New Jersey Public Health and Environmental Laboratories Technical Bulletin

May 13, 2016

Zika Virus Testing for New Jersey Patients-UPDATE

Effective Wednesday, May 18, 2016, The Public Health and Environmental Laboratories (PHEL) will begin in-house testing for Zika virus and two other related viruses - Dengue and Chikungunya - in clinical specimens.

Zika Virus Disease continues to be a priority for public health preparedness and response especially as mosquito season approaches. PHEL has the capability to perform three diagnostic CDC-developed tests for Zika virus: Zika reverse transcription Real-Time PCR (rtRT-PCR) assay, Trioplex real time rtRT-PCR assay, and the Zika MAC ELISA IgM test. The Zika rtRT-PCR test is specific for the detection of Zika virus and the Trioplex assay allows for the detection and differentiation between RNA from Zika, Dengue, and Chikungunya viruses. The Zika MAC ELISA IgM assay is a qualitative test for the presumptive detection of IgM antibodies to Zika virus. All presumptive positive or equivocal IgM results will be confirmed using the Plaque Reduction Neutralization Test (PRNT). PHEL will continue to refer positive/equivocal IgM tests to the Centers for Disease Control and Prevention (CDC) or another State Public Health Laboratory for the PRNT test. PHEL will not report presumptive positive or equivocal results until confirmatory results are received.

Newly available data have shown that Zika RNA persists in urine longer than in serum and at a slightly higher concentration. PHEL continues to require submission of serum and urine collected at the same date for each Zika virus test request. Other specimens requested for testing (*e.g.* cerebrospinal fluid and amniotic fluid) must also be accompanied by serum collected at the same time as the fluids.

Please note that all tests are not applicable to all the specimen types listed above. The tests that are ultimately performed are determined by PHEL. Test selection will be dependent on the type of specimen received, the patient's clinical history and/or CDC Zika virus epidemiological criteria, and any initial results obtained.

There are two notable changes to the collection and submission of specimens:

1. Only one set of urine and serum specimen is now needed; duplicate aliquots of urine and sera are no longer required.
2. Serum and urine specimens collected Monday-Thursday should be refrigerated at 4ºC and shipped on cold packs using overnight next day priority delivery. (Receipt at PHEL within 24hrs of collection.)
3. Collection and shipment of specimen on Fridays should be avoided – call if must ship Fridays.

Additional details on the collection and submission of specimen are provided on page 2 of this bulletin

- **To request Zika testing, contact:**
  Communicable Disease Service at (**609)-826-5964** Monday through Friday from 8:00 AM to 5:00 PM.

- **For laboratory questions relating to Zika Virus, email the PHEL Zika Team at:**
  Zika.phel@doh.nj.gov

  General laboratory information, contact: (609)-530-8516 or the PHEL website at:
  http://NJ.gov/health/phel/index.shtml

  NOTE: Per NJAC 8:57, clinicians and laboratories must report confirmed cases of all arboviral diseases (e.g. Zika, Chikungunya, West Nile, and Dengue) to the local health department (LHD) where the tested person resides. http://localhealth.nj.gov If the LHD cannot be reached at the number in the directory, please call the CDS at 609-826-5964, Monday through Friday from 8:00 AM – 5:00 PM.

