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Source: Journal of Wildlife Diseases, 38(4) : 713-719

Published By: Wildlife Disease Association

URL: https://doi.org/10.7589/0090-3558-38.4.713

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Journal of Wildlife Diseases, 38(4), 2002, pp. 713–719
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INNATE RESISTANCE TO EPIZOOTIC HEMORRHAGIC DISEASE IN WHITE-TAILED DEER

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ABSTRACT: Differences in innate disease resistance at the sub-species level have major implications for wildlife management. Two subspecies of white-tailed deer, *Odocoileus virginianus borealis* and *O. virginianus texanus* were infected with epizootic hemorrhagic disease (EHD) viruses. These viruses are highly virulent pathogens of white-tailed deer and are endemic within the range of *O. virginianus texanus* but not within the range of *O. virginianus borealis.* Two experimental infections were performed. Five *O. virginianus texanus* fawns and five *O. virginianus borealis* fawns were infected with $10^{7.1}$ median tissue culture infective doses ($TCID_{50}$) of EHD virus, serotype 1 and five of each subspecies were infected with $10^{7.1}$ $TCID_{50}$ of EHD virus, serotype 2. Infections with both EHD virus serotypes caused severe clinical disease and mortality in *O. virginianus borealis* fawns, whereas disease was mild or nondetectable in *O. virginianus texanus* fawns. Virus titers and humoral immune response were similar in both subspecies suggesting that differences in innate disease resistance explain the differences seen in clinical disease severity. In white-tailed deer, innate disease resistance may vary at the subspecies level. Should this phenomenon occur in other species, these findings have major implications for managing wildlife populations, both endangered and non-endangered, using tools such as translocation and captive propagation.

Key words: Captive propagation, epizootic hemorrhagic disease virus, EHD, hemorrhagic disease, HD, innate resistance, *Odocoileus virginianus,* translocation, white-tailed deer.

INTRODUCTION

Hemorrhagic disease (HD), the most important infectious disease of white-tailed deer (*Odocoileus virginianus*), is caused by orbiviruses in the epizootic hemorrhagic disease (EHD) virus or bluetongue (BT) virus serogroups (Nettles and Stallknecht, 1992). Of the two serogroups, the EHD viruses are most often associated with infection in white-tailed deer populations in the southeastern United States (Nettles et al., 1992). Clinical HD, initiated by viral replication in vascular endothelial cells, is characterized by intravascular thrombosis, disseminated intravascular coagulation, and hemorrhagic diathesis, often resulting in mortality (Tsai and Karstad, 1973; Howerth et al., 1988).

Spatially, HD is not uniformly distributed throughout the United States. Epizootics in northern latitudes are infrequent and characterized by severe clinical disease and mortality, whereas epizootics in southern latitudes are more frequent and often result in mild or inapparent disease (Davidson and Doster, 1997). Extreme southern latitudes in the US such as in Texas are thought to represent areas of enzootic stability, where EHD and BT viruses are endemic, but disease is rarely seen due to a near perfect host-virus relationship (Stallknecht et al., 1996). Interactions among vectors (*Culicoides* spp.), virus strain and virulence, and herd immunity have been suggested as causes for these distinct geographic patterns of HD distribution (Nettles and Stallknecht, 1992). We hypothesized that these factors have ultimately influenced evolution of the host, which results in extreme differences in

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host innate resistance (Nettles and Stallknecht, 1992). *Odocoileus virginianus borealis* is a northern white-tailed deer subspecies found outside of the known endemic area for EHD viruses (Baker, 1984), and *O. virginianus texanus* is a southern subspecies that occurs within the known EHD virus-endemic area (Baker, 1984). We infected fawns from Pennsylvania (*O. virginianus borealis*; PA fawns) and fawns from Texas (*O. virginianus texanus*; TX fawns) with both EHD virus serotypes known to occur in the United States to determine if innate resistance to clinical disease varied between these white-tailed deer subspecies.

