It is unlikely that the channels of a given population can be formed by different venom proteins. It may therefore be assumed that all four of the fractions that we investigated contained the same substance with a minimum molecular weight of 5000 dalton and below. This substance may indeed act presynaptically but it is activated or acquires a specific direction in the presence of another component with a higher molecular weight. In actual fact, the low-molecular-weight fraction IV increases the frequency of MEPPs when it is added to a preparation of frog and cockroach synapses, but this effect is observed only after a latent period lasting from 30 to 60 min, which can be explained by its less effective interaction with the presynaptic membrane in the absence of a high-molecular-weight support (promoter). In confirmation of this alternative we must draw attention to the results of the repeat gel filtration of fraction I from which likewise a low-molecular-weight fraction analogous to fraction IV in the molecular dimension of the components present in it and its action on synapses at bilayer membranes, in which it formed the same channels, was separated.

We obtained completely identical results in an investigation of the venom of the spider Lactrodectus mactans.

Thus, in spite of the fact that the two spiders belong to different genera of the family Theridiidae, their venoms have a similar active principle – a low-molecular-weight channel-forming component. On the basis of the results that we have obtained, it is possible to conclude that the structures of the presynaptic neurotoxins of the venoms of the spiders Lithyphantes paykullianas and Latrodectus mactans consist of a promotor of high molecular weight ensuring the selective action of the presynaptic membrane of synapses either of mammals or insects, and a low-molecular-weight peptide (5000 ± 500 dalton) which is a channel-forming agent. This point of view is in harmony with a now forgotten hypothesis [5] relating to a low-molecular-weight toxin of the spider Latrodectus mactans associated with another high-molecular-weight protein.

LITERATURE CITED

1. H. E. Longenecker, W. P. Hurlbut, A. Mauro, and A. W. Clark, Nature (London) 225, 701 (1970).
2. S. Bettini and M. Maroli, in: Arthropod Venoms (Vol. 48 of Handbook of Experimental Pharmacology), S. Bettini, ed., Springer, Heidelberg (1978), Chapter 8, p. 149.
3. A. Finkelstein, L. L. Rubin, and M. C. Tzeng, Science, 193, 1009 (1976).
4. P. V. Krasil'nikov, V. I. Ternovskii, and B. A. Tashmukhamedov, Biokhimiya, 27, 72 (1982).
5. J. D. McCrone, Am. Zool., 9, 153 (1969).

ACTION OF TRICYCLAZOLE ON THE BIOSYNTHESIS OF MELANIN IN SOME FUNGI OF THE GENUS Verticillium

L. N. Ten, N. N. Stepanichenko, S. Z. Mukhamedzhanov, and A. V. Khotyanovich

UDC 547.651:576.809.8+632.428

It is known that the systemic fungicide tricyclazole (5-methyl-1,2,4-triazolo[3,4-c]benzothiazole) blocks different stages of the formation of melanin in Verticillium dahliae, depending on the concentration, thereby causing the accumulation in a culture of the fungus of biosynthetic precursors of this polymer and their transformation products [1]. This property of tricyclazole has been used to prove the identity of melaninogenesis in the fungi Thielaviopsis basicola, Pyricularia oryzae, and V. dahliae [1-3].

In the present communication we give the results of the use of this fungicide for a comparative study of some stages of the biosynthesis of melanin in the fungi V. tricorpus, V. nigrescens, and V. dahliae.

We used natural isolates of V. tricorpus P-24 and P-26, V. nigrescens XI 681 and Khl-763, and V. dahliae KhL-1,3 and Khl-17 obtained from the laboratory of the genetics of cottonplant immunity of the Division of General Genetics of the Cotton Plant of the Academy of Sciences of the Tadzh SSR.

V. I. Lenin Tashkent State University. All-Union Scientific-Research Institute of Agricultural Microbiology, Leningrad.

Translated from Khimiya Prirodnykh Soedinenii, No. 3, pp. 401-402, May-June, 1983. Original article submitted November 30, 1982.

0009-3130/83/1903-0384$07.50

The cultivation of the fungi and the isolation and identification of the pentaketide metabolites was carried out by methods described in the literature [1, 4]. Technical tricyclazole (EL-291) was added to the medium in the form of an ethanolic solution, and the corresponding amount of ethanol was added to the control.

It was established that the treatment of all isolates of V. tricorpus V. nigrescens, and V. dahliae with tricyclazole in concentrations of 0.1-1.0 μg/ml led to the suppression of the synthesis of melanin in the dormant structures of these fungi. From all the cultures we isolated and identified as the main pentaketide metabolite 2.5-dihydroxy-1,4-naphthoquinone (2-hydroxyjuglone) (I). The accumulation of the latter shows the blockage by tricyclazole of the conversion of 1,3,8-trihydroxynaphthalene (II) into vermelone in the chain of biochemical reactions involved in the formation of melanin, and the autooxidation of (II) to (I), as shown by Tokousbaliedes and Sisler [1].

An increase in the concentration of tricyclazole to 10 μg/ml was accompanied by the appearance in the cultures of all the isolates of V. tricorpus, V. nigrescens, and V. dahliae of 2,5,7-trihydroxy-1,4-naphthoquinone (flaviolin) (III) and a decrease in the amount of (I). This unambiguously shows the presence on the biosynthetic route to the formation of melanin in V. tricorpus and V. nigrescens of a stage of the reduction of 1,3,6,8-tetrahydroxynaphthalene (IV) to acytalone with secondary sensitivity to tricyclazole, since (III) is formed as the result of the autooxidation of (IV), as has been shown for V. dahliae [1].

The results on the inhibition by tricyclazole of the biosynthesis of melanin in isolates of V. dahliae Khl-1,3 and Khl-17 agree with those given by Tokousbaliedes and Sisler [1] for the strain ATCC 22924 of this fungus. We are the first to have described a similar effect of the fungicide used on V. tricorpus and V. nigrescens.

The blockage of the biosynthesis of melanin in V. tricorpus and V. nigrescens by the same concentrations of tricyclazole as for V. dahliae and the appearance as the result of this in cultures of these fungi of the same pentaketide metabolites indicates that the initial stages of the biosynthesis of melanin up to the formation of (II) in V. tricorpus and V. nigrescens are identical with those in V. dahliae [4-6].

LITERATURE CITED

1. M. C. Tokousbalides and N. D. Sisler, Pestic. Biochem. Physiol., 11, 64 (1979).
2. C. P. Woloshuk et al., Pestic. Biochem. Physiol. 14, 256 (1980).
3. M. H. Wheeler and R. D. Stipanovic, Exp. Mycol., 3, 340 (1979).
4. L. N. Ten et al., Khim. Prir. Soedin., 393 (1980).
5. A. A. Bell et al., Tetrahedron, 32, 1363 (1976).
6. R. D. Stipanovic and A. A. Bell, Mycologia, 69, 164 (1977).