

ANTIBODY RESPONSES OF COYOTES INOCULATED WITH VENEZUELAN EQUINE ENCEPHALITIS VIRUS

Authors: LUNDGREN, D. L., and SMART, K. L.

Source: Bulletin of the Wildlife Disease Association, 5(1) : 39-42

Published By: Wildlife Disease Association

URL: <https://doi.org/10.7589/0090-3558-5.1.39>

BioOne Complete (complete.BioOne.org) is a full-text database of 200 subscribed and open-access titles in the biological, ecological, and environmental sciences published by nonprofit societies, associations, museums, institutions, and presses.

Your use of this PDF, the BioOne Complete website, and all posted and associated content indicates your acceptance of BioOne's Terms of Use, available at www.bioone.org/terms-of-use.

Usage of BioOne Complete content is strictly limited to personal, educational, and non - commercial use. Commercial inquiries or rights and permissions requests should be directed to the individual publisher as copyright holder.

BioOne sees sustainable scholarly publishing as an inherently collaborative enterprise connecting authors, nonprofit publishers, academic institutions, research libraries, and research funders in the common goal of maximizing access to critical research.

ANTIBODY RESPONSES OF COYOTES INOCULATED WITH VENEZUELAN EQUINE ENCEPHALITIS VIRUS

The geographical distribution of Venezuelan equine encephalitis (VEE) virus includes many South and Central American countries and the southeastern and possibly the western desert areas of the United States (Sidwell, *et al.*, 1967. *Bacteriol. Rev.*, 31:65-81). As summarized by these workers, the VEE virus has a wide host range that includes domestic animals, birds, a marsupial, bats, rodents, and carnivores. To this list has been added the coyote (Lundgren and Smart, 1968. *Am. J. Trop. Med. and Hyg.*, in press) which has various subspecies distributed throughout most of North and Central America (Hall and Kelson, 1959. *Mammals of North America*. Vol. II:843-846. The Ronald Press Co.) where they range into areas where VEE is known to occur.

We reported that coyote pups, *Canis latrans lestes* Merriam, were susceptible to experimental infections with as few as $10^{1.7}$ 21-day old mouse intracerebral 50% lethal (MICLD₅₀) doses of VEE virus as determined by the development of a viremia (Lundgren and Smart, *op. cit.*). Because of the magnitude and duration of the viremia in the experimentally infected coyotes, it was concluded that this animal could have a role in the cycle of VEE in nature. To better understand the possible role of this animal in the epidemiology of VEE it was suggested that coyotes be included in future serological surveys for the incidence of VEE in wildlife, especially in areas where the virus is known to be endemic. To aid in interpreting the results of such a survey, the present study of the antibody responses of the experimentally infected coyotes mentioned above was undertaken using VEE and related antigens.

Materials and Methods

Animal collection and handling have been described (Lundgren and Smart, *op. cit.*). Briefly, the coyotes were divided into two different age groups which were in turn divided into subgroups of six animals each that were then inoculated with different ten-fold serial dilutions of VEE virus by the subcutaneous route. Fifty-four animals in one group (nine subgroups of six each) were inoculated when 1 to 2 months of age, and 18 pups in a second group (three subgroups of six each) were inoculated when 6 to 7 months of age. The younger coyotes were inoculated with virus concentrations ranging from $10^{1.7}$ to $10^{9.7}$ mouse intracerebral MICLD₅₀ and the older coyotes with $10^{0.2}$, $10^{3.2}$ and $10^{6.2}$ MICLD₅₀. Sera were collected from blood drawn from the jugular vein. All coyotes were bled prior to inoculation and at 1, 2, 3, 4 and 6 weeks after inoculation. Seven of the 1 to 2 month old coyotes were selected at random from the surviving animals and were also bled for sera at 7, 21, 31 and 41 weeks. The latter group consisted of one animal inoculated with $10^{9.7}$, three with $10^{6.7}$, two with $10^{3.7}$, and one with $10^{1.7}$ MICLD₅₀ doses of VEE virus. All sera were stored in a mechanical freezer at -65°C until tested. In most instances all of the different sera collected from one coyote were tested at the same time to minimize the minor day to day variations in serology.