MATERIALS AND METHODS

Pennsylvania fawns came from five counties (Lackawanna, Luzerne, Monroe, Pike, and Wayne) in northeast Pennsylvania (USA) with permission from the Pennsylvania Game Commission. Texas fawns (*O. virginianus texanus*) came from the Kerr Wildlife Management Area (Donnie E. Harmel White-tailed Deer Research Facility, Texas, USA) with permission from the Texas Parks and Wildlife Department.

Ten fawns of each subspecies were moved by 2 wk of age to an indoor facility at the University of Georgia (Athens, Georgia, USA) in June and July 2000. Fawns were first tested for antibodies to EHD and BT viruses on July 17 using agar gel immunodiffusion (Veterinary Diagnostic Technology, Inc., Wheatridge, Colorado, USA) and serum neutralization tests as previously described (Stallknecht et al., 1995). At that time PA fawns were negative for antibodies to EHD and BT viruses, whereas TX fawns had maternal antibodies to EHD, or BT viruses, or both, confirming that they were derived from EHD virus nonendemic and endemic areas, respectively. Fawns were maintained indoors and were manually restrained and bled by jugular venipuncture weekly until they were negative for EHD antibodies by both agar gel immunodiffusion (AGID) and serum neutralization tests at a dilution of $\leq$1:2.

In November, five PA and five TX fawns were experimentally infected with $10^{7.1}$ tissue culture infective doses ($TCID_{50}$) of EHD virus, serotype 1 (EHDV-1). The EHDV-1 virus used in this study was originally isolated from a blood sample obtained from a clinically ill white-tailed deer from Walton County, Georgia (USA) in 1999 and was propagated and passed once on baby hamster kidney cells (BHK_{21} cells; American Type Culture Collection, Rockville, Maryland, USA). It was serotyped as EHDV-1 by virus neutralization as previously described (Quist et al., 1997).

A separate group of five PA and five TX fawns were experimentally infected with $10^{7.1}$ $TCID_{50}$ of EHD virus, serotype 2 (EHDV-2). The EHDV-2 virus used was isolated on BHK_{21} cells from a lymph node sample from a clinically ill white-tailed deer from Clarke County, Georgia. A white-tailed deer fawn, serologically negative for antibodies to EHD and BT viruses was used to prepare the inoculum. The deer was subcutaneously (SC) and intradermally (ID) inoculated with a sonicated third passage BHK_{21} cell suspension-containing $10^{7.4}$ $TCID_{50}$. Coinciding with the first febrile episode on day 5 post-infection, the deer was anesthetized with 4.4 mg/kg of tiletamine and zolazepam (Telazol®, Fort Dodge Animal Health, Fort Dodge, Iowa, USA) intramuscularly (IM) and 340 ml of blood was collected in sodium citrate. Platelet rich plasma was removed and blood cells were washed and resuspended twice with Dulbecco's phosphate-buffered saline (DPBS). This was sonicated and frozen in 10 ml aliquots at −70 C. The viral titer of this inoculum was $10^{6.3}$ $TCID_{50}$ per ml as determined by endpoint titration. It was serotyped as EHDV-2 by virus neutralization as previously described (Quist et al., 1997).

For both inoculations, deer were sedated with approximately 0.5 mg/kg xylazine (Xylazine-100®, Butler Company, Columbus, Ohio, USA) administered IM. Half of the viral inoculum was administered SC and half ID at multiple sites on the neck. Post inoculation, sedation was reversed with 2–5 mg yohimbine (Yobine®, Lloyd Laboratories, Shenandoah, Iowa) administered IM.

Beginning 4 days pre-infection and continuing every other day through post-infection day 14 (PID 14) deer were manually restrained, examined, body temperature was measured rectally, and blood was drawn via jugular venipuncture for complete blood counts, platelet counts, plasma protein, fibrinogen, viral isolation, serology and blood coagulation analyses.

