



Best Practices for Use of Polymerase Chain Reaction (PCR) for Diagnosing *Haemophilus influenzae* and *Neisseria meningitidis* Disease and Public Health Importance of Identifying Serotype/Serogroup

In the clinical setting the diagnosis of invasive disease caused by *Haemophilus influenzae* (Hi) or *Neisseria meningitidis* (Nm) is based on clinical presentation, as well as a variety of laboratory tests, including culture and polymerase chain reaction (PCR). Culture remains the gold standard laboratory test for identification of Hi and Nm with virtually 100% specificity. However, both pathogens have fastidious growth requirements and culture has poor sensitivity in specimens that are not handled properly and in specimens from persons who have received antibiotics. PCR is a rapid test and has high sensitivity and specificity. A major advantage of PCR is that it allows for detection of Hi and Nm from clinical samples in which the organism cannot be detected by culture methods, such as when a patient has been treated with antibiotics before a clinical specimen is obtained for culture. Even when the organisms are nonviable following antimicrobial treatment, PCR can still detect Hi and Nm DNA if a sufficient amount is present in the specimens [1]. The 2015 Council for State Territorial Epidemiologists (CSTE) case definitions for both Hi and Nm classify patients PCR-positive for Hi or Nm as confirmed.

Determining serotype and serogroup is crucial for identifying potential outbreaks and determining appropriate public health responses, such as chemoprophylaxis for contacts of cases of *Haemophilus influenzae* type b (Hib) disease or meningococcal conjugate (MenACWY) versus serogroup B meningococcal (MenB) vaccine administration in outbreaks of Nm [2-4]. Validated, specific real-time PCR assays capable of detecting all six serotypes (a-f) of Hi and 6 serogroups (A, B, C, W, X, and Y) of Nm are available at CDC; these assays can be used directly on cerebrospinal fluid (CSF) and bacterial isolates with high sensitivity and specificity, as well as on DNA extracted from blood and other clinical specimens.

State public health laboratories with Hi and Nm PCR capacity are strongly encouraged to continue performing culture and/or to save clinical specimens for further testing. Hi and Nm culture isolates are valuable not only for serotyping/serogrouping but also for monitoring antimicrobial susceptibility and for conducting whole genome sequencing, which is necessary for strain comparisons during outbreak investigations and to monitor vaccine effectiveness over time [4].

State public health laboratories considering PCR for Hi and Nm should select assays capable of detecting and differentiating all Hi serotypes (serotypes a-f) and all Nm serogroups common in the United States (serogroups B, C, W, and Y). If a public health laboratory is not able to perform serotyping/serogrouping by PCR and a culture isolate is not available, the laboratory should send specimens to the CDC Bacterial Meningitis Laboratory or one of the Association of Public Health Laboratories (APHL) Vaccine Preventable Diseases Reference Laboratories for serotype/serogroup testing (more information available at <http://www.cdc.gov/meningococcal/laboratory.html> and http://www.aphl.org/programs/infectious_disease/Pages/VPD.aspx).

Several commercial multiplex PCR assays capable of simultaneously testing a single specimen for an array of pathogens that cause blood infections or meningitis/encephalitis are now available, primarily for clinical settings (e.g., FilmArray® Blood Culture Identification Panel and FilmArray® Meningitis/Encephalitis [ME]

Panel from BioFire Diagnostics and Meningitis/Encephalitis Panel by PCR from ARUP Laboratories¹) [5,6]. While such assays can rapidly identify Hi and Nm species, most do not determine serotype or serogroup. **Therefore, it is important for laboratories using assays that do not determine serotype/serogroup to perform either a simultaneous culture or a reflex culture if Hi or Nm is identified.** If obtaining a culture is not possible, at a minimum, laboratories should collect and maintain an adequate clinical sample for further testing at a laboratory with a PCR assay that can detect serotype/serogroup.

1. CDC. Laboratory Methods for the Diagnosis of Meningitis. Available at: <https://www.cdc.gov/meningitis/lab-manual/index.html>.
2. CDC. Prevention and control of *Haemophilus influenzae* type b disease: Recommendations of the Advisory Committee on Immunization Practices (ACIP) (<http://www.cdc.gov/mmwr/preview/mmwrhtml/rr6301a1.htm>). *MMWR*. 2013;63(RR01):1–14.
3. Prevention and Control of Meningococcal Disease: Recommendations of the Advisory Committee on Immunization Practices (ACIP) (<http://www.cdc.gov/mmwr/preview/mmwrhtml/rr6202a1.htm>). *MMWR*. 2013;62(RR02):1–22.
4. CDC. Guidance for the Evaluation and Public Health Management of Suspected Outbreaks of Meningococcal Disease. Available at: <https://www.cdc.gov/meningococcal/downloads/meningococcal-outbreak-guidance.pdf>.
5. Biofire FilmArray® Panels. Available at: <http://www.biofiredx.com/products/the-filmarray-panels/>.
6. ARUP Meningitis/Encephalitis Panel by PCR. Available at: <http://ltd.aruplab.com/Tests/Pub/2013305>.

¹ Disclaimer: Use of trade names or commercial sources is for identification only and does not imply endorsement by the Centers for Disease Control and Prevention or the Department of Health and Human Services.