

The HbBC of the same serum increases after 10 min oxygenation.

The method of oxygenation influences the results. The treatment with a smaller quantity of oxygen leads to an increase of lesser degree. It seems to be necessary to standardize the conditions of oxygenation. Only in this manner would it be possible to settle definitely whether the rate of HbBC increased by oxygen treatment is correlated with the phenotype or not. It may be assumed that a relation exists between the two features of haptoglobins described: the sensibility to heat and the dependency from oxygen. At present there is no explanation available for these phenomena.

Résumé. Sous l'effet de l'oxygène l'«hemoglobine binding capacity» des haptoglobines du sérum natif augmente.

Ö. HEVÉR

State Institute Fodor József, Postafiók 1,
H-1528 Budapest 123 (Hungary), 20 June 1974.

Effect of Methylendioxyphenyl Synergists on Metabolism of Carbaryl by *Aspergillus terreus*¹

Methylenedioxyphenyl compounds were originally developed for use with the pyrethrin insecticides as synergists. However, it was found that they also possess the ability to synergize carbamate insecticides. Especially, these synergists are capable of enhancing the toxicity of carbaryl (1-naphthyl N-methylcarbamate) toward houseflies which efficiently detoxicate the insecticide as an inhibitor of cholinesterase.

When methylenedioxyphenyl synergists were added to aldrin-treated soils, the conversion of aldrin to dieldrin was inhibited². In experiments with culture media con-

taining microorganisms, it appeared that the inhibition of the conversion of aldrin to dieldrin occurred through an effect of sesamex on microorganisms, primarily bacteria. Lichtenstein et al. concluded that the chemical may either inhibit the enzyme system responsible for the

¹ Authorized for publication on 10 July 1974 as paper number 4732 in the journal series of the Pennsylvania Agricultural Experiment Station.

² E. P. LICHTENSTEIN, K. R. SCHULZ and G. T. COWLEY, J. econ. Entom. 56, 485 (1963).

Concentration of sesamex (ppm)

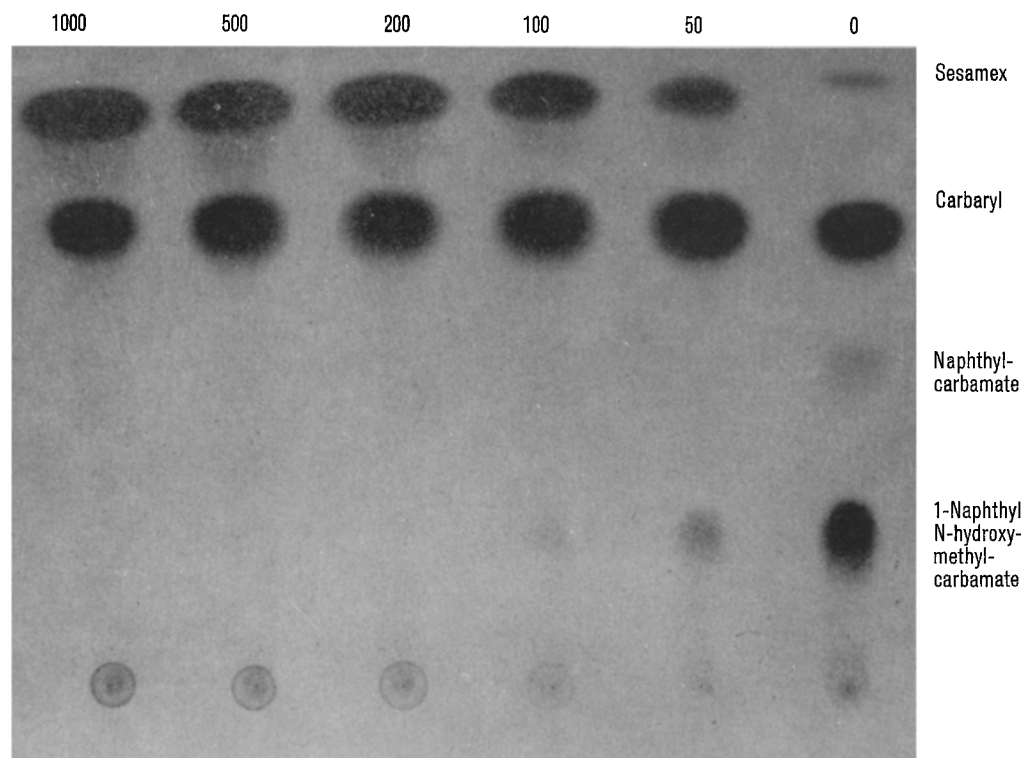


Fig. 1. Effect of different concentrations of sesamex on metabolic transformation of carbaryl by *Aspergillus terreus* (Thin-layer chromatography of ether extract from growth medium after 10 days of growth).

