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Prevalence of EEEV Antibodies among White-Tailed Deer Populations in Maine

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Abstract

During fall 2010, 332 deer serum samples were collected from 15 of the 16 (93.8%) Maine counties and screened for eastern equine encephalitis virus (EEEV) antibodies by using Plaque Reduction Neutralizing Tests (PRNTs). The aim was to detect and map EEEV activity in the state of Maine. Forty-seven of the 332 (14.2%) sera were positive for EEEV antibodies showing a much wider distribution of EEEV activity in Maine than previously known. The percentage of EEEV antibody positive deer sera was ≥10% in six counties Piscataquis (100%), Somerset (28.6%), Waldo (22.2%), Penobscot (21.7%), Kennebec (13.7%) and Sagadahoc (10%). Positive sera were detected in all the 6 counties (Somerset, Waldo, Penobscot, Kennebec, Cumberland and York) that were positive in 2009 suggesting endemic EEEV activity in these counties. EEEV antibodies were not detected in sera collected in five counties: Franklin, Knox, Lincoln, Oxford and Washington which was either due to low sample size or lack of EEEV activity in these counties. Our data suggest higher EEEV activity in central Maine compared to southern Maine whereas EEEV activity in Maine has historically been associated with the southern counties of York and Cumberland.

Keywords

Odocoileus virginianus ; Eastern Equine Encephalitis Virus; White-Tailed Deer; Plaque Reduction Neutralization Test; Maine

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Introduction

The distribution of eastern equine encephalitis virus (EEEV) activity in Maine is currently not well understood. Before 2005 EEEV had only been detected in an American goldfinch (*Carduelis tristis*) in York County (Lubelcyck et al. 2013). Between 2005 and 2007 EEEV was detected in birds, horses, and mosquito pools in York and Cumberland counties in southern Maine and at that time it was thought that southern Maine represented the northern limits of EEEV activity in the North America (Lubelcyck et al. 2014). However, in 2009 an epizootic of EEE occurred in 5 counties in southern (York and Cumberland Counties) and central Maine (Kennebec, Waldo and Penobscot Counties) and it involved fatalities in at least 15 horses, a llama and 3 flocks of pheasants (Gibney et al. 2011, Lubelcyck et al. 2013). This suggested a much broader distribution of EEEV activity in the state. Following the outbreak in 2009, we conducted a pilot study and found high EEEV antibodies levels in free-ranging deer (*Odocoileus virginianus*) sera in central Maine which suggested that EEEV is endemic in this region (Mutebi et al. 2011). Since then deer serosurveys have been utilized to study the distribution of EEEV activity and other arboviruses in other states in Northeastern United States (Berl et al. 2013, Nofchissey et al. 2013).

In 2010, we expanded the deer serosurvey studies to include most of the counties in Maine to get a more complete picture of the distribution of EEEV activity in the state. In the course of these studies we noticed that the number of deer tagged was significantly reduced in northern Maine corresponding to lower densities in that region of the state (Lavigne 1997). We also noticed that the numbers of moose (*Alces alces*) tagged increased in northern Maine suggesting increased moose population size in that part of the state (Wattles and DeStefano 2011, Lubelcyck et al. 2014). We screened the moose sera and detected EEEV antibodies in approximately 11% of the samples which showed that moose were exposed to EEEV infections in northern Maine (Lubelczyk et al. 2014). Previously we had observed that 29% of the moose sera collected in northern Vermont were positive for EEEV antibodies (Mutebi et al. 2012). Taken together these observations suggest that similar to deer, moose can be used as sentinels to detect EEEV activity. In the present study, EEEV antibody positive moose sera were detected near Fort Kent very close to the Canadian border suggesting EEEV activity throughout the state of Maine (Lubelczyk et al. 2014). However there are large areas in the state of Maine where EEEV activity is unknown. Since the distribution of EEEV activity in North America is patchy and not uniform throughout the distribution range (Morris 1988) it is essential to investigate all areas of the state to obtain an accurate picture of EEEV activity. In this manuscript we present and discuss our observations of the expanded deer serosurvey in Maine in 2010.

Materials and Methods

Serum collection.

