

With regard to the resulting effect on ammonium-uptake, it was found that addition of this weak acid tended to counteract the effect of excision of the shoot, although it did not do so completely. Again a decrease in uptake was found, but to a lesser extent (only about 35 per cent). This work is being continued with other acids.

I thank the Director of the Laboratorium voor Bloembollenonderzoek at Lisse for the use the 'Uras' and the 'Magnos' apparatus.

A. L. KOSTER

Botanical Laboratory,  
Department of Experimental Botany,  
University of Leyden.

<sup>1</sup> Bach, M. K., Magee, E. W., and Burriss, R. H., *Plant Physiol.*, **33**, 118 (1958).

<sup>2</sup> Bond, G., *J. Exp. Biol.*, **7**, 387 (1956).

<sup>3</sup> Wieringa, K. T., and Bakhuis, J. A., *Plant and Soil*, **8**, 254 (1957).

<sup>4</sup> Bond, G., *Ann. Bot.*, N.S., **14**, 245 (1950).

<sup>5</sup> Moore, S., and Stein, W. H., *J. Biol. Chem.*, **178**, 367 (1948).

<sup>6</sup> Hoagland, D. R., and Arnon, D. I., *Circular 347* (Univ. Calif., 1950).

<sup>7</sup> Becking, J. H., *Acta Botan. Neerl.*, **5**, 1 (1956).

<sup>8</sup> Allport, N. L., *Colorimetric Analysis* (Chapman and Hall, London, 1947).

<sup>9</sup> Hylmø, B., *Physiol. Plantarum*, **6**, 333 (1953).

<sup>10</sup> Jensen, G., *Physiol. Plantarum*, **15**, 363 (1962).

<sup>11</sup> Lycklama, J. C. (to be published).

### Kinetin as an Antagonist of the Toxic Effect of *Pseudomonas tabaci*

The wildfire disease of tobacco, caused by *Pseudomonas tabaci* (Wolf and Foster) Stevens, is characterized by the development of chlorotic halos around the infection sites. This symptom is induced by a bacterial toxin. The toxic compound proved to be a structural analogue of methionine<sup>1</sup>. Strangely enough, its effect could be overcome by methionine only in model experiments with *Chlorella vulgaris*<sup>2</sup>. The mode of action of the wildfire toxin in tobacco leaves, therefore, remained obscure.

To throw some light on this problem, investigations of the metabolic alterations in tobacco leaves induced by the toxic effect of *Ps. tabaci* were undertaken<sup>3,4</sup>. In this communication results of investigations on the protein metabolism of tobacco tissues treated with toxin-containing culture filtrates are presented. It was found that in leaves of intact tobacco plants injected with culture filtrates of *Ps. tabaci* the soluble protein content decreases, whereas the free amino-acid and ammonia content increases (Table 1). Proteins were determined according to Lowry *et al.*<sup>5</sup>, total free amino-acids were assayed by a copper method<sup>6</sup>, and ammonia content was estimated by micro-diffusion in Conway units.

Table 1. PROTEIN, FREE AMINO-ACID AND AMMONIA CONTENT OF TOBACCO HALF-LEAVES TREATED WITH TOXIN-CONTAINING CULTURE FILTRATE OF *Pseudomonas tabaci* AND WITH NUTRIENT MEDIUM RESPECTIVELY

Exp. No.	Protein mg/g fresh wt.		Amino-N µg/g fresh wt.		Ammonia µg/g fresh wt.	
	C	T	C	T	C	T
1	14.0	7.3	51	87	24	570
2	9.0	6.6	26	120	10	460
3	9.7	6.6	40	105	17	510

C. Tissues treated with Czapek-Dox nutrient medium; T, tissues treated with toxin-containing culture filtrate.

The results indicate that the balance of protein synthesis and breakdown is shifted in favour of increased decomposition of protein. The development of the chlorotic symptom and the changes of protein metabolism which occur under the effect of the wildfire toxin are very similar to those occurring in detached (or senescent) leaves. As in the latter both chlorosis and breakdown of

