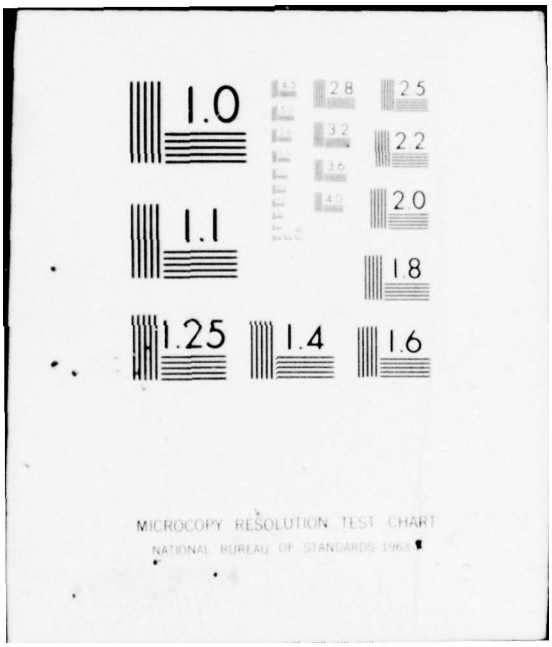
AD-A034 590 EDGEWOOD ARSENAL ABERDEEN PROVING GROUND MD F/G 6/20
RESISTANCE OF THE OPOSSUM (DIDELPHIS VIRGINIANA) TO ENVENOMATIO--ETC(U)
DEC 76 R M WERNER, J A VICK

UNCLASSIFIED EB-TR-76104 NL

1 OF 1
AD A034590

END

DATE FILMED
2-77


1.0
1.1
1.25
1.4
1.6
2.8
2.5
3.2
2.2
3.6
2.0
4.0
1.8
MICROCOPY RESOLUTION TEST CHART
NATIONAL BUREAU OF STANDARDS 1963

AD

12

B.S.

EDGEWOOD ARSENAL TECHNICAL REPORT

EB-TR-76104

RESISTANCE OF THE OPOSSUM (*DIDELPHIS VIRGINIANA*) TO ENVENOMATION BY SNAKES OF THE CROTALIDAE FAMILY

by

Robert M. Werner, CPT, VC
James A. Vick, LTC, MSC

Biomedical Laboratory

December 1976

DEPARTMENT OF THE ARMY
Headquarters, Edgewood Arsenal
Aberdeen Proving Ground, Maryland 21010

Disclaimer

The findings in this report are not to be construed as an official Department of the Army position, unless so designated by other authorized documents.

Disposition

Destroy this report when it is no longer needed. Do not return it to the originator.

UNCLASSIFIED

SECURITY CLASSIFICATION OF THIS PAGE (When Data Entered)

REPORT DOCUMENTATION PAGE

READ INSTRUCTIONS BEFORE COMPLETING FORM

1. REPORT NUMBER: EB-TR-76104	2. GOVT ACCESSION NO.
3. RECIPIENT'S CATALOG NUMBER	
4. TITLE (and Subtitle): RESISTANCE OF THE OPOSSUM (*DIDELPHIS VIRGINIANA*) TO ENVENOMATION BY SNAKES OF THE CROTALIDAE FAMILY.	5. TYPE OF REPORT & PERIOD COVERED: Technical Report. December 1974–August 1975.
	6. PERFORMING ORG. REPORT NUMBER
7. AUTHOR(s): Robert M. Werner, CPT, VC James A. Vick, LTC, MSC	8. CONTRACT OR GRANT NUMBER(s)
9. PERFORMING ORGANIZATION NAME AND ADDRESS: Commander, Edgewood Arsenal Attn: SAREA-BL-VA Aberdeen Proving Ground, Maryland 21010	10. PROGRAM ELEMENT, PROJECT, TASK AREA & WORK UNIT NUMBERS: Task 1W762718AD2104
11. CONTROLLING OFFICE NAME AND ADDRESS: Commander, Edgewood Arsenal Attn: SAREA-TS-R Aberdeen Proving Ground, Maryland	12. REPORT DATE: December 1976 13. NUMBER OF PAGES: 19
14. MONITORING AGENCY NAME & ADDRESS (if different from Controlling Office)	15. SECURITY CLASS. (of this report): UNCLASSIFIED 15a. DECLASSIFICATION/DOWNGRADING SCHEDULE: NA

16. DISTRIBUTION STATEMENT (of this Report)

Approved for public release; distribution unlimited.
Copies available from: National Technical Information Service, Springfield, Virginia 22151.