The sera were tested for hemagglutination inhibiting (HI) with VEE, Western equine encephalitis (WEE) and Eastern equine encephalitis (EEE) antigens and VEE virus neutralizing (SN) antibody by standard procedures (Hammon and Work, 1964. In Lennette and Schmidt (ed.) *Diagnostic Procedures for Viral and Rickettsial Diseases*: 268-311. Third Ed. American Public Health Association, Inc.). The kaolin

adsorption technique was used for the treatment of sera for the HI test. The complement fixation (CF) test for VEE antibody was done according to the procedures outlined by Thorpe, *et al.*, 1965 (Proc. Soc. Exptl. Biol. and Med., 118:179-181).

Results and Discussion

The antibody response of the seven coyotes held for 41 weeks after the inoculation of VEE virus are illustrated in Figures 1 and 2. The VEE HI antibody responses were rapid with peak titers reached by one week after inoculation (Figure 1). The VEE CF antibody was not detectable until the second week and peak titers were reached by the third week. Significant VEE SN antibody levels were detectable by one week and reached peak neutralizing indices at three weeks. All antibody levels remained within the same general ranges during the 41 weeks that these animals were observed. These findings are similar to the antibody responses in man except that in humans the CF titer tends to decrease a few months after infection (Hammer and Work, *op. cit.* and Work, 1964. In Lennette and Schmidt (ed.) Diagnostic Procedures for Viral and Rickettsial Diseases: 312-355. Third Ed. American Public Health Association, Inc.).

In the coyotes antibody that cross reacted with WEE and EEE antigens in the HI test reached peak titers at three weeks as compared to the one week for the VEE antibody (Figure 2). The WEE and EEE HI titers were lower at the seventh week but the titers at 21 and 31 weeks remained in the same general range. At 41 weeks the WEE titers had decreased only slightly while nearly all of the EEE titers were negative. The magnitude of these cross reactions is similar to that observed in mice (Casals and Brown, 1954. J. Exptl. Med., 99:429-449). The VEE CF and HI antibody responses and the WEE and EEE HI cross reacting responses of all other coyotes, both the younger and older animals, that developed a viremia fell within the ranges of titers illustrated in Figures 1 and 2. No SN antibody determinations were made in these animals.

Two of the 1 to 2 month old coyotes that were in the subgroup that received the lowest dose of virus, $10^{1.7}$ MICLD₅₀, were the only animals in this group that did not develop a viremia (Lundgren and Smart, *op. cit.*). One of these coyotes developed only transient low levels of CF and HI titers and the second developed titers in the same ranges as those of the infected animals. Two of the 6 to 7 month old coyotes inoculated with the lowest dose of virus, $10^{0.2}$ MICLD₅₀, given in their age group did not develop a viremia. Both animals, however, developed CF and HI antibody titers at levels similar to the animals that had demonstrable viremia.

It is evident from this study that VEE virus infection in coyotes can readily be diagnosed by any one of the three methods used within at least 41 weeks of the initial exposure. These findings again support the need expressed by others for the use of more than one antigen when conducting a serological survey for the incidence of arboviruses (Hammon and Work, *op. cit.* and Sidwell, *et al.*, *op. cit.*).

