CHANGES IN THE NUCLEIC ACID CONTENT IN MOUSE TISSUES DURING THE ACTION OF THE VENOM OF Vipera lebetina L. AND OF ANTIVIPER SERUM

A. S. Meshalov

UDC 615.94+ 615.373.39]:598.12]-092:612.015.348

The author's previous investigations [7] showed that, in response to the injection of a minimal lethal dose (MLD) of the venom of Vipera lebetina L., the nucleic acid (NA) content in the blood and spleen of albino mice falls sharply, and the animals die at the 8th hour of the experiment.

Investigations of the enzymic activity of the venoms of several snakes have shown that the venom of V. lebetina possesses phosphodiesterase and 5-nucleotidase (phosphomonoesterase) activity [3]. The desoxyribonuclease activity of snake venom has also been established [2]. Hence, snake venom contains enzymes active against NA, i.e., against the structures responsible for many different forms of protective reactions.

The best protection against snake bite is given by the antivenin known as "Antigyurza." The mechanism of action of this serum has not been analyzed in the literature [6]. Only in the reports of some investigations [1] is it mentioned that this serum reduces the hemolytic, coagulatory, and neurotoxic effects of the venom.

The author has attempted to discover whether the antiviper serum suppresses the nuclease activity of the venom. For this purpose, the changes in the NA content were studied in the organs and tissues of animals exposed to the action of the venom of V. lebetina L. alone, of the venom and the antiviper serum together, and of the serum alone.

EXPERIMENTAL METHOD

Experiments were conducted on albino mice weighing 18-20 g. The dried viper venom was dissolved in neutral physiological saline and injected subcutaneously into the animals in a dose of 1 MLD. At the same time, the animals received antiviper serum intraperitoneally in a dose neutralizing 1 MLD of venom (0.1 ml of serum in 0.4 ml physiological saline). In another case, the antiviper serum alone was injected. The results obtained were compared with the changes in the NA content observed under the influence of 1 MLD of venom. The mice were sacrificed at different times: on the 2nd and 8th hour and the 1st, 2nd, 3rd, 5th, and 7th day of the experiment. The NA content was determined in a weighed sample of fresh liver and spleen tissue, in two parallel samples, by A. S. Spirin's spectrophotometric method [5]. The same method was used to determine the NA content in the blood, but in P. V. Simakov's modification [4].

Experiments in which the venom and the antiviper serum were injected at the same time were carried out on 79 mice. Five of these animals were sacrificed at each of the following times: 2, 4, 5, and 6 h and 1, 2, and 5 days after the beginning of the experiment; 7 mice at each of the following times: 3, 7, and 8 h and 3 days; and 4 mice on the 7th day. Twenty control animals were sacrificed. Altogether, 136 samples of blood, 156 of spleen, and 156 of liver were tested.

Antivenin alone was injected into 74 mice. Five of these animals were sacrificed at each of the following times: 2, 3, 5, 7, and 8 h and 1, 3, 5, and 7 days, 6 mice at the 6th hour, 7 at the 4th hour, and 4 on the 2nd day after the beginning of the experiment. Twenty control animals were sacrificed. Altogether, 136 samples of blood, 148 of spleen, and 143 of liver were tested.

Department of General Biology, I. M. Sechenov First Moscow Medical Institute. (Presented by Active Member of the Academy of Medical Sciences of the USSR V. M. Zhdanov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 62, No. 8, pp. 36-39, August, 1966. Original article submitted December 17, 1964.