17. DISTRIBUTION STATEMENT (of the abstract entered in Block 20, if different from Report)

18. SUPPLEMENTARY NOTES

Animal studies in relation to chemical agents

19. KEY WORDS (Continue on reverse side if necessary and identify by block number)

Opossum	Viperidae
Envenomation	Hydrophidae
Crotalidae	Resistance
Elapidae	

20. ABSTRACT (Continue on reverse side if necessary and identify by block number)

Whether the North American opossum, *Didelphis virginiana*, has a natural resistance to envenomation by 12 species of snakes from the families Crotalidae, Viperidae, Elapidae, and Hyprophidae was studied. Challenge of the anesthetized opossum was by actual snakebite or intravenous injection of 4 to 20 times the dose known to be lethal to susceptible mammals. Some animals that survived the snakebite were given venom intravenously 40 to 90 minutes after the bite. Heart and respiratory rate, EKG, and blood pressure were monitored for 2 hours after envenomation, and surviving animals were observed for signs of poisoning for 30 days. The opossum

DD FORM 1 JAN 73 1473 EDITION OF 1 NOV 65 IS OBSOLETE

UNCLASSIFIED

SECURITY CLASSIFICATION OF THIS PAGE (When Data Entered)

401007

20. ABSTRACT (Contd.)

survived the venom of the following snakes of the Crotalidae family: eastern diamondback rattlesnake, western diamondback rattlesnake, copperhead, cottonmouth moccasin, Mexican copperhead, and Central American moccasin. It died when challenged with the venom of the Indian cobra, Chinese cobra, coral snake, cape cobra, puff adder, and sea snake. These findings show promise of the opossum being an excellent experimental animal for venom research, particularly by identifying its protective mechanisms.

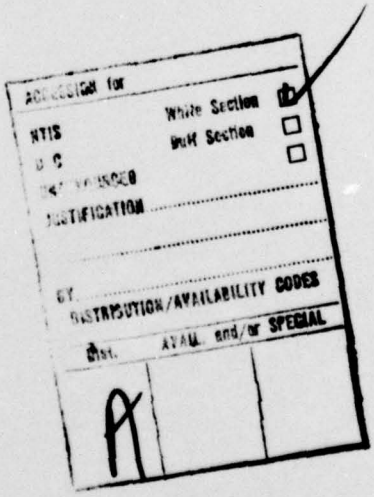
PREFACE

The work described in this report was authorized under Task 1W762718AD2104, Animal Studies Related to Chemical Agents. This work was started in December 1974 and completed in August 1975.

In conducting the research described in this report, the investigators adhered to the "Guide for the Care and Use of Laboratory Animals" as promulgated by the Committee on Revision of the Guide for Laboratory Animals Facilities and Care of the Institute of Laboratory Animal Resources, National Research Council.

Acknowledgment

The authors acknowledge the technical assistance of Mr. Harry Froehlich and the assistance of Mrs. Marion Royston in the preparation of this manuscript.



CONTENTS

LIST OF TABLES

Tables

LIST OF FIGURES

Figures

RESISTANCE OF THE OPOSSUM (*DIDELPHIS VIRGINIANA*) TO ENVENOMATION BY SNAKES OF THE CROTALIDAE FAMILY

I. INTRODUCTION.

The opossum, *Didelphis virginiana*, has received a good deal of attention in recent years as an experimental animal, with highly varied applications to scientific investigations.[1] From their evolutionary history, marsupials represent a specialized side branch of the mammalian class, diverging from mammalian stocks at an early stage. The opossum is considered a primitive member of the class Mammalia which developed to its present form in the Cretaceous period.

Our interest in the opossum began when Mr. Jack Kilmon, a Baltimore herpetologist, told us that the opossum is believed to eat venomous snakes and may be resistant to snake venom.[2] Certainly, this slow moving, nocturnal animal would be an easy target for snakes and probably would have difficulty surviving if it had no protective mechanism. If the opossum did show resistance, it might prove to be useful as an experimental animal for venom and therapy research.

