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Survey for Vesicular Stomatitis Virus Neutralizing Antibodies in Serums of White-tailed Deer *Odocoileus Virginianus* of the Southeastern United States

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Abstract

New Jersey type vesicular stomatitis (VS) antibodies were found in 14 of 677 deer serums tested by neutralization tests in embryonated chicken eggs. Twelve positive serums were received from Louisiana and two from Georgia. Eight of the positive deer serums from Louisiana were collected in the area of the only reported case of VS during 1967.

Clinical VS has not been diagnosed in the east coast states since 1964. Two positive deer serums were collected on Ossabaw Island, Georgia, and three positive serums, one each from a hog, bull, and sheep, were collected from young animals on the island. These findings indicated subclinical infection or a nidus of New Jersey VS in Georgia. The low percentage of New Jersey type VS antibodies in deer and the distribution parallel the low incidence of VS since 1964. No antibodies were present for Indiana type VS virus.

Introduction

Reports of VS antibodies in deer^{18,14,21} led us to consider this species in the search for hosts having a role in the maintenance and epizootiology of VS. Currently we have tested 677 deer serums for VS. Vesicular stomatitis neutralizing antibodies persist for many years following infection and are evidence of VS infection some time during the life of the animal.

Vesicular stomatitis is a virus disease affecting cattle, swine, horses, and occasionally people and deer. Vesicles are produced on the feet, mouth, udder or

snout of livestock and people develop symptoms resembling influenza infection. Ecological studies were made by Hanson,^{5,6} Karstad, Adams, Hanson, and Ferris,¹³ and Hanson and Karstad,¹⁴ mostly in Georgia during the fifties. During the summer of 1965, the National Communicable Disease Center investigated an outbreak of the Indiana type in New Mexico and Colorado, which included eight human cases reported by Fields and Hawkins,¹ and Sudia, Fields and Calisher.¹⁸ Jonkers¹⁰ isolated the Cocal subtype of Indiana VS and studied enzootics

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involving rodents in Trinidad, British West Indies. Forty-three percent of 122 wild Rio Grande turkeys trapped at the Welder Wildlife Refuge in south Texas during 1964-65 had antibodies for New Jersey VS virus although no clinical disease was reported in livestock.⁴ Hanson and Karstad reported that 79 percent of 58 feral swine tested in Georgia had antibodies to New Jersey VS virus.⁵ Feral swine are common in marshy areas of the coastal states from North Carolina to eastern Texas. Recently arbovirus workers in Panama have been studying VS virus.^{17, 19, 20}

Nidi of VS infection appear to exist in tropical and semi-tropical America and the viruses spread from these areas under favorable conditions. Viremia has been difficult to detect following experimental infection of animals. The absence of viremia or at most the presence of a

very transient viremia in domestic livestock and wild mammals decreases the probability that the principal transmission of VS is by blood-sucking arthropods.²⁰ Sentinel mice¹¹ and sentinel monkeys¹⁷ have on some occasions become infected.

Virus isolations have been made from arthropods: *Phlebotomous* (sandflies),¹⁶ *Gigantolaelaps* (mites),¹⁰ mosquitoes^{11, 18} and *Hippelates pusio* (eye gnats).⁸

Many investigators consider the VS viruses to be arthropod-borne, a few suggest the possibility of transmission by fungi and some think that VS is a plant virus,⁸ possibly altered by insect passage.^{9, 21} The rapid spread or simultaneous recognition of VS on premises several miles apart without apparent contact has led Jonkers to propose that VS virus is present on the premise and triggered to infect animals by favorable and as yet undefined conditions.¹²

Materials and Methods

The Southeastern Cooperative Wildlife Disease Study (SCWDS) was the source of 446 of the deer serums tested. These were collected between February 1965, and May 1969, as part of the wildlife disease studies in various wildlife management areas and military reservations of ten southeastern states: Alabama, Arkansas, Florida, Georgia, Louisiana, Maryland, Mississippi, North Carolina, South Carolina and Virginia.

The Louisiana Wildlife and Fisheries Commission, representatives of the Louisiana Livestock Sanitary Board and United States Department of Agriculture, Animal Health Division, collected 231 serums between December 1967, and February 1969.

Deer serums were tested by the neutralization test. One half (0.5) ml quantities of a 1:4 dilution of serums were mixed with equal volumes of a vesicular stomatitis virus dilution containing 100 to 1000 LD₅₀ units. The serum virus

mixtures were allowed to react 30 minutes at room temperature. Fifteen hundredths (.15) ml doses of serum-virus mixtures were then inoculated into the allantoic cavity of 8-day embryonated chicken eggs. Tris buffered tryptose broth containing 5000 units of penicillin G and 5 mg of streptomycin sulfate per ml was used as diluent for the virus and serum controls. Broth was used for serum dilutions with the following exception. Frequently equal quantities of four undiluted serums were pooled and .5 ml of the serum pool was mixed with .5 ml of virus. Test eggs were incubated at 35 C and examined daily throughout the 72 hour test period. When neutralization was detected in serum pools, the serums were tested individually. Positive serums were titrated in a fourfold dilution series and tested by complement-fixation as described previously.^{3, 7} Results were accumulative on samples received at NADL between May 1967, and February 1969.

Results

In Table 1 we have summarized the neutralization tests with New Jersey type VS. Twenty-three deer samples collected

during 1964 were negative to New Jersey type VS. There were 2 positive serums among 117 collected in 1965, 3/39 in

1966, 8/109 in 1967, 0/213 for 1968, and 1/176 for 1969. Positive samples were found only from Ossabaw Island near Savannah on the Georgia coast (2 positive serums), and from Louisiana (12 positive serums).