Blood samples were collected from deer carcasses by using the methods previously described by Mutebi et al. (2011). Briefly whole blood was collected either from the heart or from blood pools in body cavities of the disemboweled carcasses by using sterile syringes or pipettes and placed into 5 or 10ml vacutainer tubes (Fischer Scientific, Pittsburg, PA). In the field, vacutainer tubes were kept on ice in Styrofoam chests and transported on ice to the lab

at the end of each day. In the lab, the vacutainer tubes were centrifuged at 3000 rpm for 5 – 10 min to separate serum from the blood clot and stored frozen at −20°C. Deer tag numbers were used as ID numbers and the approximate age of deer was estimated and reported as juvenile or adult using teeth wear (Cain 2010). The approximate locations where the deer were killed were pointed out and marked on high resolution area maps by the hunters. At all times during the blood collection process, standard universal precautions against potential blood borne pathogens were taken.

Blood samples were collected at 42 deer tagging stations: Greene, Livermore, Minot and Sabattus in Androscoggin County; Ashland, Fort Kent, Houlton, Island Falls, Linneus, Monticello, New Limerick, Portage Lake and Presque Isle in Aroostook County; Gray, Sebago, and Standish in Cumberland County; Centerville in Franklin County; Gouldsboro in Hancock County; Benton, West Gardiner, Windsor and Winthrop in Kennebec County; Thomaston in Knox County; Waldoboro in Lincoln County; Dexter, Eddington, Millinocket, Newport in Penobscot County; Greenville in Piscataquis; Bowdoin in Sagadahoc County; Skowhegan and Jackman in Somerset County; Freedom and Morrill in Waldo County; Jonesport, Machias, and Whiting in Washington County; Acton, South Berwick and Wells in York County. These stations were selected to include as much of the state as possible. In addition large numbers of deer had consistently been registered at most of these stations in the previous years and their inclusion increased the possibility of obtaining representative samples. We started sampling on 30 October 2010 (the beginning of the firearm hunting season) and continued through January 2011.

Serologic tests.

Deer serum samples were diluted 1:10 and screened for EEEV-neutralizing antibodies by plaque-reduction neutralization assay (Beaty et al. 1995). Positive specimens and all specimens neutralizing over 70% were titrated in duplicate for confirmation. Serum samples were considered positive for EEEV antibodies if they neutralized 80% of a challenge dose of ≈100 plaque-forming units of EEE-Sindbis chimeric virus (Wang et al. 2007).

Results and Discussion

Three hundred and thirty-two (332) deer serum samples were collected from 15 of the 16 (93.8%) ME counties and 47 (14.2%) were positive for EEEV antibodies by PRNT (Tables 1 and 2). This shows a much wider distribution of EEEV activity in Maine than previously reported by Mutebi et al. (2011) and Lubelczyk et al. (2014). The percentage of EEEV antibody positive deer sera was ≥10% in six counties Piscataquis (100%), Somerset (28.6%), Waldo (22.2%), Penobscot (21.7%), Kennebec (13.7%) and Sagadahoc (10%) (Table 1). Although the highest percentage of EEEV positive sera (100%) was detected in Piscataquis County, only 2 deer samples were collected in that County and therefore the high percentage may be attributed to the extremely low sample size. In counties where more than 20 samples were collected the highest percentage of EEEV antibody sera was detected in Somerset County (28.6%) followed by Waldo (22.2%), Penobscot (21.7%), Kennebec (13.7%) and Cumberland (7.3%). Positive sera were detected in all the 6 counties (Somerset, Waldo, Penobscot, Kennebec, Cumberland and York) that were positive in 2009 (Mutebi et al. 2011)

which suggest endemic EEEV activity in these counties. The high percentage of positive sera detected in Somerset County in both 2009 (19%) and 2010 (28.6%) suggest consistently high EEEV activity in this County. Similarly the percentages of positive deer sera detected in Cumberland, York and Penobscot Counties (9.4%, 5% and 12.5% respectively) were very similar to those observed in 2009 (7.3%, 2.9% and 21.7% respectively) (Mutebi et al. 2011) suggesting consistent EEEV activity in this county as well as well. However, in Kennebec and Waldo counties, the percentage of EEEV positive sera detected was substantially lower in 2010 (3.8% and 7.9% respectively) when compared to what was detected in 2009 (13.7% and 22.2% respectively) (Mutebi et al. 2011) which suggests that EEEV activity may be a recent expansion into these counties. As epizootic activity of the virus was also higher in 2009 than 2010 in this region of Maine (Gibney 2011), the parallels seen in cervids may also speak to the usefulness of this surveillance method.