A search of the literature revealed that the resistance of various animals to snake venom is well documented. However, most of the reports deal with the resistance of reptiles.[2-8] As an example, Philpot and Smith[8] showed that the king snake, *Lampropeltis getulus*, has a high degree of resistance to several snake venoms. They also reported that the serum of the king snake has a greater ability to neutralize venom from the water moccasin, *Agkistrodon piscivorus*, when mixed *in vitro* and injected into mice or injected separately from the venom than does Wyeth's polyvalent antivenin. The goal of this field of research is to discover a naturally occurring antidote to venom or to develop an antiserum that can be obtained more easily than the present method of preparing antivenins by immunizing horses over a period of months.

There are some reports of the resistance of mammals to venomous snakebite.[9-11] Vellard has done an extensive study of the resistance of various South American mammals, including species of Didelphis. Our study was undertaken to investigate whether the North American opossum is resistant to snake venoms and to single out, if possible, to what species of snakes this resistance extends.

We have established a colony of opossums, which adapt well and seem content when confined in standard cages for other animals of equal size (about 8 to 10 pounds). They thrive on a diet of regular commercial cat food. Our colony consists of opossums that were captured south of Edgewood, Maryland, on the Gunpowder Neck, which is located on the western shore of the Maryland Coastal Plain Province. The only two poisonous snakes normally found in Maryland are of the family Crotalidae: the northern copperhead (*Agkistrodon contortrix mokeson*) and the eastern timber rattlesnake (*Crotalus h. horridus*).[12] Only the northern copperhead has been reported on the western shore of the Maryland Coastal Plain Province, and occasional encounters of the northern copperhead in this area could be expected.* We were, therefore, particularly interested in what effect Crotalidae venoms would have on the opossum.

II. MATERIALS AND METHODS.

The opossums were anesthetized by one of two methods. In the initial studies, the animals were given 37 mg/kg of pentobarbital sodium intraperitoneally. Additional doses of

* Harris, H. S., Curator of Herpetology, Natural Historical Society of Maryland. Personal communication. 1976.

anesthetic were administered through a catheter placed in the jugular or femoral vein. As it was difficult to maintain a constant level of anesthesia with this method, an alternate was used in subsequent studies. Ketamine HCl was administered intramuscularly at a dose of 100 mg/kg. This allowed intubation and subsequent administration of methoxyflurane with a Foregger small-animal anesthetic machine.

The opossums were envenomated either by actual snakebite into the biceps femoris muscle of the left hind leg or by intravenous or intramuscular administration of 4 to 60 times the dose of venom known to be lethal to susceptible mammals.[13] In some instances, when an animal survived a snakebite, it was also given venom intravenously 40 to 90 minutes after the bite. The live snakes were obtained from Biologicals Unlimited, Baltimore, Maryland. A lyophilized form of the venoms given intravenously was obtained from the same supplier and reconstituted with 0.9% physiological saline before use. In our studies, all of the venoms utilized were titrated for the approximate lethal dose by ip injection into mice (see table 1). In all cases, the lethal doses in milligrams per kilogram of body weight were similar to those published by other authors.[13,14] Rabbits, anesthetized with ketamine HCl and methoxyflurane, were given the venoms intravenously and served as additional venom controls (see table 1). Here, again lethal doses in the rabbits were approximately the same as those published for other species of animals.[13,14] Approximate lethal doses for susceptible animals are listed in tables 2 and 3. The doses of venom administered to test opossums were multiples of approximate lethal doses for dogs and monkeys.[13]

In the opossum, either the femoral or carotid artery was catheterized for recording arterial blood pressure with a Statham pressure transducer. Electrocardiogram, heart rate, and respiration were monitored using needle-tipped electrodes placed in either side of the chest wall and recordings were made on a Narco physiograph. Physiological functions were monitored for 2 to 8 hours after envenomation.

The opossums were given no drugs or other treatment after envenomation. Survivors were observed closely for clinical signs of toxicity for 30 days. At the end of that period, they were either released into the wild or kept as a part of the colony but were not used for further envenomation studies.

III. RESULTS.

A. Mortality.

1. Crotalidae Family. Table 2 shows the species of Crotalidae used, the approximate lethal doses normally expected for susceptible animals, the methods of challenge, the doses administered intravenously to the opossum, and the outcome. Two opossums were challenged with the venom of the cottonmouth moccasin, two were challenged with the venom of the Western diamondback rattlesnake, and two were challenged with the venom of the Eastern diamondback rattlesnake; one opossum per venom was challenged with the other three venoms. None of the animals challenged showed local signs of hemorrhage, swelling, or tissue necrosis. None of the animals died nor were there any irreversible changes in blood pressure, heart rate, respiration, or EKG.

