

## West Nile Virus Laboratory Issues

by Jodi Garrett, MT (ASCP)SM, Amy Armbrust, MT (ASCP),  
and Steven Hinrichs, MD

West Nile Virus has hit the state of Nebraska with a vengeance and a number of public health efforts are in effect. The NPHL offers the IgM antibody screening test (**Figure 1**) that utilizes West Nile Virus (WNV) reagents provided by Focus Technologies. The role of the NPHL is to assist in determining the severity and extent of disease throughout the state. The NPHL is working with the state program that provides WNV testing at no charge. Once the full extent of WNV disease is determined, the “no charge” program may end. A termination date has not yet been determined.

A number of questions have arisen about the testing procedures and the interpretation of the index value that is reported. To assist you in responding to telephone calls from medical staff, the following explanations have been provided:

*What's the difference between a laboratory presumptive positive and a CDC confirmed result?*

All IgM screening tests, including the Focus Laboratories test used at NPHL incorporate a WNV antigen that in some cases will also detect antibodies to other flaviviruses including St. Louis Encephalitis Virus (SLE). Therefore, to rule out these other possibilities, a number of confirmatory procedures may be used, including determination of specific antibodies to SLE. A monitoring program is in place for SLE in the state and at the present time there is no activity, therefore, SLE infection in humans is unlikely. The State Epidemiologist, Dr. Tom Safranek may request some cases to be referred to the CDC when SLE is a consideration. Currently, positive results from the NPHL should be considered presumptive.

*What does an indeterminate result mean?* In some situations a cross reaction can be due to heterophile antibodies, in which case the result is reported as indeterminate. Heterophile antibodies are non-specific antibodies that react with antigens in the test kit. If the clinical condition warrants, it may be appropriate to obtain an additional sample for retesting.

*How long does it take to get results?* Although the laboratory initially intended to perform the test only twice a week, the volume has grown so rapidly that it is now performed daily, Monday through Friday. Results are typically available 48 to 72 hours after specimens are received. The NPHL reports results to the state epidemiologist on specimens submitted through the state program.

*How many positive human cases of WNV have been identified?* The laboratory has reported over 300 positive cases as of August 22 and approximately 20 to 30 new positives are reported each day. Therefore, the total number of positives is rapidly changing and this frequency may increase until Nebraska has its first hard frost this fall.

*Why does NPHL only test for the IgM antibody and not IgG.* Dr. Safranek does not recommend screening for West Nile IgG antibody at the present time. All requests for IgG will be forwarded to a reference laboratory for testing and will be billed to the submitter.

*Does the NPHL provide other surveillance testing for WNV in Nebraska?* To date, the NPHL laboratory has tested 1890 pools of mosquitoes. Each mosquito pool consists of 50 mosquitoes (Genus *Culex*). 683 or 36% of the pools have tested positive. Analysis has shown that 18 of the 21 counties tested have positive WNV mosquitoes in them.

*How can I send specimens to the laboratory?* If your laboratory is not serviced by a regular courier route to the NPHL, specimen transport arrangements may be made by calling NPHL Client Services at 800-334-0459 or 402-559-2440.

West Nile Virus Antibody IgM Test Requirements

Method: Capture IgM ELISA  
Availability: Monday through Friday  
STAT Testing is not available  
Specimen: Blood or CSF  
CSF must be paired with a serum specimen  
Collect: 6.0 ml SST/ red/black clot tube  
Volume: Serum: 2.0 ml serum or 0.5 ml minimum  
CSF: 0.5 ml minimum  
Transport: Refrigerated.  
If sample cannot be transported to the laboratory the same day as collected, please centrifuge and separate serum from cells. Arrangements for specimen Transportation can be made by calling NPHL at 800-334-0459 or 402-559-2440. Frozen specimens are acceptable.

CPT: Serum: 86317  
CSF/Serum: 86317 x 2

Reference Serum: Negative: <0.9 Index  
Equivocal:  $\geq 0.9 - \leq 1.1$  Index  
Positive: >1.1 - >15.0 Index

Reference CSF: Negative: <0.9 Index  
Equivocal:  $\geq 0.9 - \leq 1.1$  Index  
Positive: >1.1 - >15.0 Index