Complete blood counts were performed on a Baker System 9000 automated cell counter (Baker Instrument Corporation, Allentown, Pennsylvania). White blood cell differential counts and platelet counts were performed manually. Plasma protein was measured using a hand-held refractometer. Fibrinogen was measured by heating the plasma to 56 C for 3 min, remeasuring plasma protein post-heating, and recording the difference between the two measurements. Plasma was frozen at −70 C for later evaluation of activated partial thrombo-

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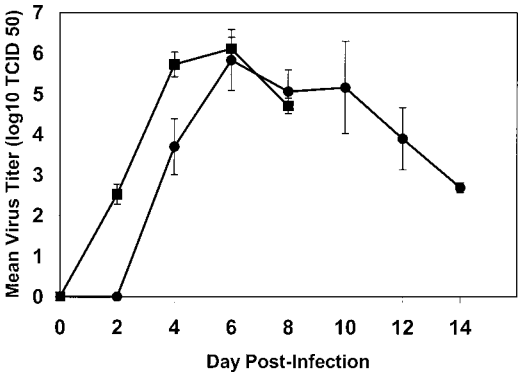

FIGURE 1. Mean virus titers and standard deviations in *O. virginianus borealis* (PA fawns), and *O. virginianus texanus* (TX fawns) experimentally infected with EHDV-1 (■=PA fawns, ●=TX fawns).

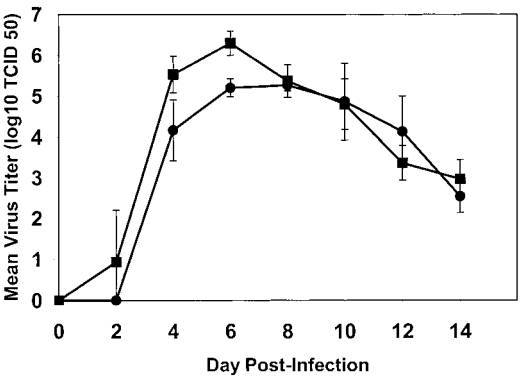

FIGURE 2. Mean virus titers and standard deviations in *O. virginianus borealis* (PA fawns), and *O. virginianus texanus* (TX fawns) experimentally infected with EHDV-2 (■=PA fawns, ●=TX fawns).

plastin time (APTT), prothrombin time (PT), and fibrin degradation product. All samples from each deer were run en-bloc using an Amelung KC4A™ Micro Coagulation Analyzer (Sigma, St. Louis, Missouri, USA) to evaluate changes in APTT and PT over the course of the experiment. Deer were tested for fibrin degradation using the D-di Test® (Diagnostica Stago, Parsippany, New Jersey, USA) as described by the manufacturer. Using blood samples collected in acid citrate, virus isolation, endpoint titration, and virus identifications were performed as previously described (Quist et al., 1997) excepting that cattle pulmonary artery endothelial (CPAE) cells (American Type Culture Collection) were used for virus isolation instead of BHK_{21} cells. Clinical disease severity scores (CDSS) were calculated for each deer based on the presence of certain physical and clinical parameters. Scatter plots were created for the following measured parameters for each serotype: plasma protein, APTT, PT, platelet count, lymphocyte count, fibrinogen, hematocrit, and rectal temperature. Deer were assigned one point for having at least one outlier in a parameter as determined by visually examining individual scatter plots that were created for each of the measured parameters. Deer were also allotted one point if they had degradation of fibrin in plasma at any time during the infection and one point for exhibiting each of the following clinical signs: depression, erythema of lightly haired areas, and salivation or oral lesions. The sum of all points per deer was taken as that deer's CDSS with 12 being the highest possible score and higher scores indicating more severe disease.

An overdose of pentobarbital solution was administered to all fawns for euthanasia (Beuthanasia®-D Special, Schering-Plough Animal Health Corporation, Union, New Jersey). Necropsies were performed on all deer within 1 hr following euthanasia or immediately upon being found dead.