2. Elapidae Family. Table 3 shows the elapid species used, the approximate lethal doses normally expected for susceptible animals, the method of challenge, the doses

Table 1. Assay of Specimens of Venom Used in This Study

A. MICE. (Injections were by intraperitoneal route; mice were 25 to 30 grams in weight.)

Type of venom	Amount of venom given ip	Number of mice per dose of venom	Number of mice surviving at 48 hours	Approximate lethal dose range
	mg			mg/kg
Crotalus atrox	0.1	4	3	3-10
Western diamondback rattlesnake	0.2	4	0	
	0.3	4	1	
	Saline	4	4	
Crotalus atrox	0.1	4	4	3-6
Western diamondback rattlesnake	0.2	4	0	
	0.3	4	0	
	Saline	4	4	
Crotalus adamanteus	0.05	4	4	1.6-3
Eastern diamondback rattlesnake	0.1	4	0	
	0.15	4	0	
	Saline	4	4	
Crotalus adamanteus	0.05	4	4	1.6-3
Eastern diamondback rattlesnake	0.1	4	1	
	0.15	4	0	
	0.2	4	0	
	Saline	4	4	
Agkistrodon c. contortrix	0.1	4	3	3-10
Copperhead	0.2	4	1	
	0.3	4	1	
	Saline	4	4	
Agkistrodon piscivorus	0.1	4	4	3-6
Cottonmouth moccasin	0.2	4	0	
	0.3	4	0	
	Saline	4	4	
Micrurus fulvius	0.01	4	4	0.4-1.2
Coral snake	0.02	4	2	
	0.03	4	0	
	Saline	4	4	

B. RABBITS. (Intravenous doses were given to rabbits anesthetized with methoxyflurane; one rabbit per venom was used.)

Type of venom	Dose given	Results
	mg/kg	
Naja naja Indian cobra	0.32	Death 5 minutes after injection
Naja nivea Cape cobra	0.31	Death 10 minutes after injection
Laticaudata semifasciata Sea snake	0.1	Death 45 minutes after injection

Table 2. Lack of Effect of Crotalid Venoms on the Opossum

Scientific and common name of snake	Approximate lethal dose for susceptible animals*	Challenge to opossum**
	mg/kg	
Crotalus adamanteus Eastern diamondback rattlesnake	1.58	15.5 mg/kg, iv 105.3 mg/kg, im
Crotalus atrox Western diamondback rattlesnake	3.03	Snakebite followed by 13.7 mg/kg, iv 63.7 mg/kg, iv
Agkistrodon c. contortrix Copperhead	5.36	Snakebite followed by 43.4 mg/kg, iv
Agkistrodon piscivorus Cottonmouth moccasin	2.46	Snakebite Snakebite followed by 12.5 mg/kg, iv
Agkistrodon h. brevicaudata Korean mamushi		Snakebite
Agkistrodon bilineatus Central American moccasin		Snakebite

* Approximate lethal doses for dogs and monkeys.[13]
** All challenged opossums survived.

Table 3. Effects of Elapid, Viperid, and Hydrophid Venoms on the Opossum

Scientific and common name of snake	Approximate lethal dose for susceptible animals*	Challenge to opossum	Results
	mg/kg		
	Elapidae		
Naja naja Indian cobra	0.28	Snakebite 1.07 mg/kg, iv	Died in 45 min Died in 30 min
Naja naja atra Chinese cobra		Snakebite	Died in 45 min
Naja nivea Cape cobra	0.5	1.38 mg/kg, iv	Died in 1 hr
Micrurus fulvius Coral snake	0.34	1.70 mg/kg, iv	Died in 6 hr
	Viperidae		
Bitis arietans Puff adder		Snakebite	Died in 24 hr
Laticaudata semifasciata Sea snake	0.08	0.34 mg/kg, iv	Died in 45 min

* Approximate lethal doses for dogs and monkeys.[13]

administered intravenously, and the outcome. Two opossums were challenged with the venom of the Indian cobra; one opossum per venom was challenged with the other three venoms. All animals died within 1/2 to 24 hours after envenomation, and they all showed severe and irreversible changes in the physiological functions monitored.

3. Hydrophidae Family. The one opossum given sea snake venom (intravenous dose of 0.34 mg/kg) died 45 minutes after envenomation (table 3). There were no significant changes in blood pressure, EKG, or heart rate. However, there was a slow, progressive decrease in respiratory rate and volume which led to complete respiratory paralysis and death.