Table 2. EFFECT OF KINETIN ON THE PROTEIN CONTENT OF TOBACCO LEAVES TREATED WITH TOXIN-CONTAINING CULTURE FILTRATE OF *Pseudomonas tabaci*

Exp. No.	Protein mg/g fresh wt.	
	Half-leaves sprayed with Water	10 <sup>-5</sup> M kinetin
1	5.1	9.9
2	4.3	9.9
3	6.5	12.0

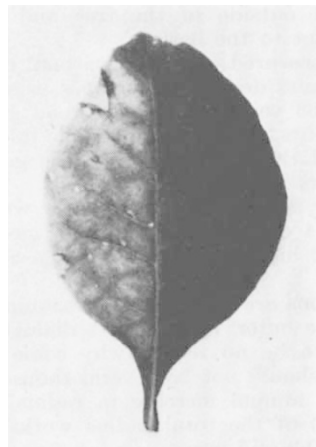


Fig. 1. Counter-action by kinetin of the toxic effect induced by culture filtrate of *Pseudomonas tabaci* injected into tobacco leaf. Left half sprayed with water, right half sprayed with 10<sup>-5</sup> M kinetin

protein can be counteracted by kinetin<sup>7,8</sup> the effect of this compound on the metabolism of tobacco leaves treated with toxin-containing culture filtrates was investigated. Leaves were injected with culture filtrates of *Ps. tabaci* and half-leaves were sprayed with 10<sup>-5</sup> M kinetin. Control halves were sprayed with water. The treatments were repeated three times in 24-h intervals. It may be seen from Fig. 1 that in the kinetin-treated half-leaf the toxin-induced chlorosis fails to develop. Further investigations have shown that the deleterious effect of the wildfire toxin on the protein-level is also fully counteracted by kinetin (Table 2).

The results described here clearly indicate that the protein metabolism of the host is adversely affected by the toxin of *Ps. tabaci* but still do not give a final answer to the problem of the primary point of attack.

The results obtained are compatible with the idea that the toxin-induced damage to protein metabolism in tobacco may be due to competitive antagonism. However, serious consideration must be given to alternative explanations as well. Recent results indicate that both the decrease of protein-level in detached leaves and its reversal by kinetin are associated with a concomitant change in ribonucleic acid (RNA) concentration. The RNA/protein ratio remains always constant<sup>9</sup>. These results suggest that the alterations of protein metabolism both in detached and in kinetin-treated leaves may be secondary in nature and may be evoked by primary changes in RNA metabolism. The similarity of protein metabolism in detached and in intact toxin-treated leaves is striking. Their response to kinetin is also identical. Therefore, investigations on the effect of the wildfire toxin on RNA metabolism seem to be necessary. Investigations in this direction are prompted particularly by the recent observations that the ribonuclease activity in extracts from tobacco tissues treated with toxin-containing culture filtrates of *Ps. tabaci* is markedly increased.

L. LOVREKOVICH  
G. L. FARKAS

Research Institute for Plant Protection,  
Budapest.

<sup>1</sup> Woolley, D. W., Schaffner, G., and Braun, A. C., *J. Biol. Chem.*, **215**, 485 (1955).

<sup>2</sup> Braun, A. C., *Phytopath.*, **45**, 659 (1955).

<sup>3</sup> Farkas, G. L., Lovrekovich, L., and Klement, Z., *Naturwiss.*, **50**, 22 (1963).

<sup>4</sup> Lovrekovich, L., Klement, Z., and Farkas, G. L., *Nature*, **197**, 917 (1963).

<sup>5</sup> Lowry, O. H., Rosebrough, N. J., Farr, A. L., and Randall, R. J., *J. Biol. Chem.*, **193**, 265 (1951).

<sup>6</sup> Belozerskij, A. N., and Proskurjakov, N. I., *Praktikum der Biochemie der Pflanzen* (VEB Deut. Verlag d. Wiss., Berlin, 1956).

<sup>7</sup> Richmond, A. E., and Lang, A., *Science*, **125**, 650 (1957).

<sup>8</sup> Mothes, K., *Naturwiss.*, **47**, 337 (1960).

<sup>9</sup> Osborne, D. J., *Plant Physiol.*, **37**, 595 (1962).