Clinical severity scores were compared by *t*-test (Proc TTEST, SAS®, version 8e, SAS Institute Inc., Cary, North Carolina, USA). Viral and antibody titers, hematology results and clotting times were analyzed by analysis of variance (Proc GLM, SAS®), with the repeated measures split plot in time model including subspecies, deer (subspecies), day post-experimental infection, and day×subspecies interaction.

RESULTS

Deer became viremic by post-inoculation day (PID) 4 in both infections. Virus titers increased earlier (EHDV-1, $P<0.001$; EHDV-2, $P=0.003$) and were higher (EHDV-1, $P<0.001$; EHDV-2, $P=0.06$) in PA fawns than in TX fawns (Figs. 1, 2). Clinical disease severity scores differed between PA and TX fawns for both the EHDV-1 ($P=0.0048$) and EHDV-2 ($P=0.0032$) infections, with PA fawns exhibiting more severe clinical disease than TX fawns (Fig. 3). All five PA fawns infected with EHDV-1 and one of five infected with EHDV-2 died, whereas all TX fawns survived infection. Specifically, following EHDV-1 infection, two PA fawns became moribund on PID 7 and were euthanized. One PA fawn was found dead on PID 8, and the remaining two PA fawns

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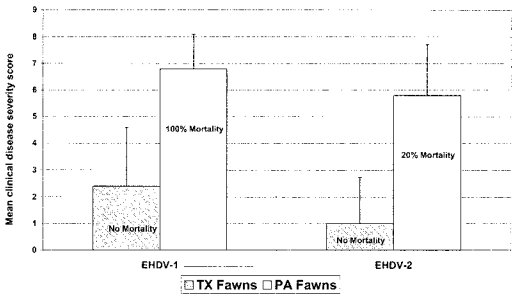

FIGURE 3. Clinical disease severity scores (mean and standard deviation) of *O. virginianus borealis* (PA fawns), and *O. virginianus texanus* (TX fawns) following experimental infection with EHDV-1 and EHDV-2 (higher scores indicate more severe disease). Scores differed between deer subspecies ($P<0.005$).

became moribund and were euthanized on PID 9. During the EHDV-2 infection, one PA fawn became moribund on PID 13 and was euthanized. All fawns exhibited postmortem lesions consistent with HD infection.

In both infections, PA fawns showed greater decreases in lymphocyte counts (EHDV-1, $P=0.053$; EHDV-2, $P<0.001$) and plasma protein concentrations (EHDV-1, $P=0.001$; EHDV-2, $P<0.001$), and greater prolongation of the APTT (EHDV-1, $P=0.006$; EHDV-2, $P=0.026$) and PT (EHDV-1, $P=0.022$; EHDV-2, $P<0.001$). In the EHDV-1 infection, PA fawns also exhibited a greater decrease in platelet counts ($P=0.076$). The humoral immune response in fawns surviving to seroconversion did not vary between subspecies ($P>0.10$).

DISCUSSION

In the past it has been thought that pathogen evolution will eventually result in a pathogen with decreased virulence for its host (Allison, 1982). This is being replaced with the idea that pathogen evolutionary adaptation to a host can result in either decreased or increased harm to the host as long as the adaptation favors successful pathogen passage into future generations (May, 1985; Lipsitch et al., 1995; Ewald, 1995). This suggests that pathogens can evolve towards increased virulence, as long as the evolution does not result in decreased pathogen transmission. In addition to changes in pathogen virulence, pathogen-host evolution also may result in changes in host resistance as has been demonstrated in the coevolution of myxoma virus and feral Australian rabbits (Parer et al, 1994; Best et al., 2000). The white-tailed deer–EHD virus association represents selection for host resistance without concurrent attenuation of viral virulence. Although EHD mortality is rarely reported in native Texas deer (Stallknecht et al., 1996), there is clear evidence that EHDV-2 viruses circulating in Texas are virulent, despite the lack of reported clinical disease in native Texas deer. In fall 2000, EHDV-2 was isolated from white-tailed deer that died of EHD after being imported into Leon and Hunt Counties, Texas, as captive breeding stock. Despite this EHD epizootic in captive imported deer, HD was not detected in local native deer (SCWDS, unpubl. data). If EHD viruses acted as a selection force in the early evolution of Texas deer, it is possible that genetic selection favored deer with innate resistance to these viruses. Conversely, the absence, or rare presence of EHD viruses probably prevented these viruses from acting as a selection force in evolution of Pennsylvania deer.