4. Viperidae Family. The one opossum bitten by a puff adder showed early changes in blood pressure and heart rate that returned toward control values at 8 hours; however, the animal died within 24 hours (table 3). Tissue edema and cutaneous hemorrhage at the site of the bite were evident, indicating envenomation was the cause of death.

B. Physiological Effects.

The effect of a copperhead snakebite on the physiological functions of the anesthetized opossum is shown in figure 1. Immediately after bite challenge there were subtle changes in respiration and blood pressure; however, within 5 minutes these functions had returned to control levels. Subsequent alterations in the physiological parameters did not occur until the animal was challenged intravenously with eight lethal doses of reconstituted whole venom from the copperhead 40 minutes after the original envenomation. Again, changes in blood pressure and respiration were noted, as well as a decrease in heart rate. Yet, as had occurred after the snakebite, these changes reverted to control or near control values within 15 to 30 minutes.

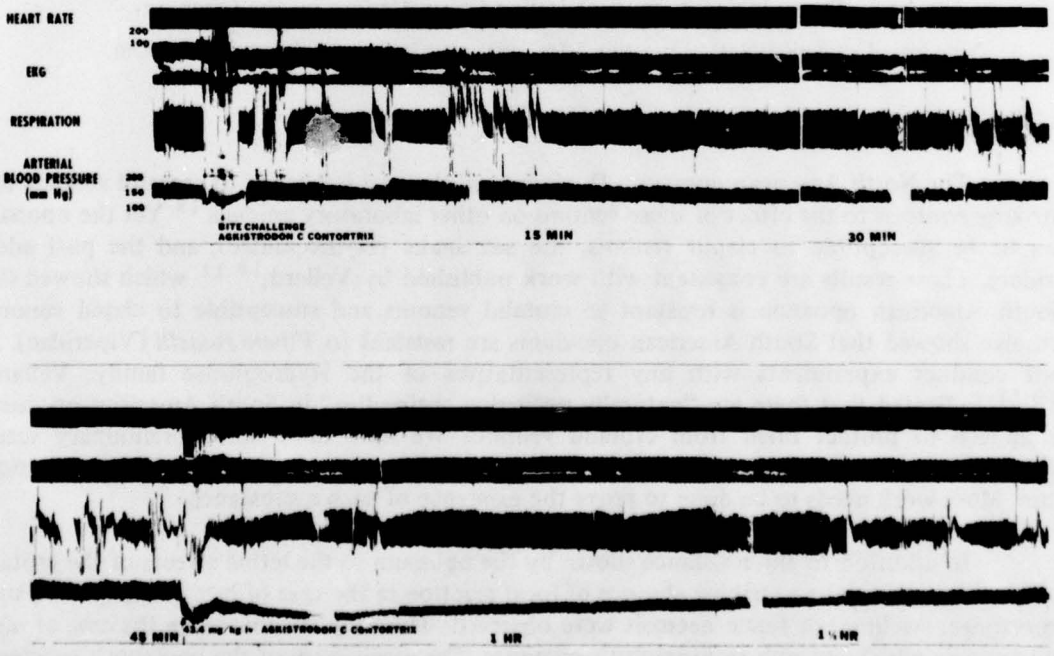


Figure 1. Physiological Effects of Copperhead Venom on the Opossum.
Note return to control levels following bite challenge as well as intravenous challenge.

Figure 2 demonstrates the effect of four lethal doses of Indian cobra venom given intravenously to the anesthetized opossum. Little or no change was noted for 15 minutes in heart rate, EKG, respiration, or blood pressure. Then blood pressure became very unstable, heart rate decreased, and respiration progressively deteriorated into complete apnea. The opossum died 30 minutes after envenomation.

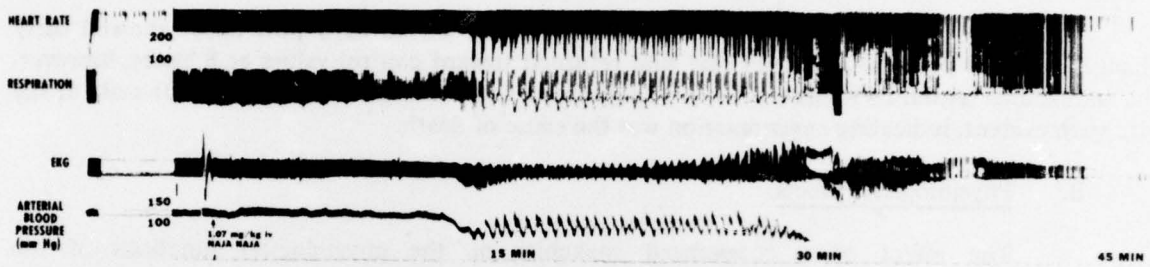


Figure 2. Physiological Effects of Indian Cobra Venom on the Opossum.