Although positive sera has been detected in York and Cumberland in both 2009 and 2010 and EEEV activity has historically been associated with these two southern counties, the percentages of positive sera detected in these two counties were substantially lower than those detected in some counties in central Maine such as Somerset, Waldo and Penobscot (Table 1) (Mutebi et al. 2011). This suggests that the highest EEEV activity in the state may not be in southern Maine, despite historical viral activity in veterinary cases (Lubelczyk 2013). Most of the positive sera were detected at the junction where five counties, Somerset, Piscataquis, Penobscot Waldo and Kennebec come together in central Maine suggesting that this may be a focus of EEEV activity (Fig. 1).

EEEV antibodies were not detected in sera collected in five counties: Franklin, Knox, Lincoln, Oxford and Washington (Table 1, Fig. 1). However, with the exception of Washington County where 20 serum samples were collected, the sample sizes from the other four counties were limited to one or 2 samples (Table 1) and the inability to detect positive samples in these counties may be attributed to the small sample sizes. Although there was a sizable number of samples from Washington County (20) (Table 1), all these samples were collected in the coastal areas in the south and none from the central or northern areas of this county (Fig. 1). The absence of positive deer sera in southern Washington County is intriguing especially because the coastal areas of this county have numerous ideal habitats for *Cs. melanura* and other EEEV vectors.

Only 17 (5.1%) deer sera were collected from the northern part of the state; 15 (4.5%) from Aroostook County, and 2 of the 23 samples from Penobscot County (Table 1, Fig. 1). Our observations based on deer tagging data suggest that deer populations are not uniformly distributed throughout Maine but rather more restricted to the southern, costal and central parts of the state (Rand et al. 2003). These observations are consistent with the conclusions by Krom (http://www.umaine.edu/cfru/Events/Munsungan_DWA_12.07/Krohn_HistoricalEcology.pdf). Krom compiled available information on the distribution of white-tailed deer in Maine, from the time of European settlement to the early 2000s and found significant distribution variations of deer populations over time; currently deer populations are abundant in the south, central and coastal areas of Maine and very low in the North.

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References

Beaty BJ, Calisher CH and Shope RE. 1995. Arboviruses. In Diagnostic Procedures for Viral, Rickettsial and Chlamydial Infections, 7th Edition. Lennette EH, Lennette DA and Lennette ET (eds.). American Public Health Association, Washington, DC, pp. 169–188.

Berl E, Eisen RJ, MacMillan K, Swope BN, Saxton-Shaw KD, Graham AC, Turmel JP, Mutebi JP. 2013. Serological Evidence for Eastern Equine Encephalitis Virus Activity in White-Tailed Deer, Odocoileus virginianus, in Vermont, 2010. American Journal of Tropical Medicine and Hygiene, 88(1): 103–107.

Cain A A guide to age determination of White-Tailed deer. Texas Parks and Wildlife, Wildlife Region IV. ww.tpwd.state.tx.us/publications/pwdpubs/media/pwd_bk_w7000_0755.pdf, 2010.

Gibney KB, Pelletier AR, Robinson S, Mutebi JP, Nett RJ, Staples JE, Fischer M. 2011. Eastern Equine Encephalitis: An Emerging Arboviral Disease Threat — Maine, 2009. Vector Borne and Zoonotic Diseases, 11(6):637–639. [PubMed: 21254938]

Krohn WB History of the White-Tailed Deer in Maine. http://www.umaine.edu/cfru/Events/Munsungan_DWA_12.07/Krohn_HistoricalEcology.pdf, 2007.

Lavigne GL. 1997. White-tailed deer assessment and strategic plan. Maine Department of Inland Fisheries and Wildlife. August, ME. 111p.

Lubelczyk C, Mutebi JP, Robinson S, Elias SP, Smith LB, Juris S, Foss K, Lichtenwalner A, Shively KE, Hoenig D, Sears S, Smith RP Jr. 2013. An outbreak of eastern equine encephalitis virus, Maine, U.S.A. in 2009: Outbreak description and entomological studies. Journal of the American of Tropical Medicine and Hygiene, 88(1): 95–102.