It appears that evolutionary selection for host resistance has not resulted in decreased viral transmission. Viremias were detected a day earlier in PA fawns and peaked higher than TX fawns (Figs. 1, 2), but this may not be biologically significant. Viral titers in both groups of animals were sufficient to infect *Culicoides* (Smith et al., 1996) beyond which vectorial capacity is probably more important to EHD virus transmission than subtle differences in virus titers in the vertebrate host.

The fact that humoral immune responses were nearly identical for both subspecies implies that innate resistance rather than acquired immunity is responsible for the dramatic variation in clinical disease seen between white-tailed deer subspe-

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cies. The underlying mechanism of innate resistance is unknown, but differences in cell-mediated immunity or cellular response to infection might play roles in the different clinical responses seen in the two subspecies. Inherent differences in the susceptibility of cattle and sheep microvascular endothelial cells to infection by bluetongue virus, an orbivirus closely related to EHD virus, has been suggested as a reason why this virus usually causes asymptomatic infection in cattle, but severe disease in sheep (DeMaula et al., 2001).

Differences in innate genetic resistance to diseases have been reported in wildlife at the species level. Variations in innate resistance to avian malaria have been reported in different species of native Hawaiian honeycreepers (Atkinson et al., 2000). It also has been hypothesized that innate resistance to clinical disease caused by morbillivirus infection may explain differences in the epidemiology of this virus in harbor (*Phoca vitulina*) and gray (*Halichoerus grypus*) seals (Duignan et al., 1995). Additionally, differences have been reported in domestic animals. For example, it has been reported (Howell, 1963) and verified by several experimental infections (Bida and Eid, 1974; Neitz, 1975; Tomori, 1980) that breeds of African and Asiatic domestic sheep are more resistant to infection by bluetongue virus than are European breeds. Differences in innate resistance to disease also extend to the subspecies level in wild animals as demonstrated here with EHD and white-tailed deer.

Differences in innate genetic resistance to HD are one aspect of the enzootic stability hypothesized to occur for EHD and BT viruses in native Texas deer. Should differences in innate genetic resistance to HD extend to other white-tailed deer subspecies, these differences also may help explain why severe clinical disease and mortality characterize HD epizootics, specifically EHD, in northern latitudes whereas infections in southern latitudes often result in mild or inapparent disease.

Differences in innate resistance to disease that vary on the subspecies level have major implications in managing free-ranging wildlife populations. Captive propagation, a management tool often used for endangered wildlife, is designed to retain reproductive fitness and maintain a genetically diverse population for eventual reintroduction into the wild (Montgomery et al., 1997). Because often it is unknown to which diseases animals are innately resistant, it is a difficult proposition for captive breeding programs to account for preserving innate resistance to endemic diseases. Animals that are removed from native areas for captive propagation also may be removed from frequent and intense challenge by pathogens, a mechanism that may be responsible for maintenance of population-wide innate resistance to those pathogens. Subsequent propagation of multiple generations in captivity could result in a loss of innate resistance and a population that may never be suitable for reintroduction.