Note loss of vital physiological function 30 minutes after intravenous injection of venom.

IV. DISCUSSION.

The North American opossum, *D. virginiana*, shows a resistance to crotalid venoms that is in striking contrast to the effect of these venoms on other laboratory animals.[13] Yet the opossum appears to be susceptible to elapid venoms, the sea snake (Hydrophidae), and the puff adder (Viperidae). These results are consistent with work published by Vellard,[10,11] which showed that the South American opossum is resistant to crotalid venoms and susceptible to elapid venoms. Vellard also showed that South American opossums are resistant to *Vipera russelli* (Viperidae). He did not conduct experiments with any representatives of the Hydrophidae family. Vellard's work[10,11] indicated that there are "naturally occurring antibodies" in South American opossums which appear to protect them from crotalid venoms. We have done some preliminary serum neutralization studies that indicate a similar serum protective substance in the North American opossum. More work needs to be done to prove the existence of such a substance.

In addition to the resistance shown by the opossum to the lethal effects of the crotalid family of snakes, there was a striking absence of local reaction in the case of bite challenge. No signs of hemorrhage, swelling, or tissue necrosis were observed. This is quite unusual in the case of most crotalid venoms which are rich in proteolytic enzymes. The mechanism of the opossum's resistance to tissue damage by crotalid venoms is unknown but its elucidation could have far-reaching implications in the treatment of snakebites.

Although the lethal doses of venom Vick[13] cites were an average of those used in dogs and monkeys and Russell[14] did not mention the animals in which he derived lethal doses, the intravenous doses used in the present study were multiples of those cited by Vick and shown to be lethal in our potency assays. We therefore have no doubt of their lethality to susceptible mammals on a weight-of-venom per kilogram-of-body weight basis. The validity of results with so few animals per venom may be questioned, but the "all-or-none" response of the opossum to the crotalid and elapid venoms lends credence to the phenomenon of resistance. It is hoped that these preliminary findings will stimulate others in the field of venom research to pursue the study of the opossum's protective mechanism with the possible goal of application to humans.

LITERATURE CITED

1. Jurgelski, William. The Opossum (*Didelphis virginiana kerr*) as a Biomedical Model. Lab. Anim. Sci. *24*, 375 (1974).

2. Klauber, L. M. Susceptibility, Resistance and Immunity to Rattlesnake Venom. *In*: Rattlesnakes. L. M. Klauber, editor. pp. 883-891. University of California Press, Los Angeles, California. 1972.

3. Bonnett, D. E., and Guttman, S. I. Inhibition of Moccasin (*Agkistrodon piscivoris*) Venom Proteolytic Activity by the Serum of the Florida King Snake (*Lampropeltis getulus Floridana*). Toxicon *9*, 417 (1971).

4. Clark, W. C. Venom Neutralization by Rattlesnake Serum Albumin. Science *164*, 1402 (1969).

5. Kellaway, C. H. Snake Venom. III. Immunity. Bull. *Johns Hopkins Hosp. 60*, 159 (1937).

6. Noguchi, H. The Action of Snake Venom upon Cold-Blooded Animals. Carnegie Institute, Washington, D.C. 1904.

7. Philpot, V. B. Neutralization of Snake Venom *in Vitro* by Serum from the Nonvenomous Japanese Snake *Elaphe quadririrgata*. Herpetologica *10*, 158 (1954).

8. Philpot, V. B., and Smith, R. G. Neutralization of Pit Viper Venom by King Snake Serum. Proc. Soc. Exp. Biol. Med. *74*, 521 (1950).

9. Boquet, P. Venins de serpents et antivenins. Flammarion, Paris, France. 1948.

10. Vellard, J. Investigaciones sobre inmunidad natural contra los venenos de serpientes. Part I. Mus. Hist. Nat. 'Javier Prado', Ser A, Zool. *1*, 1-16 (1949).