NJDOH-PHEL **Laboratory Information for Zika Virus Testing** **(May 13, 2016)**

Test Information	Description	Comments
Test Approval:	**Requests for Zika testing must be preapproved** by the New Jersey Department of Health (NJDOH) Communicable Disease Service (CDS). Contact CDS during regular business hours (Monday–Friday 8 AM-5 PM) at (609)-826-5964. If approved, an NJDOH case number will be assigned.	Specimen will not be processed without an NJDOH case number.
Submission of Test Request:	If testing is approved, CDS will provide the clinician with a laboratory test request form (SRD-1) for the patient to take with them to the hospital laboratory for specimen collection.	
Biosafety:	Follow all OSHA Blood borne Pathogen healthcare requirements for blood collection and sharps disposal. For manipulation of specimens within the hospital laboratory follow Biosafety Level 2 practices and procedures. Perform aliquoting of serum and urine within a Biosafety Cabinet Class II and utilize appropriate PPE. http://www.cdc.gov/biosafety/publications/bmbl5/	Refer to the ***Biosafety in Microbiological and Biomedical Laboratories*** **(BMBL) 5th Edition**
Specimen Requirements:	**SERUM and URINE are required for each test request** **SERUM: 3 ml aliquot of sera in a leak-proof tube** **URINE: 3 ml aliquot of a urine specimen without preservative in a leak-proof tube**	Label urine tube with the word "URINE". Label serum tube with the word "SERUM".
Specimen Collection:	**SERUM AND URINE** **Serum:** Collect sufficient blood in a serum separator tube (tiger top, speckle top, or gold top) to obtain a total of 3 ml of serum. Allow blood to clot for 30 min.at room temp. before centrifugation. Aspirate 3 ml of serum into a leak-proof, screw-capped tube. **Urine:** Collect urine in a clean container, pour off 3ml into a clean plastic, leak proof, screw capped tube. Ensure cap is properly aligned to avoid leakage in transit. **Label all specimens with patient's full name, DOB, date and time of collection, and type of specimen.** Store all specimens at 4ºC until ready for packaging and shipping. **Amniotic, cerebrospinal fluids, cord blood, and placental specimens are acceptable on CDS approval.**	**UNACCEPTABLE:** Blood in anticoagulant or plain red top tubes. Urine in a tube with preservative or submitted in a cup. Other specimens: Contact zika.phel@doh.nj.gov
Test Methods:	Zika Virus RNA by rtRT-PCR; Trioplex Real Time rtRT-PCR; Zika MAC (IgM) ELISA; Positive IgM serum screen samples confirmed by Plaque Reduction Neutralization Test (PRNT). When applicable, follow-up testing on **convalescent specimens performed with CDS approval (Contact: [609]-826-5964).**	CDC approved methods and New York State Wadsworth Center approved methods are performed.
Specimen Storage:	**PHEL encourages Monday through Thursday specimen collection only.** Specimens collected Mon–Thursday: Store at 4º C until shipped, Mon-Thurs (within 24 h) Specimens collected Friday–Sunday: Freeze at -70º to -80º C for dry ice shipment on Mon	If -70º C freezer is lacking & Friday draw is unavoidable, email: zika.phel@doh.nj.gov
Specimen Packaging:	**Mon-Thurs Specimen Collection**: Condition the serum and urine at 4º C for a minimum of one hour. Fill the box with refrigerated cold packs, and follow the International Air Transport Association (IATA) packaging instructions 650 for Biological Substances, Category B, UN 3373. Packaging must be conducted by certified personnel in accordance with IATA regulations. **Fri – Sun Specimen Collection**: Specimens collected Fri–Sun must be frozen at -70º to -80º C and shipped on dry ice on Monday. Follow instructions for shipping specimens on Dry Ice in the Zika Technical Bulletin Supplement on Packaging and Shipping Zika Specimens 3/4/16. http://nj.gov/health/phel/index.shtml	Specimens are kept at 4ºC to ensure the stability of potentially low levels of virus during transit. Please use sufficient cold packs to maintain refrigeration during transit. DO NOT FREEZE AT -20ºC.
Specimen Shipping:	Ship to PHEL **Monday–Thursday ONLY** at: **NJ Public Health and Environmental Laboratories, 3 Schwarzkopf Drive, Ewing, NJ 08628. ATTN: Dr. Nelson Delgado (609)-209-9004**	Emergency contact for shipments in transit is (609)-209-9004
Specimen Transport:	**Specimens are received at PHEL Monday–Friday, 8 AM–5 PM** Special arrangements must be made for Saturday receipt at PHEL. If special arrangements have been made with PHEL for Saturday receipt, be sure to request Overnight ***Delivery for Saturday***. Carriers do not routinely deliver on Saturday.	Saturday delivery requests are discouraged. Contact zika.phel@doh.nj.gov for more information.
Results Reporting:	Results reporting vary depending on the combination of tests that are performed. Report are not finalized until all indicated tests are completed. Links to CDC Fact sheets are provided	
Turnaround Time (TAT):	For RT-PCR, urine and/or serum is 7 days Serum IgM if negative is 7 days Serum IgM, if positive is 20 days Serum PRNT is 20 days	Times stated excludes weekends and holidays
Report Generated:	Via secured email or fax to CDS, ordering physician, submitting hospital laboratory, and if positive, Centers for Disease Control and Prevention (CDC).	
Specimen Rejection Policy:	Specimens may be rejected if all the requirements are not met. All attempts will be made to resolve any issues before rejecting a specimen, *i.e.*, tubes leaking, broken in transit, unlabeled, or mislabeled, & unapproved request, incorrect name, no specimen identified, or a urine cup etc.	Questions regarding specimen acceptability: Zika.phel@doh.nj.gov

For questions relating to any information on this bulletin, please contact PHEL Zika Team at zika.phel@doh.nj.gov

Note: The guidelines and protocols for laboratory handling and processing of Zika specimens are dynamic at this time. This technical bulletin may be updated as necessary. Retain copies of all forms prepared for your records