

confirmed by our results revealing anticoagulant activity in phospholipase A₂ III-3 with pI 6.73 and not detecting it in phospholipase A₂ III-2 with pI 7.57.

It may be assumed that other features of their structure are important for the manifestation of anticoagulant properties in phospholipases A₂.

LITERATURE CITED

1. K. Stocker, in: Natural Toxins, Proceedings of the 6th Symposium on Animal, Plant, and Microbial Toxins, Uppsala, 1979 (1980), p. 111.
2. D. N. Sakhinov, V. M. Sorokin, and L. Ya. Yukel'son, The Chemistry and Biochemistry of Snake Venoms [in Russian], Tashkent (1972).
3. M.-C. Boffa, J. Dachary, H. M. Verheij, C. Rothen, J. Dufoureaud, R. Verger, and G. H. de Haas, *Toxicon*, 20 (1982), 305.
4. M.-C. Boffa, C. Rothen, H. M. Verheij, R. Verger, and G. H. de Haas, in: Neutral Toxins, Proceedings of the 6th Symposium on Animal, Plant, and Microbial Toxins, Uppsala, 1979 (1980), p. 131.
5. G. E. Lyubimtseva and L. Ya. Yukel'son, Abstracts of Lectures at the IIIrd Conference of Biochemists of the Republics of Central Asia and Kazakhstan [in Russian], Dushanbe (1981).
6. L. Ya. Yukel'son and G. E. Lyubimtseva, *Khim. Prir. Soedin.*, 389 (1980).

A LOW-MOLECULAR-WEIGHT CHANNEL-FORMING COMPONENT OF THE NEUROTOXINS OF THE VENOMS OF SPIDERS OF THE FAMILY THERIDIIDAE

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The presynaptic effect of the venoms of spiders of the genus *Latrodectus* (family Theridiidae) is well known [1]. It is considered [2] that it is due to high-molecular-weight toxins which, on interacting with a membranous nerve termination cause the massive liberation of a mediator. It has been shown [3, 4] that the whole venom of the spider *Latrodectus mactans* and a purified toxin isolated from it with a molecular weight of 130,000 form conductivity channels for K⁺, Na⁺, and Ca²⁺ ions in a model membrane, and this has been made the basis of their presynaptic action.

We have investigated the venom of the spider *Lithyphantes paykulliana* from another genus of the family Theridiidae. In preliminary experiments it was shown that like the venom of spiders of the genus *Latrodectus*, this venom acts toxically on mammals and insects, with LD₅₀ for white mice and cockroaches of 5.5 and 5.2 mg/kg, respectively. Using nerve-muscle preparations of the frog and locust it has been established that the venom of the spider *Lithyphantes paykulliana* also causes a massive liberation of mediator, and in a model membrane it forms a homogeneous population of conductivity channels for cations with an amplitude of 20-350 pS, depending on the concentration and type of cation in the medium.

In order to ascertain the components responsible for the presynaptic and channel-forming effect of the venom, it was separated on Sephadex G-100. On using 0.01 M phosphate buffer with pH 8.0 containing 0.01 M sodium chloride, it was possible to separate the whole spider venom into four fractions containing proteins of the following molecular sizes: 100,000 dalton and above - fraction I; 60,000-70,000 dalton - fraction (II); 30,000-40,000 dalton - fraction (III); and 5000 ± 500 dalton - fraction (IV).

Test on preparations of the synapses from mammals and insects showed that fraction (I) increased the frequency of MEPPs and acted presynaptically only on vertebrates, while fraction (III) acted similarly only on the synapses of insects. All four fractions, including the one that contained components with a molecular weight of about 5000 dalton and below increased the integral conductivity of bilayer membranes and formed channels characterized by the same ionic selectivity and the same amplitude.

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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 Latrodectus 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 paykullianus 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*

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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.

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