Lubelczyk C, Elias SP, Kantar L, Albert J, Hansen S, Godsey MS, Saxton-Shaw K, MacMillan K, Smith LB, Eisen R, Smith RP, Mutebi JP. 2014. Detection of eastern equine encephalitis antibodies in moose (Alces americanus), Maine, U.S.A. Vector-Borne and Zoonotic Diseases, 14(1):77–81. [PubMed: 24359417]

Morris CD 1988. Eastern equine encephalomyelitis. In The Arboviruses: Epidemiology and Ecology, Monath TP (ed.). CRC Press Boca Raton, Florida, pp. 1–20.

Mutebi JP, Lubelczyk C, Eisen R, Panella N, Macmillan K, Godsey M, Swope B, Young G, Smith RP, Kantar L, Robinson S and Sears S. 2011. Using Wild White-Tailed Deer to Track Eastern Equine Encephalitis virus in Maine. Vector Borne and Zoonotic Diseases 11: 1403–1409. [PubMed: 21736489]

Mutebi JP, Swope BN, Saxton-Shaw KD, Graham AC, Turmel JP, Berl E. 2012. Eastern equine encephalitis in moose (Alces americanus) in northeastern Vermont. Journal of Wildlife Diseases, 48(4): 1109–1112. [PubMed: 23060520]

Nofchissey RA, Deardorff ER, Blevins TM, Anishchenko M, Brault AC, D. Ebel GD, Lubelczyk C, Berl E, Mutebi JP, Magnarelli. 2013. Seroprevalence of Powassan virus in New England Deer, 1979–2010. American Journal of Tropical Medicine and Hygiene, 88(6): 1159–1162.

Rand PW, Lubelczyk C, Lavigne GR, Elias SP, Holman MS, Lacombe EH, and Smith RP Jr. 2003 Deer density and the abundance of Ixodes scapularis (Acari: Ixodidae). J. Med. Entomol 40 (2): 179–184. [PubMed: 12693846]

Wang E, Petrakova O, Adams AP, Aguilar P, Kang W, Paessler S, Volk SM, Frolov I and Weaver SC. 2007. Chimeric sindbis/eastern equine encephalitis vaccine candidates are highly attenuated and immunogenic in mice. Vaccine 25: 7573–7581. [PubMed: 17904699]

Wattles DW, DeStefano S Status and management of moose in the northeastern United States. Alces 2011; 47: 53–68.

