HETEROLOGOUS ANTIVENIN IN NEUTRALIZATION OF NORTH AMERICAN CORAL SNAKE VENOM

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NEUTRALIZATION of heterologous venoms by snake antivenins is well known. The importance of this paraspecific action in antivenin production has been discussed by Criley (*1*) and Grasset (*2*). It has also been shown, most recently by Keegan and Yoshino (*3*), that the relationship of snake species involved is not always a practical basis for accurate prediction of neutralization. Actual performance of a neutralization test or use of the antivenin in therapy are the only sure methods of determining whether paraspecificity exists.

Presented here are results of neutralization tests conducted to determine whether an antivenin produced with venom of a South American snake of genus *Micrurus* would neutralize venom of the North American coral snake, *Micrurus fulvius fulvius* (Linnaeus, 1766). Although Shannon (*4*), and undoubtedly many others, speculated as to whether such neutralization would take place, test results to support such speculation have never been published.

Materials and Methods

Dried coral snake venom used in this study was obtained from the Miami Serpentarium, Miami, Fla. (lot No. 1256).

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Sôro Antielapídico, the antivenin tested, is a product of the Instituto Butantan, São Paulo, Brazil. This antivenin is in liquid form, and is packaged in 10-ml. ampules. According to information in a brochure supplied by the producer, Sôro Antielapídico is a solution of specific globulins of serums from horses hyperimmunized against venom of *Micrurus corallinus*, a snake which is widely distributed in Brazil. Printed instructions on each box of antivenin state that the product is for use in treatment of bites by *M. corallinus* and also by *Micrurus frontalis*, a coral snake found in southern Brazil and Argentina. All antivenin used in this study was of lot No. 49 (expiration date, May 8, 1964).

The 14–17 gm. white mice used in all venom potency and neutralization tests were of a single strain supplied by the Euers Mouse Farm, Austin, Tex.

A stock solution of venom for potency and neutralization tests was prepared by dissolving the dried venom in physiological saline in the amount of 10 mg. per milliliter.

Potency tests were conducted by injecting varying amounts of venom in a constant volume of 0.5 ml. of saline solution intraperitoneally into white mice. Injections were made with a 24-gauge needle into the lower left quadrant of the abdomen. Mice receiving equal amounts of venom were caged together and observed for 24 hours. At this time the LD_{50} of the venom, expressed in terms of milligrams per mouse, was determined by the Reed-Muench (*5*) cumulation method. The method of Pizzi (*6*) was used in determining the standard error of the log of the LD_{50}.

Potency of North American coral snake venom and its neutralization by Brazilian coral snake antivenin in white mice

Series No.	Venom potency		Antivenin potency			
	LD_{50} (mg. per mouse)	Standard error log LD_{50}	LD_{50} for mice protected by 0.25 ml. of antivenin (mg. of venom)	Standard error log LD_{50}	LD_{50}'s neutralized by—	
					0.25 ml.	1 ml.
1	0. 0115	0. 0952	0. 218	0. 0440	18. 0	72. 0
2	. 00971	. 0406	. 178	. 0300	17. 3	69. 2

Neutralization tests were conducted by injecting varying amounts of venom (dissolved in 0.25 ml. saline) mixed with a constant volume of 0.25 ml. of undiluted antivenin into white mice. The venom-saline solutions and antivenins were mixed and allowed to stand at room temperature (approximately 80° F.) for 1 hour before injection. Mice which received equal amounts of venom were caged together. Test animals were observed for a period of 24 hours. Survival at this time was the criterion for determination of neutralization. Venom potency and neutralization tests were performed on the same day in each of two series of tests.

Results

Preliminary tests having shown that 0.25 ml. of antivenin would neutralize between 10 and 20 mouse LD_{50}'s of venom, two complete neutralization tests were performed with appropriate dilutions of venom and antivenin. Results of these tests are shown in the table.

