July 28, 2016

Dear Colleague,

Since January 2016, we have identified 48 cases of lab-confirmed Zika virus infection in travelers returning to Maryland from Zika endemic areas; to date, we have not identified any cases of locally transmitted mosquito-borne Zika virus infections in Maryland. With your help, we are working to prevent local mosquito-borne transmission from occurring here by disseminating educational materials, updating providers on prevention and diagnostic guidelines, and conducting mosquito control measures in response to identified cases or elevated mosquito populations in partnership with other state and local agencies. Please continue to regularly and frequently check http://zika.maryland.gov and http://cdc.gov/zika for updated information.

Today, we are writing with several important updates regarding Zika virus, including: 1) updated algorithms for the evaluation and diagnosis of Zika virus in Maryland residents, 2) updated guidance on the care of pregnant women exposed to Zika virus, 3) updated guidance for the prevention of sexual transmission of Zika virus, and 4) updated recommendations for the collection of infant specimens at the time of birth.

1) Updated Testing Algorithms for Maryland Residents

The Maryland Department of Health and Mental Hygiene (DHMH) has updated its testing algorithms for all individuals, as described in the enclosed flow charts. Consultation with and approval by your local health department is still required for testing in all individuals. Important changes to the evaluation of Maryland residents for Zika virus include the following:

- **All persons** approved for any Zika testing should have both serum AND urine collected and sent to the Maryland DHMH Laboratories Administration with the Serological Testing Form available at http://dhmh.maryland.gov/zikalabs. **All requested fields in the form must be completed or testing of the specimen will be delayed.** As previously communicated, urine samples will not be tested without an accompanying serum specimen.

- PCR testing for all symptomatic individuals on both serum and urine will be extended to **3 weeks** (as opposed to 2 weeks noted in CDC guidance) after symptom onset or last possible Zika virus exposure.

- For all pregnant women, regardless of the presence of symptoms, PCR testing will be performed within the first 3 weeks after their symptom onset or last possible Zika virus exposure.
• For pregnant women who present more than 3 weeks after their symptom onset or last possible Zika virus exposure, reflex PCR testing will be performed when a positive or equivocal anti-Zika or anti-dengue IgM antibody result is obtained. Reflex neutralizing antibody (PRNT) testing will continue to be conducted to rule out false-positive results.
• For individuals presenting more than 12 weeks after symptom onset or last possible Zika virus exposure, testing approval is considered on a case-by-case basis, though we will continue to approve testing in high-risk individuals, including pregnant women.

2) Updated Interim Guidance for Health Care Providers Caring for Pregnant Women with Possible Zika Virus Exposure

On Monday, July 25, 2016, the Centers for Disease Control and Prevention (CDC) released a Morbidity and Mortality Weekly Report (MMWR) with updated guidance on the evaluation and care of pregnant women potentially exposed to Zika virus. It is available at http://www.cdc.gov/mmwr/volumes/65/ww/mm6529e1.htm?s_cid=mm6529e1_w. This guidance has been incorporated into DHMH’s updated Maryland-specific testing algorithms already described above. Additional important highlights from this report include:

• Recognition of the need to ask all pregnant women about possible Zika virus exposures—including travel to Zika endemic areas and sexual contact with persons who have traveled to Zika endemic areas—at every prenatal appointment
• Recommendations to collect both cord blood AND infant serum for infants born to women with possible Zika virus or unspecified flavivirus infection
• Emphasis on testing potentially exposed infants for Zika virus infection regardless of the presence or absence of phenotypic abnormalities
• Clarification that a positive PCR or immunohistochemical staining on the placenta indicates the presence of maternal infection; it does not necessarily mean that the infant was infected
• Description of additional sexual activities that might expose individuals to Zika virus infection

3) Updated Interim Guidance on the Prevention of Sexual Transmission of Zika Virus

On Monday, July 25, 2016, CDC released a MMWR with updated guidance on preventing sexual transmission of Zika virus. It is available at http://www.cdc.gov/mmwr/volumes/65/ww/mm6529e2.htm?s_cid=mm6529e2_w. Important highlights in this report include:

• Recognition that Zika virus can be transmitted from both men and women to their sexual partners
• Expansion of CDC’s existing recommendations to cover all pregnant couples, including pregnant women with female sex partners
• Emphasis that testing for the assessment of risk for sexual transmission is of uncertain value and not currently recommended
4) Updated Recommendations for Testing of Infant Specimens at Time of Birth

CDC has updated its website with more detailed instructions on the collection and submission of specimens from infants born to mothers who have tested positive for possible Zika or unspecified flavivirus exposure.

- Do not ship any specimens directly to CDC; all specimens should be sent to the DHMH Laboratories Administration.
- DHMH will obtain testing pre-approval from CDC for you.
- When submitting multiple tissue specimens, each individual specimen should be clearly labeled as to its source.
- Disregard the CDC guidance on the CDC website to transfer infant cord blood/serum to new containers; continue to submit these specimens in the original vacutainers in which they were collected and DHMH will process as appropriate.

Please continue to contact the DHMH Zika Team at (410) 767-6700 with any questions or concerns. We appreciate your attention to this evolving and important matter.

Sincerely,

[Signature]

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Maryland Department of Health and Mental Hygiene Zika Diagnostic Testing Algorithm

Collect serum AND urine for all patients

- < 3 weeks after symptom onset or exposure
  - rRT-PCR on urine and serum
    - + Confirmed Zika infection
    - +/-equivocal Get convalescent serum specimen and test for IgM
      - +/-equivocal PRNT*
      - + Confirmed Zika infection ruled out
      - - Refer to PRNT results*

- 3-12 weeks after symptom onset or exposure
  - Dengue and Zika IgM
    - - ZIka virus infection ruled out
    - +/-equivocal rRT-PCR on urine and serum
      - +/-equivocal PRNT*
      - - Confirmed Zika infection
      - + Refer to PRNT results*

- >12 weeks after symptom onset or exposure
  - Fetal abnormality?
    - Yes
      - Consider dengue and Zika IgM
    - No
      - Confirmed Zika infection
      - Refer to IgM and PRNT results*

*For interpretation of PRNT results, see: Rabe IB, Staples JE, Villanueva I, et al. Interim Guidance for Interpretation of Zika Virus Antibody Test Results. MMWR Morb Mortal Wkly Rep 2016;65. DOI: http://dx.doi.org/10.15585/mmwr.mm6521e1
Maryland Department of Health and Mental Hygiene Zika Diagnostic Testing Algorithm

Collect serum AND urine for all patients

< 3 weeks after symptom onset or exposure
  - rRT-PCR on urine and serum
    + Confirmed Zika infection
    - Dengue and Zika IgM
      - +/equivocal
        - PRNT*
        - +/equivocal
          - PRNT*
        - Zika virus infection ruled out

3-12 weeks after symptom onset or exposure
  - Dengue and Zika IgM
    - +/equivocal
      - PRNT*
      - Zika virus infection ruled out
    - -

>12 weeks after symptom onset or exposure
  - Dengue and Zika IgM
    - +/equivocal
      - PRNT*
      - Zika virus infection ruled out
    - -
  - Consider dengue and Zika IgM on case-by-case basis

*For interpretation of PRNT results, see: Rabe IB, Staples JE, Villanueva J, et al. Interim Guidance for Interpretation of Zika Virus Antibody Test Results. MMWR Morb Mortal Wkly Rep 2016;65. DOI: http://dx.doi.org/10.15585/mmwr.mm6521e1