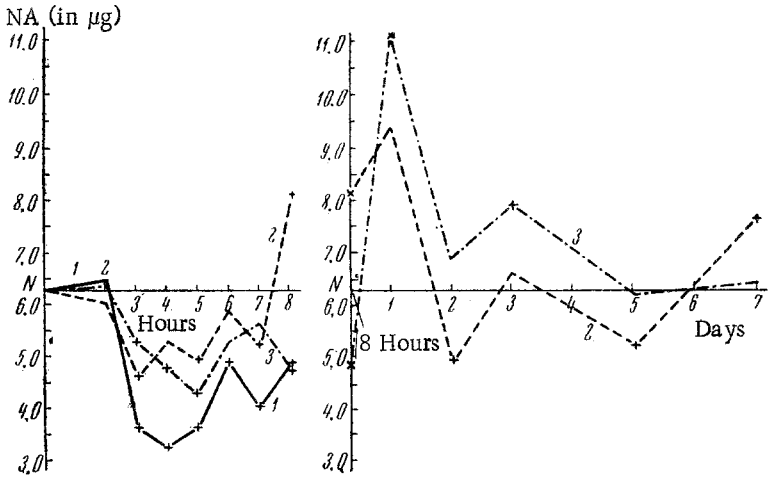

Fig. 1. Changes in the NA content in the blood of albino mice as a result of the action of 1 MLD of V. lebetina L. venom alone, of venom and antiviper serum together, and of serum alone. Here and in Figs. 2 and 3: N) normal content of NA; 1) action of venom alone; 2) action of viper venom and antiviper serum; 3) action of antiviper serum; x) statistically significant deviation.

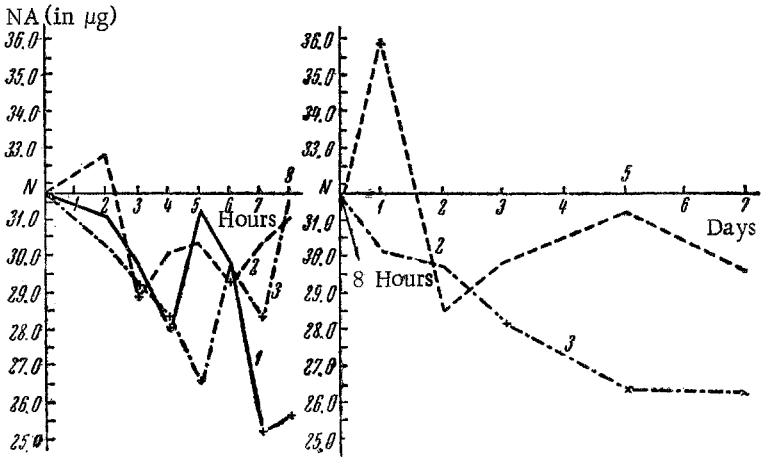

Fig. 2. Changes in the NA content in the spleen of albino mice as a result of the action of 1 MLD of V. lebetina L. venom alone, of venom and antiviper serum together, and of serum alone.

EXPERIMENTAL RESULTS

During the first hours of the experiment, a statistically significant decrease in the NA content in the blood was observed under the influence of venom, venom plus serum, and serum alone (Fig. 1). After injection of the venom alone, the NA level subsequently remained low, and the animal died at the 8th hour of the experiment. In the case of the combined administration of venom and serum, the NA content rose by a statistically significant degree to a maximum at the 8th hour of the experiment. The NA content then fell, but again increased on the 7th day. All these changes were statistically significant.

After injection of the serum alone, two statistically significant increases in the NA concentration in the blood took place on the 1st and 3rd days, after which the NA content returned to normal by the end of the experiment. In the spleen, the picture was similar, but with the difference that when serum alone was given the NA content fell after the 3rd day of the experiment (Fig. 2).

In the liver, no significant changes took place in the NA content after injection of venom alone and of venom with serum, but there was a distinct tendency for it to increase after the first 24 h. When serum alone was given,

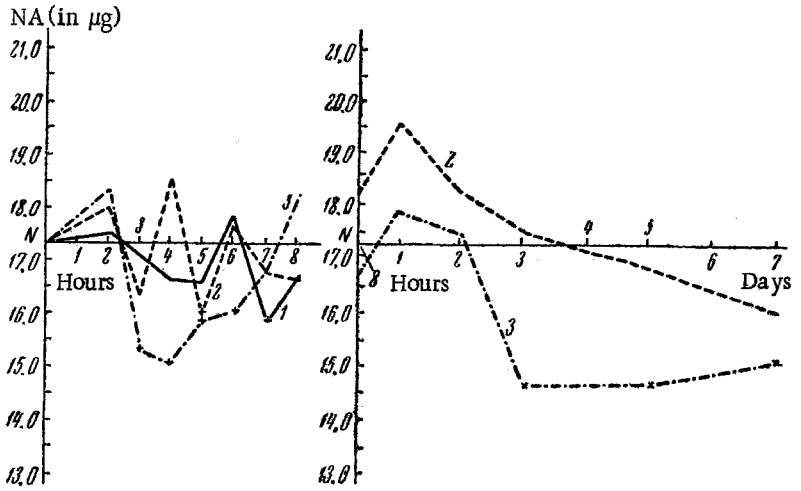

Fig. 3. Changes in the NA content in the liver of albino mice as a result of the action of 1 MLD of V. lebetina L. venom alone, of venom and antiviper serum together, and of serum alone.

the picture observed was almost completely identical with that of the spleen following injection of serum alone (Fig. 3).