CARBARYL				CARBARYL + SESAMEX				CARBARYL + SESAMOL				
D A Y S				D A Y S				D A Y S				
0	4	8	12	0	4	8	12	0	4	8	12	
				⊗	⊗	⊗	○	⊗	⊗	○		Synergist
⊗	⊗	⊗	○	⊗	⊗	⊗	⊗	⊗	⊗	⊗	⊗	Carbaryl
		○	⊗									1-Naphthyl- Carbamate
	○	⊗	⊗			○	⊗				○	1-Naphthyl N-Hydroxymethyl Carbamate
○	○	○	○	○	○	○	○	○	○	○	○	

Fig. 2. Influence of *Aspergillus terreus* on the transformation of carbaryl without and with sesamex and sesamol after various time intervals. (Thin-layer chromatography of ether extract from growth medium.)

biological oxidation of aldrin, or reduce the number of microorganisms containing this enzyme.

Methylcarbamate compounds are particularly interesting for synergistic studies because many of them respond strongly to synergists³. Since it was shown in our laboratory that various soil microorganisms are capable of decomposing the insecticide carbaryl⁴⁻⁶, it was of interest to elucidate whether methylenedioxyphenyl synergists interfere in the metabolic activity of these microorganisms.

For this purpose a strain of the fungus *Aspergillus terreus* whose carbaryl-decomposing characteristics were previously described⁷ served as the test organism. Sesamex and its anti-oxidant breakdown product sesamol were used as methylenedioxyphenyl synergists. The fungus was grown in a nutrient medium⁷ containing carbaryl or a synergist alone or a combination of the two compounds. All chemicals were sterilized by membrane filtration (0.22 µm pore size, Millipore Corp., Bedford, Mass.). The media were inoculated with spores from a slant culture and incubated on a rotary shaker at 28°C. The culture filtrates were extracted with ethyl ether for identification of the metabolites. An aliquot of the concentrated ether extract was analyzed by thin-layer chromatography employing ether:hexane (4:1, v/v) as developing system. Subsequently, the plates were sprayed with a 15% potassium hydroxide solution, air-dried, sprayed with 1 M acetic acid in methanol, and 0.1% methanolic *p*-nitrobenzenediazonium fluoborate for visualization of naphthol-related spots.

Carbaryl was metabolized by the fungus *A. terreus* to 1-naphthyl carbamate and 1-naphthyl N-hydroxymethylcarbamate after 8 days of incubation, but the addition of the synergist sesamex at a concentration of 50 and 100 ppm caused partial inhibition and a concentration of 200–500 ppm completely inhibited the conversion of carbaryl to its metabolites (Figure 1). When *A. terreus* was cultured in a growth medium containing sesamex and

sesamol, the synergists disappeared after prolonged incubation. This was apparently caused by the metabolic activity of the fungus. In an experiment in which sesamex and sesamol were added at a concentration of 100 ppm together with carbaryl, sesamex was considerably reduced whereas sesamol was not detectable after 12 days incubation (Figure 2). However, it appears that sesamol was more effective as an inhibitor than sesamex, since only after 12 days the occurrence of 1-naphthyl N-hydroxymethylcarbamate was detectable, while with sesamol its formation was observed after 8 days.

The described experiments lead to the conclusion that methylenedioxyphenyl synergists exert an inhibitory effect on the transformation of carbaryl by *A. terreus*, but since they can also be metabolized by the fungus, their effectiveness as synergists is limited by their fallibility to microbial attack.

Zusammenfassung. Die Methylenedioxyphenyl-Synergisten Sesamex und Sesamol hemmen die Umwandlung des Insektizids Carbaryl (1-Naphthyl-N-Methylcarbamat) zu N-Hydroxymethylcarbamat und 1-Naphthylcarbamat bei *Aspergillus terreus*. Die Wirkung der beiden Synergisten ist jedoch von begrenzter Dauer, da sie durch den Pilz metabolisiert werden.

J.-M. BOLLAG and KANG-CHIEN LIU

Laboratory of Soil Microbiology, Department of Agronomy, The Pennsylvania State University, 119 Tyson Building, University Park (Pennsylvania 16802, USA), 23 July 1974.

³ J. E. CASIDA, J. Agric. Food Chem. 18, 753 (1970).

⁴ J.-M. BOLLAG and S.-Y. LIU, Soil Biol. Biochem. 3, 337 (1971).

⁵ J.-M. BOLLAG and S.-Y. LIU, Nature, Lond. 236, 177 (1972).

⁶ S.-Y. LIU and J.-M. BOLLAG, J. Agric. Food Chem. 19, 487 (1971).

⁷ S.-Y. LIU and J.-M. BOLLAG, Pestic. Biochem. Physiol. 1, 366 (1971).