

# West Nile Virus Titers in a Semi-Feral Herd of Ponies in Pennsylvania

Sue M. McDonnell,<sup>1</sup> Elkanah H. Grogan,<sup>1</sup> Amy L. Glaser,<sup>3</sup> and Pamela A. Wilkins<sup>2</sup>

<sup>1</sup>Equine Behavior Laboratory, Section of Reproduction, Department of Clinical Studies - New Bolton Center

<sup>2</sup>Department of Clinical Studies - New Bolton Center  
School of Veterinary Medicine, University of Pennsylvania

<sup>3</sup>Animal Health Diagnostic Laboratory, Cornell University, Ithaca, NY

West Nile Virus (WNV) first appeared in horses in Pennsylvania in 2000. Natural exposure and antibody titer responses to WNV within a stable population of equids over time have not been previously reported. A herd of Shetland-type ponies (n=50-65), maintained continuously at pasture in Chester County Pennsylvania since 1994, with no known WNV disease and not vaccinated against WNV, was studied. Serum samples were obtained in Autumn of 2003 after mosquito season for this geographic region (A03), Spring of 2004 before mosquito season (S04) and Autumn of 2004 after mosquito season (A04). Microtiter serum neutralization testing (MSNT) was performed by the Veterinary Diagnostic Laboratory at Cornell University in S04 and A04. MSNT titers were considered positive at 1:16 or greater. ELISA was performed by the Pennsylvania Animal Diagnostic Laboratory System (PADLS) in A03, S04 and A04. ELISA titers were reported as either positive or negative at 1:400 only.

In A03, 23/61 (38%) ponies were ELISA positive, including 2/12 (17%) foals. In S04, 19/47 (40%) ponies were MSNT positive and 8/34 (24%) ELISA positive. Of yearlings, 3/12 (25%) were MSNT positive, including the 2 ELISA positive A03 foals. Of ponies  $\geq 2$  years old (mature), 16/33 (48%) were MSNT positive, including 14 ELISA positive in A03. Of the 11 mature A03 ELISA seropositive ponies available for retesting in S04, 7 (64%) remained ELISA positive. In A04, 40 of 63 (63%) ponies were MSNT positive while 9 of 63 (14%) were ELISA positive. A04 MSNT positives included 5/14 (36%) foals, 6/13 (46%) yearlings and 29/36 (81%) mature ponies, while ELISA positives included 0/14 foals, 1/13 (8%) yearlings and 8/36 (22%) mature ponies. For 30 tested on all occasions and both assays, results for 7 (23%) were consistently positive and for 8 (27%) consistently negative. There were no instances of an MSNT seropositive pony becoming MSNT seronegative (6-month period); there were 11 instances of ELISA seropositive ponies that became ELISA seronegative (1-year period). For the A04 sample set, 31 ponies had discrepant MSNT and ELISA results, all with MSNT positive and ELISA negative.

In Pennsylvania, 2003 was the peak year so far for equine clinical cases of WNV, with > 500 equine cases reported. For 2004, Pennsylvania had only 9 reported cases with none in Chester County. For this herd, MSNT positives increased from 40% before to 63% after the 2004 mosquito season. Of 43 yearlings and mature ponies tested both in S04 and A04, 12 of 20 (60%) that were MSNT negative in S04 were MSNT positive in A04. These data demonstrate persistence of MSNT positivity, and increased herd seropositivity due to natural exposure, over a 6-month period during mosquito season in a WNV endemic area. Animals with positive ELISA titers may become seronegative during periods of no mosquito activity and remain seronegative during the next mosquito season, possibly related to the large titer cut-off.

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