11. Vellard, J. Investigaciones sobre inmunidad natural contra los venenos de serpientes. Part II. Mus. Hist. Nat. 'Javier Prado', Ser A, Zool. *4*, 1 (1950).

12. Harris, H. S. Distributional Survey: Maryland and the District of Columbia. Bull. Md. Herpetol. Soc. *11*, 73 (1975).

13. Vick, J. A. Effect of Actual Snakebite and Venom Injection on Vital Physiological Function. *In*: Toxins of Animal and Plant Origin. Vol 3. A. Devries and E. Kochva, editors. pp 1001-1011. Gordon and Breach, Science Publications, Inc., New York, New York. 1972.

14. Russell, F. E. Pharmacology of Animal Venoms. Clin. Pharmacol. Ther. *8*, 849 (1967).

DISTRIBUTION LIST 3

Names	Copies
EDGEWOOD ARSENAL	
FOREIGN INTELLIGENCE OFFICER	1
CHIEF, LEGAL OFFICE	1
CHIEF, SAFETY OFFICE	1
CHIEF, PLANS OFFICE	
Attn: SAREA-PL	1
PUBLIC HEALTH SERVICE LO	1
AUTHOR'S COPY, Biomedical Laboratory	2
BIOMEDICAL LABORATORY	
Attn: SAREA-BL-M	1
Attn: SAREA-BL-O	1
Attn: SAREA-BL-Q	1
Attn: SAREA-BL-R	1
Attn: SAREA-BL-RC	1
Attn: SAREA-BL-RE	1
Attn: SAREA-BL-RI	1
Attn: SAREA-BL-V	1
CHEMICAL LABORATORY	
Attn: SAREA-CL	1
Attn: SAREA-CL-B	1
Attn: SAREA-CL-BS	1
Attn: SAREA-CL-D	1
Attn: SAREA-CL-P	1
Attn: SAREA-CL-TE	1
DEVELOPMENT AND ENGINEERING DIRECTORATE	
Attn: SAREA-DE-D	1
Attn: SAREA-DE-M	1
MANUFACTURING TECHNOLOGY DIRECTORATE	
Attn: SAREA-MT-C	1
Attn: SAREA-MT-D	1
Attn: SAREA-MT-E	1
DIRECTOR OF PRODUCT ASSURANCE	
Attn: SAREA-PA-A	1
Attn: SAREA-PA-P	1
Attn: SAREA-PA-Q	1
DIRECTOR OF TECHNICAL SUPPORT	
Attn: SAREA-TS-R	2
Attn: SAREA-TS-L	3
Attn: SAREA-TS-E	1
DEPARTMENT OF DEFENSE	
Administrator Defense Documentation Center Attn: Accessions Division Cameron Station Alexandria, VA 22314	12
Director Defense Intelligence Agency Attn: DB-4GI Washington, DC 20301	1

Names	Copies
DEPARTMENT OF THE ARMY	
HQDA (DAMO-SSC) WASH DC 20310	1
Director Defense Civil Preparedness Agency Attn: RE	1
Attn: PO(DC) Washington, DC 20301	1
CINCUSAREUR Attn: AEAGC-DS APO New York 09403	1
Chief, Office of Research, Development & Acquisition Attn: DAMA-CSM-CM	1
Attn: DAMA-ARZ-D Washington, DC 20310	1
Headquarters, Sixth US Army NBC Office Attn: AFKC-OP-PN Presidio of San Francisco, CA 94129	1
HQDA (DAMI-FIT) WASH, DC 20310	1
CINCUSAREUR Attn: AEAMD-PM APO New York 09403	1
OFFICE OF THE SURGEON GENERAL	
Commander US Army Medical Bioengineering Research & Development Laboratory Attn: SGRD-UBD-A Fort Detrick, Bldg 568 Frederick, MD 21701	1
US ARMY HEALTH SERVICE COMMAND	
Commander US Army Environmental Hygiene Agency Attn: HSE-AA-L, Librarian, Bldg 2100 APG-Edgewood Area	1
Superintendent Academy of Health Sciences US Army Attn: HSA-CDC	1
Attn: HSA-IHE Fort Sam Houston, TX 78234	1

DISTRIBUTION LIST 3 (Contd)