Wildlife translocation, a common wildlife management tool (Griffith et al., 1989), may be undertaken to augment a dwindling population or to re-establish an extirpated population (Gaydos and Corn, 2001). Failing to consider innate resistance to disease when using this tool can result in wildlife translocation failures. When considering translocation, one concern is the potential for exposure of translocated wildlife to pathogens already present at the release area (Gaydos and Corn, 2001). Although it is not known if it was due to variations in susceptibility at the species or subspecies level, examples exist where translocated animals became infected by pathogens at the release site and died, resulting in translocation failure (Gaydos and Corn, 2001). We propose that when translocation is used to augment a dwindling population, differences in innate resistance to virulent pathogens between subspecies or differences between the same species originating from different geographic locations can pose a threat to translocation

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success. The movement of non-resistant stocks to augment a remnant resistant population can cause the eventual genetic dilution of the beneficial disease-resistant trait held by the remnant population. Currently this is largely a theoretical consideration because so little is known about differences in disease resistance between wildlife subspecies or even between wildlife of the same species originating from different geographic locations.

ACKNOWLEDGMENTS

We thank D. Frels, G. Fuchs, the Texas Parks and Wildlife Department, J. Huffman, K. Dubin, R. Boyd, and the Pennsylvania Game Commission for assistance in obtaining deer. We also thank V. F. Nettles and J. R. Fischer for their scientific input into the development of this project and SCWDS staff for support. This project was supported through sponsorship from fish and wildlife agencies in Alabama, Arkansas, Florida, Georgia, Kentucky, Louisiana, Maryland, Mississippi, Missouri, North Carolina, Puerto Rico, South Carolina, Tennessee, Virginia, and West Virginia. Funds were provided by the Federal Aid to Wildlife Restoration Act (50-Stat. 917) and through Grant Agreement 14-45-0009-94-906, National Biological Service, U.S. Department of the Interior.

LITERATURE CITED

ALLISON, A. C. 1982. Co-evolution between hosts and infectious disease agents and its effects on virulence. *In* Population biology of infectious diseases, R. M. Anderson and R. M. May (eds.). Springer, Berlin, Germany, pp. 245–268.

ATKINSON, C. T., R. J. DUSEK, K. L. WOODS, AND W. M. IKO. 2000. Pathogenicity of avian malaria in experimentally-infected Hawaii amakihi. Journal of Wildlife Diseases 36: 197–204.

BAKER, R. H. 1984. Classification and distribution. *In* White-tailed deer ecology and management, L. K. Halls (ed.). Stackpole Books, Harrisburg, Pennsylvania, pp. 1–18.

BEST, S. M., S. V. COLLINS, AND P. J. KERR. 2000. Coevolution of host and virus: Cellular localization of virus in myxoma virus infection of resistant and susceptible European rabbits. Virology 277: 76–91.

BIDA, S. A., AND F. I. A. EID. 1974. Blue Tongue of sheep in Northern Nigeria. Journal of the Nigerian Veterinary Medical Association 3: 12–16.

DAVIDSON, W. R., AND G. L. DOSTER. 1997. Health characteristics and white-tailed deer population density in the southeastern United States. *In* The science of overabundance: Deer ecology and population management, W. J. McShea, H. B. Underwood and J. H. Rappole (eds.). Smithsonian Institution Press, Washington D.C., pp. 164–184.

DEMAULA, C. D., M. A. JUTILA, D. W. WILSON, AND N. J. MACLACHLAN. 2001. Infection kinetics, prostacyclin release and cytokine-mediated modulation of the mechanism of cell death during bluetongue virus infection of cultured ovine and bovine pulmonary artery and lung microvascular endothelial cells. Journal of General Virology 82: 787–794.

DUIGNAN, P. J., J. T. SALIKI, D. J. ST. AUBIN, G. EARLY, S. SANDOVE, J. A. HOUSE, K. KOVACS, AND J. R. GERACI. 1995. Epizootiology of morbillivirus infection in North American harbor seals (*Phoca vitulina*) and gray seals (*Halichoerus grypus*). Journal of Wildlife Diseases 31: 491–501.

EWALD, P. W. 1995. The evolution of virulence: A unifying link between parasitology and ecology. Journal of Parasitology 81: 659–669.

GAYDOS, J. K., AND J. L. CORN. 2001. Health aspects of large mammal restoration. *In* Large mammal restoration: Ecological and sociological challenges in the 21st century, D. S. Maehr, R. F. Noss and J. L. Larkin (eds.). Island Press, Covela, California, pp. 149–162.