In computing the number of LD_{50}'s neutralized by 0.25 ml. of antivenin, it was assumed that one LD_{50} of venom was not neutralized. Hence, the number of LD_{50}'s of venom in the neutralization test was decreased by one in calculating the number of LD_{50}'s neutralized by 0.25 ml. of antivenin. For example: In test No. 1 the LD_{50} for mice protected by 0.25 ml. of antivenin was 0.218 mg. of venom. The LD_{50} for unprotected mice was determined to be 0.0115 mg. Hence, the quantity 0.218 mg. represents 19.0 LD_{50}'s (0.218/0.0115). Since 50 percent of the mice were killed, 1 LD_{50} was not neutralized, and 18.0 LD_{50}'s were neutralized by the protective dose of antivenin. The number of LD_{50}'s neutralized by 1 ml. of antivenin was obtained by multiplication.

These evaluations showed that Sôro Antielapídico offered a high degree of protection to white mice against venom of the North American coral snake. This protection was of the same level as that reported by Criley (*1*) for a polyvalent antivenin against venoms of crotalid snakes of the United States.

Summary

Sôro Antielapídico, an antivenin produced by the Instituto Butantan, Sáo Paulo, Brazil, for treatment of bites by the South American coral snakes *Micrurus corallinus* and *Micrurus frontalis* was effective in protecting white mice against effects of intraperitoneal injections of venom of the North American coral snake, *Micrurus fulvius fulvius* (Linnaeus, 1766). The order of protection given was comparable to that conferred against venoms of North American crotalid snakes by a polyvalent antivenin produced in the United States.

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Eighth International Cancer Congress Announced

The Eighth International Cancer Congress will be held in Moscow, U.S.S.R., July 22–28, 1962, under the sponsorship of the International Union Against Cancer.

Papers are invited on experimental or clinical aspects of cancer or on cancer control. These papers must not have been published or presented prior to July 22, 1962. An abstract of not more than 250 words of each paper should be provided. Authors should send an original and two carbon copies to the Soviet National Organizing Committee, Moscow, and another original and five legible copies to the U.S.A. National Committee on the International Union Against Cancer.

Special facilities will be available for motion pictures and technical exhibits. Applicants planning to use such facilities should submit in sextuplet a brief description of their material in the same style as abstracts of papers.

Travel allotments will be available to a limited number of scientists and physicians residing in the United States. These grants will cover round trip economy jet fares, an 11-day per diem allowance, and reimbursement for the registration fee.

Applications for travel allotments should be submitted in sextuplet to the chairman of the U.S.A. National Committee on the International Union Against Cancer. Applications should be in the form of letters, giving age, training, academic or professional title, and institutional affiliation of the applicant, and the titles of his five most pertinent publications in cancer or related fields within the last 5 years. Six reprints of the work he considers his major contribution during this period should be included. Applications should be countersigned by the department director or administrative officer of the institution with which the applicant is affiliated.

Applicants for travel funds who are submitting papers for presentation at the congress should include with their applications an original and five copies of an abstract of their papers, as described above. Applicants who are not planning to present papers should include six copies of an abstract of their major current investigative work.

Those invited to participate in symposia or in other meetings sponsored by the International Union Against Cancer should write to the chairman of the union, in sextuplet, giving full particulars, indicating what additional expenses, if any, will be involved, and enclosing abstracts of their presentations, also in sextuplet.

Applicants will be responsible for their own passports, visas, registration, travel arrangements, and hotel reservations.

Applications should be addressed to the Chairman, U.S.A. National Committee on the International Union Against Cancer, 2101 Constitution Ave., NW., Washington 25, D.C.

The registration fee is $30, and the deadline for registration without payment of a penalty is April 1, 1962. The deadline for submission of abstracts and applications for travel allotments is November 1, 1961.

An information brochure on submission of papers and other details, and application forms for registration are available on request to Professor L. M. Shabad, General Secretary, Soviet National Organizing Committee, Academy of Sciences of the U.S.S.R., 14 Soljanka, Moscow, or to Dr. Harold F. Dorn, General Secretary, International Union Against Cancer, National Institutes of Health, Public Health Service, Bethesda 14, Md.