Names	Copies
US ARMY MATERIEL DEVELOPMENT AND READINESS COMMAND	
Commander	
US Army Materiel Development and Readiness Command	
Attn: DRCDE-DM	1
Attn: DRCLDC	1
5001 Eisenhower Ave	
Alexandria, VA 22333	
Commander	
US Army Foreign Science & Technology Center	
Attn: DRXST-IS1	2
220 Seventh St., NE	
Charlottesville, VA 22901	
Commander	
DARCOM, STITEUR	
Attn: DRXST-ST1	1
Box 48, APO New York 09710	
Commander	
US Army Science & Technology Center-Far East Office	1
APO San Francisco 96328	
Project Manager for Chemical Demilitarization and Installation Restoration	
Attn: DRCPM-DR, Bldg E4585	1
APG-Edgewood Area	
Director	
DARCOM Field Safety Activity	
Attn: DRXOS-C	1
Charlestown, IN 47111	
US ARMY ARMAMENT COMMAND	
Commander	
US Army Armament Command	
Attn: DRSAR-ASN	1
Attn: DRSAR-IMB-C	1
Attn: DRSAR-RDM	1
Attn: DRSAR-RDT	1
Rock Island, IL 61201	
Commander	
US Army Dugway Proving Ground	
Attn: Technical Library, Docu Sect	1
Dugway, UT 84022	

Names	Copies
Commander	
Rocky Mountain Arsenal	
Attn: SARRM-EA	1
Attn: SARRM-MD	1
Denver, CO 80240	
Commander	
Pine Bluff Arsenal	
Attn: SARPB-ETA	1
Pine Bluff, AR 71611	
US ARMY TRAINING & DOCTRINE COMMAND	
Commandant	
US Army Missile & Munitions Cente & School	
Attn: ATSK-CD-MD	1
Attn: ATSK-DT-MU-EOD	1
Redstone Arsenal, AL 35809	
Commandant	
US Army Military Police School	
Attn: ATZN-CD	1
Fort McClellan, AL 36205	
Commander	
US Army Infantry Center	
Attn: ATZB-CD-MS-C	1
Fort Benning, GA 31905	
Commandant	
US Army Ordnance Center & School	
Attn: ATSL-CD-MS	1
Attn: ATSL-DTG	1
APG-Aberdeen Area	
US ARMY TEST & EVALUATION COMMAND	
Record Copy	
CDR, APG	
Attn: STEAP-AD-R/RHA	1
APG-Edgewood Area, Bldg E5179	
CDR, APG	
Attn: STEAP-TL	1
APG-Aberdeen Area	
Commander	
US Army Test & Evaluation Command	
Attn: DRSTE-FA	1
APG-Aberdeen Area	

DISTRIBUTION LIST 3 (Contd)

Names	Copies
Commander US Army Tropic Test Center Attn: STETC-MO-A (Tech Library) APO New York 09827	1
Commander Dugway Proving Ground Attn: STEDP-PO Dugway, UT 84022	1
DEPARTMENT OF THE NAVY	
Chief of Naval Research Attn: Code 443 800 N. Quincy Street Arlington, VA 22217	1
Commander Naval Explosive Ordnance Disposal Facility Attn: Army Chemical Office Indian Head, MD 20640	1
Commander Naval Surface Weapons Center Attn: Tech Lib & Info Svcs Br White Oak Laboratory Silver Spring, MD 20910	1 1
Commander Naval Surface Weapons Center Dahlgren Laboratory Attn: DX-21 Dahlgren, VA 22448	1
Commander Nuclear Weapons Training Group, Atlantic Naval Air Station Attn: Code 21 Norfolk, VA 23511	1
Chief, Bureau of Medicine & Surgery Department of the Navy Attn: CODE 5 Washington, DC 20372	1
Commander Naval Weapons Center Attn: Technical Library/Code 233 China Lake, CA 93555	1

Names	Copies
DEPARTMENT OF THE AIR FORCE	
HQ Foreign Technology Division (AFSC) Attn: PDRR Wright-Patterson AFB, OH 45433	1
Commander Aeronautical Systems Division Attn: ASD/AELD Wright-Patterson AFB, OH 45433	1
HQ, USAF/SGPR Forrestal Bldg WASH DC 20314	1
Commander Armament Development & Test Center Attn: DLOSL (Technical Library) Eglin AFB, FL 32542	1
NORAD Combat Operations Center/DBN Cheyenne Mtn Complex, CO 80914	1
OUTSIDE AGENCIES	
Director of Toxicology National Research Council 2101 Constitution Ave., NW Washington, DC 20418	1