GRIFFITH, B., J. M. SCOTT, W. CARPENTER, AND C. REED. 1989. Translocation as a species conservation tool: Status and strategy. Science 245: 477–480.

HOWELL, P. G. 1963. Bluetongue. *In* Emerging diseases of animals. Food and Agriculture Organization Agricultural Studies 61: 109–154.

HOWERTH, E. W., C. E. GREENE, AND A. K. PRESTWOOD. 1988. Experimentally induced bluetongue virus infection in white-tailed deer: Coagulation, clinical pathologic, and gross pathologic changes. American Journal of Veterinary Research 49: 1906–1913.

LIPSITCH, M., E. A. HERRE, AND M. A. NOWAK. 1995. Host population structure and the evolution of virulence: A "law of diminishing returns." Evolution 49: 743–748.

MAY, R. M. 1985. Ecological aspects of disease and human populations. American Zoologist 25: 441–450.

MONTGOMERY, M. E., J. D. BALLOU, R. K. NURTHEN, P. R. ENGLAND, D. A. BRISCOE, AND R. FRANKHAM. 1997. Minimizing kinship in captive breeding programs. Zoo Biology 16: 377–389.

NEITZ, W. O. 1975. Studies on blue tongue in sheep. Onderstepoort Journal of Veterinary Science and Animal Industry 23: 93–98.

NETTLES, V. F., AND D. E. STALLKNECHT. 1992. History and progress in the study of hemorrhagic disease of deer. Transactions of the North American Wildlife and Natural Resources Conference 57: 499–516.

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———, S. A. HYLTON, D. E. STALLKECHT, AND W. R. DAVIDSON. 1992. Epidemiology of epizootic hemorrhagic disease virus in wildlife in the USA. *In* Bluetongue, African horse sickness and related orbiviruses, Proceedings of the second international symposium, T.E. Walton and B.I. Osburn (eds.). CRC Press, Boca Raton, Florida, pp. 238–248.

PARER, I., W. R. SOBEY, D. CONOLLY, AND R. MORTON. 1994. Virulence of strains of myxoma virus and the resistance of wild rabbits, *Oryctolagus cuniculus* (L.), from different locations in Australia. Australian Journal of Zoology 42: 347–362.

QUIST, C. F., E. W. HOWERTH, D. E. STALLKNECHT, J. BROWN, T. PISELL, AND V. F. NETTLES. 1997. Host defense responses associated with experimental hemorrhagic disease in white-tailed deer. Journal of Wildlife Diseases 33: 584–599.

SMITH, K. E., D. E. STALLKNECHT, AND V. F. NETTLES. 1996. Experimental infection of *Culicoides lahillei* (Diptera: Ceratopogonidae) with epizootic hemorrhagic disease virus serotype 2 (Orbivirus: Reoviridae). Journal of Medical Entomology 33: 117–122.

STALLKNECHT, D. E., V. F. NETTLES, E. A. ROLLOR, AND E. W. HOWERTH. 1995. Epizootic hemorrhagic disease virus and bluetongue virus serotype distribution in white-tailed deer in Georgia. Journal of Wildlife Diseases 31: 331–338.

———, M. P. LUTTRELL, K. E. SMITH, AND V. F. NETTLES. 1996. Hemorrhagic disease in white-tailed deer in Texas: A case for enzootic stability. Journal of Wildlife Diseases 32: 695–700.

TOMORI, O. 1980. Bluetongue and related viruses in Nigeria: Experimental infection of West African dwarf sheep with Nigeria strains of the viruses of epizootic haemorrhagic disease of deer and bluetongue. Veterinary Microbiology 5: 177–185.

TSAI, K., AND L. KARSTAD. 1973. The pathogenesis of epizootic hemorrhagic disease of deer: An electron microscopic study. American Journal of Pathology 70: 379–400.

Received for publication 26 November 2001.

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