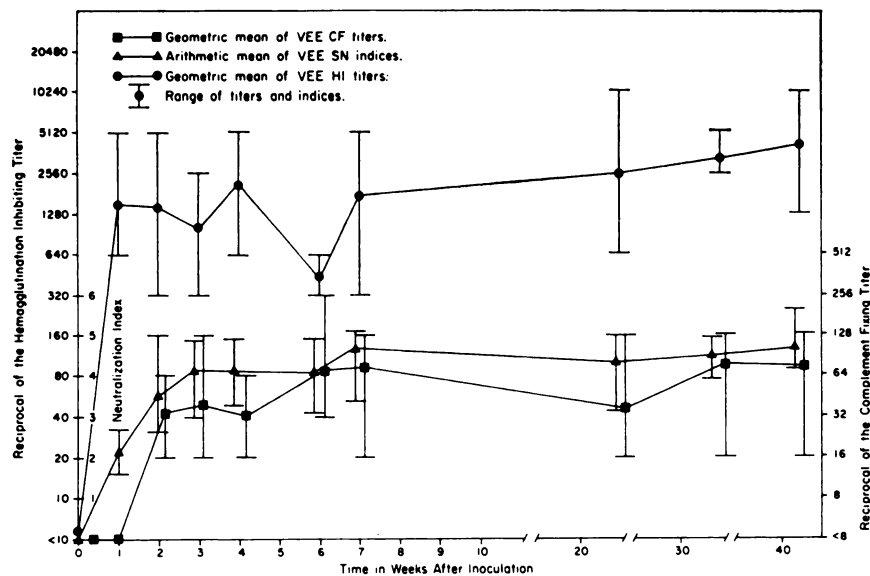


Figure 1 Complement fixing (CF), hemagglutination inhibiting (HI), and neutralizing (SN) antibody responses of seven coyote pups after the inoculation of Venezuelan equine encephalitis (VEE) virus

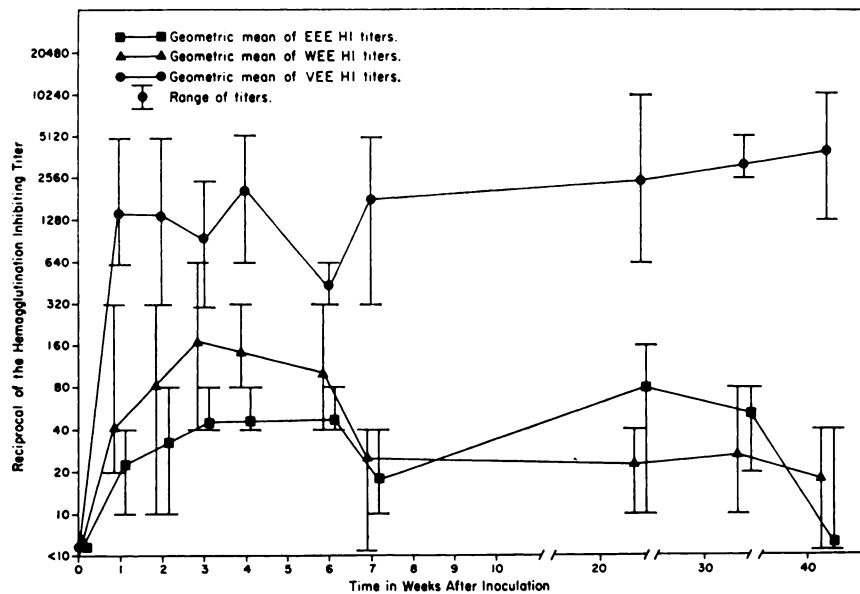


Figure 2. Hemagglutination inhibition (HI) of Venezuelan equine encephalitis (VEE), Western equine encephalitis (WEE) and Eastern equine encephalitis (EEE) viruses by the sera of seven coyotes inoculated with VEE virus.

Acknowledgements

The authors express their appreciation to Dr. B. D. Thorpe for the CF antibody determinations, to Mr. R. F. N. Woo for the HI antibody determinations, and to Mr. D. R. Terry for his assistance. The experimental animals used in these studies were housed, fed, and cared for in a humane manner and such care was supervised by a competent biologist in accordance with principles of laboratory animal care established by the National Society for Medical Research.

D. L. LUNDGREN¹ and K. L. SMART

*Ecology and Epidemiology Division, Desert Test Center
Dugway, Utah 84022*

November 1, 1968

1. Present address: *Department of Microbiology,
The Lovelace Foundation for Medical
Education and Research
Albuquerque, New Mexico 87108*

ACCOMMODATIONS LIST FOR 1969 ANNUAL WILDLIFE CONFERENCE Ames, Iowa, June 16-20

Participants: Please make your reservations directly with the Motel/Hotel. The number of each place corresponds to the position numbers on the accompanying map. All motels are of equally high standard, but vary in price somewhat. Single rooms range from \$8.50 to \$10.50, and double occupancy ranges between \$12.00 to \$18.00. It would be helpful to indicate your price limitation on your reservation request. The closest motel is approximately three miles from the National Animal Disease Laboratory. A special limited bus service to operate in the morning (8-9 A.M.), noon (12:30-1:30 P.M.), and evening (5-6 P.M.) will be available June 17-20 for participants.