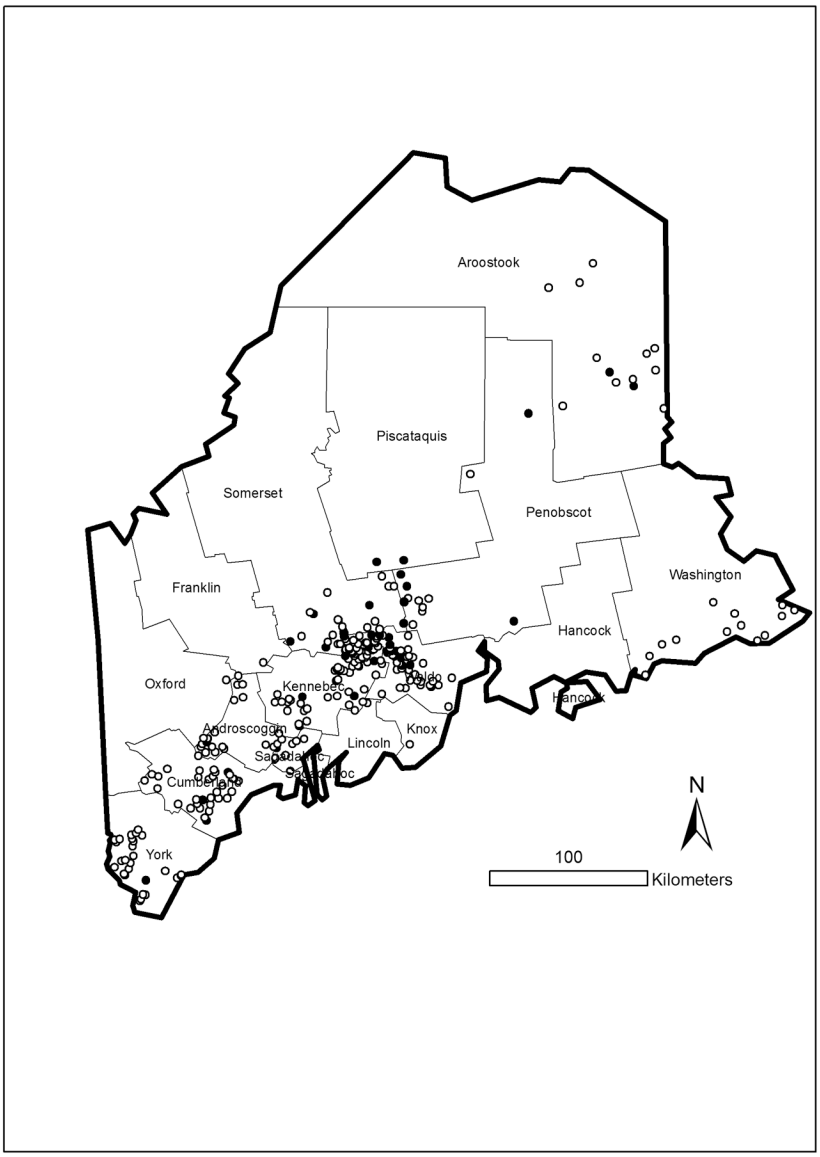

Fig. 1.
Location of sites where deer were harvested and serum samples collected in Maine, 2010.

Table 1.

Number of deer serum samples collected, number and percentage of EEEV antibody positive sera from different Counties in Maine, 2010.

County	Serum Samples Tested	EEEV Antibody Positive Samples	Percentage of EEEV Antibody Positive Samples
Androscoggin	19	1	5.3
Aroostook	15	2	13.3
Cumberland	41	3	7.3
Franklin	2	0	0.0
Kennebec	73	10	13.7
Knox	1	0	0.0
Lincoln	1	0	0.0
Oxford	1	0	0.0
Penobscot	23	5	21.7
Piscataquis	2	2	100.0
Sagadahoc	10	1	10.0
Somerset	35	10	28.6
Waldo	54	12	22.2
Washington	20	0	0.0
York	35	1	2.9
Totals	**332**	**47**	**13.6**

Table 2.

$PRNT_{80}$ and $PRNT_{90}$ EEEV antibody results for the 47 positive white-tailed deer (*O. virginianus*) in Maine, 2010.

County	Township	Serum $PRNT_{80}$	Serum $PRNT_{90}$
Androscoggin	Mechanic Falls	640	640
Aroostook	New Limerick	160	20
Aroostook	Ludlow	640	320
Cumberland	North Yarmouth	80	20
Cumberland	Windham	-	1280
Cumberland	Gorham	20	10
Kennebec	Clinton	20	-
Kennebec	Albion	≥640	320
Kennebec	Winslow	1280	640
Kennebec	Clinton	160	-
Kennebec	Benton	-	≥640
Kennebec	Vassalboro	40	10
Kennebec	Clinton	640	320
Kennebec	Benton	-	1280
Kennebec	Manchester	640	320
Kennebec	China	10	-
Penobscot	Dexter	-	320
Penobscot	Corinna	160	320
Penobscot	TAR7	320	160
Penobscot	Holden	-	80
Penobscot	Newport	80	40
Piscataquis	Parkman	10	-
Piscataquis	Sangerville	80	-
Sagadahoc	Bowdoin	320	80
Somerset	Hartland	-	640
Somerset	Mercer	80	40
Somerset	Pittsfield	640	640
Somerset	Skowhegan	2560	1280
Somerset	Fairfield	-	640
Somerset	Fairfield	-	160
Somerset	Pittsfield	-	640
Somerset	Fairfield	-	640
Somerset	Detroit	-	640
Somerset	Madison	320	160
Waldo	Burnham	-	320

County	Township	Serum $PRNT_{80}$	Serum $PRNT_{90}$
Waldo	Burnham	-	640
Waldo	Unity	80	20
Waldo	Unity	-	10
Waldo	Unity	20	10
Waldo	Unity	-	640
Waldo	Freedom	640	640
Waldo	Unity	1280	640
Waldo	Unity	5120	2560
Waldo	Knox	-	1280
Waldo	Unity	320	160
Waldo	Monroe	-	10
York	North Berwick	160	80