The results showed that three periods of the experiment could be distinguished: 1st, from the beginning to the 5th hour; 2nd, from the 5th hour to the end of the first day; and 3rd, from the 1st to the 7th day of the experiment.

In the first period, regardless of the material injected, the NA content in the blood and organs fell. This was evidently a stressor reaction. At the beginning of the 2nd period, when venom alone had been injected, the mice died (a low NA level, the specific effect of the action of venom). After the simultaneous injection of venom and antivenin, and injection of the serum alone, the protective reactions of the body began to act in the second period, and this was shown by an increase in the NA content. In relation to the 3rd period, the experiment in which antivenin alone was injected may be considered. The fall in the NA content in the liver and spleen in these circumstances evidently reflected the inhibition of the process of immunogenesis as a result of the introduction of a large amount of foreign protein into the organism in the form of "Antigyurza" serum [8,9]. Free nucleotides entered the blood stream, accounting for the high NA content of the blood. Following the simultaneous administration of the venom and antivenin, similar phenomena occurred in the 3rd period. This also accounts for the fall in the NA content, although this was to a level not statistically significantly different from normal. The NA content in the blood also fell, and then increased later, toward the 7th day of the experiment. The causes of these changes require further study.

It may be concluded from the results of these investigations that the toxic action of snake venom is due not only to hemolysis and cytolysis, but also to another property of the venom, its nuclease activity. The nucleases, by destroying NA, prevent the development of the protective reactions of the organism and, in particular, the synthesis of antienzyme antibodies. The decrease in the nuclease activity of the venom under the influence of the "Antigyurza" serum is responsible for the development of the protective reactions of the organism to injection of a foreign agent as powerful as snake venom. This facilitates the development of an active antitoxic immunity and may be important in protecting the organism against repeated poisoning with this venom.

LITERATURE CITED

1. M. Elisuiskii, in the book: Collection of Abstracts and Separate Papers on Epidemiology, Microbiology, and Hygiene from the Institute of Epidemiology, Microbiology, and Hygiene of the Ministry of Health of the Azerbaijan SSR (1944-1954) [in Russian], Baku (1956), p. 151.
2. A. S. Imamaliev, Byull. éksp. biol., 32, 1, 78 (1951).
3. A. S. Imamaliev, Khirurgiya, 2, 139 (1958).
4. I. I. Nikol'skaya and É. I. Budovskii, Vopr. med. Khim., 1, 73 (1962).
5. I. I. Nikol'skaya, N. M. Shalina, and É. I. Budovskii, Biokhimiya, 5, 759 (1963).
6. E. N. Pavlovskii, F. F. Talyzin, I. A. Val'tseva, et al., DAN SSSR, 142, 6, 1428 (1962).
7. P. V. Simakov, Vopr. Pitaniya, 6, 69 (1960).

8. A. S. Spirin, Biokhimiya, 5, 656 (1958).
9. F. F. Talyzin, K. I. Matveev, T. E. Kalinina, et al., in the book: Problems in Regional, General, and Experimental Parasitology and Medical Zoology, Vol. 8 [in Russian], Moscow (1953), p. 199.
10. F. F. Talyzin, L. B. Yurkova, M. V. Dalin, et al., Byull. éksp. biol., 5, 45 (1964).
11. M. Feldman, A. Globerson, and D. B. Nachtigel, in the book: Mechanisms of Immunological Tolerance, Prague (1962), p. 305.
12. N. Hasek and J. Hort, in the book: Mechanisms of Immunological Tolerance, Prague (1962), p. 143.

All abbreviations of periodicals in the above bibliography are letter-by-letter transliterations of the abbreviations as given in the original Russian journal. *Some or all of this periodical literature may well be available in English translation.* A complete list of the cover-to-cover English translations appears at the back of the first issue of this year.