- 1 AMES MOTOR LODGE 30 Units
Large single, double or 2-room, air conditioned, individually controlled electric heat, courtesy coffee, direct dial phones, TV, carpeted. Kitchenettes available. 2 blocks east of Junction of Highways 30 and 69. Phone 232-4315.
- 2 AMES TRAVELODGE 50 Units
Individually controlled heating and air conditioning, TV, heated pool, private patios, direct dial, extra length beds. On U.S. 69 & 30 (229 S. Duff). Phone 233-1714, Area 515.
- 3 CAMPUS COURT MOTEL 17 Units
Brick exterior. Carpeted. TV, air-conditioning and room phones. Reasonable rates. Seven blocks west of Iowa State University. 3405 Lincolnway. On U.S. 30 west. Phone 292-2116.
- 4 CASEY MOTEL 14 Units
All large carpeted rooms. Automatic heat, air conditioning, TV, room phones. Shaded playground, very quiet area. Close to North Grand Shopping Center. 2 miles north of Ames on Highway 69 and Dawes Drive. Your hosts, Mr. and Mrs. A. Stein. Phone 232-8854.
- 5 EL RANCHO MOTEL AAA 12 Units
Free TV, room phones, room controlled heat and air-conditioning, carpeted, inn room coffee, complete soft water baths in ceramic, massage mattresses, AAA restaurant nearby. Cr. cards honored. 5 min. to ISU. 1½ miles south on U.S. 79 at Airport Rd. Phone 232-3732.
- 6 HOLIDAY INN OF AMES, IOWA 121 Units
Dial 232-0280. Free advance reservations. On U.S. 69 & 30. 3 miles from 1-35. 316 South Duff Avenue. 6 blocks from downtown, free transportation. Heated pool. TV and Music, carpeted, tile bathrooms and direct dial phones in each room. 24 hr. restaurant. Beautiful new Cyclone room for banquets and meetings. Facilities to 400 people.
- 7 IOWA MOTEL 10 Units
Comfortable quiet rooms, free color TV, carpeted, room phones, hot water heat, individually controlled heat and air conditioned. In room coffee. Beauty rest mattresses, reasonable rates, close to good restaurant, ¾ miles west of University on Highway 30. Phone 292-1216.
- 8 LINCOLN LODGE 24 Units
Air conditioned, private baths, room phones, free TV. Paved parking lot. One block east of junction highway 30 & 69 on Lincoln Way. Phone 232-5464.
- 9 MALLARD MOTEL 8 Units
Close to restaurants. Quiet. Single or double. Room telephones, television. Carpeted. Central heating and air conditioning. 1 mile east of ISU on Highway 30. Phone 292-9808.
- 10 MOTEL 69 10 Units
Central heat, air conditioned, TV, radio, air foam mattresses. Kitchenettes available. Moderate rates. On Highway 69 & 30 — ½ mile south of Ames. Phone 232-5198.
- 11 NEW ENGLANDER MOTEL AAA 20 Units
West Western, breakfast restaurant, steak house adjacent. Heated swimming pool, hot water heat, air conditioned, room controlled, combination baths, carpeting, TV, room phones. 2 miles west of ISU on U.S. 30. Phone 292-1551.
- 12 NEW FRONTIER MOTEL AAA 14 Units
Conveniently located to ISU campus. Carpeted, air conditioned, phones, color TV, hot water heat, tubs and showers, inn room coffee, heated pool. Excellent restaurant with bar next door. Two miles west on Hwy. 30. Am. Exp. Phone 292-2056.
- 13 HOTEL SHELDON-MUNN 108 Units
Home of the Famous Town Cafe. Air conditioned rooms, TV, convenient free parking. Service clubs: Rotary, Lions, Kiwanis. Member Iowa Hotel & Motor Inn Association. Phone 232-6161.
- 14 SILVER SADDLE MOTEL AAA 14 Units
Tiled showers and tubs. Beauty rest mattresses, hot water heat, carpeted, room phones, TV, A.C., Inn Room courtesy coffee, swimming pool, Am. Exp. Card, Intersection 69 & 30. Phone 232-8363.