In Table 2 we have summarized the New Jersey virus neutralization results on the positive serums. All 677 deer serums were negative to the neutralization test for Indiana subtype I VS antibodies.

TABLE 1. Summary of deer serums tested for New Jersey type vesicular stomatitis antibodies.

State and Area	Date Bled					
	1964	1965	1966	1967	1968	1969
Alabama (65)						
Game Management Areas Marengo and Clark Co's.		0/20	0/5	0/15	0/15	0/10
Arkansas (13)						
Sylamore Exp. For. Ft. Chaffee		0/3		0/10		
Florida (72)						
Camp Blanding Davis Ranch		0/20			0/5	
Collier and Hernando Co's. Elgin A.F.B.			0/10		0/7	
				0/15	0/10	0/5
Georgia (67)						
Ft. Stuart and Fargo Ossabaw Island		0/9 2/14		0/14	0/20	0/10
Louisiana (268)						
L. S. U. and Sam Houston West Bay G.M.A.	0/18		0/9 3/5			
Saline G.M.A. Lawton Ranch		0/5		8/10		
Delta Nat. W. F. Refuge Atchafalaya Swamp					0/101	1/117 0/3
North Carolina (35)						
Daniel Boone G.M.A.				0/15	0/15	0/5
South Carolina (57)						
A.E.C., Savannah R.P., Forks G.M.A., Palmetto Bluff		0/15	0/1	0/15	0/15	0/11
Maryland (15)						
Worcester, Garrett and Allegheny Co's.					0/10	0/5
Mississippi (34)						
Hunington Point H.C. Merigold H.C., Cleveland		0/4 0/21	0/9			
Virginia (51)						
Camps A. P. Hill and Perry		0/11		0/15	0/15	0/10
Total Pos./Total Tested (677)	0/23	2/117	3/39	8/109	0/213	1/176

TABLE 2. Summary of New Jersey vesicular stomatitis positive deer serums.

State and Area	Antibody Titer Log ₁₀ [Ⓛ]
Georgia	
Ossabaw Island	2.1 (1:128)
Chatham County — bled Feb. 5, 1965	1.5 (1:32)
Louisiana	
West Bay G.M.A.	2.7 (1:512) [Ⓛ]
Allen Parish (undated, rec'd. June 1966)	2.4 (1:256)
	1.5
Saline G.M.A. LaSalle-Catahoula Parishes	1.5 [Ⓛ]
bled Dec., 1967 [Ⓛ]	1.2 (1:16)
	1.5 [Ⓛ]
	2.4
	2.1
	1.5 [Ⓛ]
	2.1
	1.5 [Ⓛ]
Delta National Waterfowl Refuge (Plaquemines Parish)	1.2
Bled Feb. 9, 1969	

- Ⓛ Endpoints are figured by the Karber method and expressed as the logarithm of the serum dilution
- Ⓛ Buck deer only
- Ⓛ Complete neutralization of the 1:512 serum dilution
- Ⓛ Age 1½ years, ages of other deer were not given

Discussion

Vesicular stomatitis positive serums were detected only from Louisiana, 12/268 and Georgia, 2/67. There were no positive deer serums among 347 serums tested from the states of Alabama (65 samples), Arkansas 13, Florida 72, North Carolina 35, South Carolina 57, Maryland 15, Mississippi 34, and Virginia 51.

These results are more significant when correlated with the known clinical appearances of New Jersey type VS. During the period 1952 through 1964, New Jersey VS was diagnosed in Georgia each year except 1960, and slightly less frequently along the entire coastal plain from North Carolina to Louisiana.^{10,8} There was a large outbreak of VS in western Georgia and eastern Alabama during the summer of 1963 with 356

laboratory confirmed infected herds, followed by a tenfold reduction in cases during 1964. Since 1964, VS has not reappeared clinically in the southeastern states except in Louisiana. During 1966, New Jersey VS appeared in Texas and progressed northward; at the same time, Indiana and New Jersey VS were being diagnosed in New Mexico and Colorado. During August 1967, the only case of VS recognized in the United States involved hogs in the Larto Lake area of La Salle Parish, Louisiana. The eight New Jersey VS positive serums were from ten deer killed in this Larto Lake area of the Saline Game Management Area (GMA) during the hunting season in December 1967.

During May 1968, New Jersey VS was diagnosed in several horses located at

the Saline GMA headquarters several miles within the GMA. During the next few months, VS was first diagnosed in horses 2 miles away, in pigs of an adjacent parish, then in swine and cattle of the Jena area (Catahoula and La Salle Parishes) about 15 miles north. No clinical cases of VS have been diagnosed in the United States since that time, 1968 through March 1970.

The West Bay GMA should be studied because three strongly positive deer serums came from that area of Allen Parish, which is one parish removed from the Saline GMA. Only one positive serum was detected among 218 collected at the Delta National Waterfowl Refuge located in Plaquemines Parish at the tip of the Mississippi River delta.

A limited bovine serum survey of 1079 serums from six states in 1969, and

serums collected from suspected VS cases in Louisiana have disclosed New Jersey VS antibodies in Sabine and Washington Parishes of Louisiana.

Two positive deer serums from Ossabow Island, Georgia, indicated a possible nidus in wildlife of coastal Georgia. Of 17 survey serums taken from the same island on July 31, 1969, 1 hog, age 3 years; 1 bull, 1½ years; and 1 goat, 4 years, had New Jersey VS neutralizing antibodies varying from 1.5 (1:32) for the bull to 2.1 (1:128) for the hog.

Deer are a helpful species for studying the ecology of VS in sylvan areas and may contribute to locating nidi of infection in wildlife. The assistance and observations of wildlife biologists, ecologists and veterinarians are solicited for studying other species in an effort to resolve the epizootiology of VS.

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