces of salicylic acid were also found in untreated plants.

1. INTRODUCTION

L-threo-β-phenylserine increases the resistance of cucumber plants against *Cladosporium cucumerinum*, probably via enhanced indoleacetic acid oxidase activity in the hypocotyls (VAN ANDEL, 1966a). Since phenylserine (PS) itself was inactive *in vitro* in both respects, an indirect effect is supposed, possibly leading to a decrease in IAA-oxidase inhibitors in cucumber hypocotyls and cotyledons (VAN ANDEL 1962, 1968, NIEMANN 1970a).Not much is known about the fate of PS in higher plants. Small amounts of benzoic acid were found in extracts of cucumber seedlings after application of PS (VAN ANDEL 1966b). Since the effect of benzoic acid on the fungus and its influence on IAA-oxidase after root application are low (VAN ANDEL 1966b, NIEMANN 1970a) benzoic acid does not seem to explain the PS activity. Bacteria and animal tissue have been shown to break down PS to benzaldehyde and glycine (JANEČEK *et al.* 1960, BRUNS & FIEDLER 1958). Again, both benzaldehyde and glycine have almost no PS activity (VAN ANDEL 1962, 1966b).

2. MATERIAL AND METHOD

Ten-day old cucumber seedlings, *Cucumis sativus* L. cv. "lange gele tros" were placed with their roots in 15×10^{-3} M PS or in water. After three days hypocotyls or whole plants were ground in ethanol. The ethanol solution was concentrated by evaporation and freeze-drying and washed with ligroin and chloroform. The residue was dissolved in butanol, extracted with 0.1 N ammonia, the ammonia layer was acidified and again extracted with butanol. The organic acid phase was separated by successive banding with butanol-1/27% acetic acid (1:1), butanol-1 sat. with 1.5 N ammonia and with acetone/ammonia/water (90:3:7).

3. RESULTS AND DISCUSSION

When 2-^{14}C phenylserine was fed to the roots for one day, part of it could be recovered as such from the hypocotyls. Furthermore, a number of spots were obtained in our chromatograms with R_f values similar to those obtained after feeding glycine-1-^{14}C. The latter indicates a break-down with glycine as one of the products. Since we were especially interested in the aromatic part of the molecule, these experiments were not pursued. After feeding 15×10^{-3} M phenylserine to the roots for three days three compounds were isolated (in solution) by repeated banding of whole-plant extracts on chromatography paper. The compounds were identified as phenylserine, benzoic acid and salicylic acid by comparison of R_f values, UV spectra and colour reactions with those of the authentic samples. Phenylserine and salicylic acid were also isolated from the hypocotyls; the benzoic acid concentration, apparently, was too low for detection. Salicylic acid was also found in untreated plants. The concentration, however, was about 0.06 of that in PS-treated plants. Salicylic acid, when fed to the roots, enhanced the IAA-oxidase activity of cucumber hypocotyl extracts (NIEMANN 1970b). However, the margin between active and toxic doses is rather small, and root injury also causes an increase in IAA-oxidase activity (NIEMANN 1970b). On the other hand, a small effective dose of benzoic and salicylic acid in the right place, both fungitoxic and both with a still unexplained effect on IAA-oxidase activity, could be responsible for the PS effect.

REFERENCES

ANDEL, O. M. VAN (1962): Growth regulating effects of amino acids and dithiocarbamic acid derivatives and their possible relation with chemotherapeutic activity. *Phytopath. Z.* **45**: 66–80.

— (1966a): Mode of action of L-threo-β-phenylserine as a chemotherapeutant of cucumber scab. *Nature (Lond.)* **211**: 326–327.

— (1966b): Amino acids and plant diseases. *Ann. Rev. Phytopath.* **4**: 349–368.

— (1968): Shifts in disease resistance induced by growth regulators. *Neth. J. Pl. Path.* **74**: (Suppl.) 113–120.

BRUNS, F. H. & L. FIEDLER (1958): Enzymatische Spaltung und Synthese von L-threo-β-Phenylserin und L-erythro-β-Phenylserin. *Biochem. Z.* **330**: 324–341.

JANEČEK, J., J. CHALOUPKA, K. VEREŠ & M. HAVRANEK (1960): Regulation of protein synthesis in Escherichia coli by amino acid analogues. *Folia Microbiol.* **5**: 207–216.

NIEMANN, G. J. (1970a): The effect of phenylserine and related compounds on the indoleacetic acid oxidase activity of cucumber hypocotyl and root extracts. *Acta Bot. Neerl.* **19**: 415–418.

— (1970b): A possible mechanism for the increased resistance against fungal attack as a consequence of root injury: Acta Bot. Neerl. **19**: 681–683.