Multiplex PCR for Microbial Detection in Spinal Fluid (BIOFIRE FILMARRAY Meningitis/Encephalitis Panel)

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Disclosures

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FDA-Approved Multiplex PCR Assay - Spinal Fluid
BIOFIRE FILMARRAY Meningitis/Encephalitis Panel

<table>
<thead>
<tr>
<th>Viruses</th>
<th>Bacteria</th>
<th>Fungus</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cytomegalovirus</td>
<td><em>Escherichia coli</em> K1</td>
<td><em>Cryptococcus neoformans/gattii</em></td>
</tr>
<tr>
<td>Enterovirus</td>
<td><em>Haemophilus influenzae</em></td>
<td></td>
</tr>
<tr>
<td>Herpes simplex virus 1</td>
<td><em>Listeria monocytogenes</em></td>
<td></td>
</tr>
<tr>
<td>Herpes simplex virus 2</td>
<td><em>Neisseria meningitidis</em></td>
<td></td>
</tr>
<tr>
<td>Human herpes virus 6</td>
<td><em>Streptococcus agalactiae</em></td>
<td></td>
</tr>
<tr>
<td>Human parechovirus</td>
<td><em>Streptococcus pneumoniae</em></td>
<td></td>
</tr>
<tr>
<td>Varicella zoster virus</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Evaluation of BIOFIRE FILMARRAY Meningitis/Encephalitis Panel

- 291 clinical, residual CSF samples positive by routine method(s) (e.g., bacterial culture, individual real-time PCR)
  - 76 collected during prevaccine era (March 1975 to June 1997); positive for bacterial pathogen
- Overall percent positive agreement
  - 90.1% (145/161) viruses
  - 97.5% (78/80) bacteria
  - 52% (26/50) Cryptococcus neoformans/Cryptococcus gattii

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Comparison of BIOFIRE FILMARRAY Meningitis/Encephalitis Panel to Routine Testing

<table>
<thead>
<tr>
<th>Target</th>
<th>No. of cerebrospinal fluid specimens with:</th>
<th>Expected positive result</th>
<th>Positive result by BIOFIRE FILMARRAY Meningitis/Encephalitis panel</th>
<th>TP result</th>
<th>FP result</th>
<th>FN result</th>
<th>PPA (95% CI)</th>
<th>Adjusted PPA (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Enterovirus</td>
<td>43</td>
<td>41</td>
<td>41</td>
<td>0</td>
<td>2</td>
<td></td>
<td>95.4 (83.7, 99.6)</td>
<td>95.4 (83.7, 99.6)</td>
</tr>
<tr>
<td>Herpes simplex virus 1</td>
<td>26</td>
<td>20</td>
<td>19</td>
<td>1</td>
<td>7</td>
<td></td>
<td>73.1 (53.7, 86.5)</td>
<td>82.6 (62.2, 93.6)</td>
</tr>
<tr>
<td>Herpes simplex virus 2</td>
<td>55</td>
<td>49</td>
<td>48</td>
<td>1</td>
<td>7</td>
<td></td>
<td>87.3 (75.7, 94.0)</td>
<td>96.0 (85.8, 99.7)</td>
</tr>
<tr>
<td>Cytomegalovirus</td>
<td>3</td>
<td>3</td>
<td>3</td>
<td>0</td>
<td>0</td>
<td></td>
<td>100 (38.3, 100)</td>
<td>100 (38.3, 100)</td>
</tr>
<tr>
<td>Varicella zoster virus</td>
<td>29</td>
<td>30</td>
<td>29</td>
<td>1</td>
<td>0</td>
<td></td>
<td>100 (86.1, 100)</td>
<td>100 (86.5, 100)</td>
</tr>
<tr>
<td>Human herpes virus 6</td>
<td>5</td>
<td>7</td>
<td>5</td>
<td>2</td>
<td>0</td>
<td></td>
<td>100 (51.1, 100)</td>
<td>100 (55.7, 100)</td>
</tr>
<tr>
<td>Streptococcus agalactiae</td>
<td>2</td>
<td>3</td>
<td>2</td>
<td>1</td>
<td>0</td>
<td></td>
<td>100 (29.0, 100)</td>
<td>100 (29.0, 100)</td>
</tr>
<tr>
<td>Streptococcus pneumoniae</td>
<td>27</td>
<td>30</td>
<td>27</td>
<td>3</td>
<td>0</td>
<td></td>
<td>100 (85.2, 100)</td>
<td>100 (85.7, 100)</td>
</tr>
<tr>
<td>Neisseria meningitidis</td>
<td>10</td>
<td>10</td>
<td>9</td>
<td>1</td>
<td>1</td>
<td></td>
<td>90.0 (57.4, 99.9)</td>
<td>100 (67.9, 100)</td>
</tr>
<tr>
<td>Haemophilus influenzae</td>
<td>40</td>
<td>39</td>
<td>39</td>
<td>0</td>
<td>1</td>
<td></td>
<td>97.5 (86.0, 99.9)</td>
<td>100 (89.3, 100)</td>
</tr>
<tr>
<td>Listeria monocytogenes</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>UTC</td>
<td>UTC</td>
<td>UTC</td>
</tr>
<tr>
<td>Escherichia coli K1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td></td>
<td>100 (16.8, 100)</td>
<td>100 (16.8, 100)</td>
</tr>
<tr>
<td>Cryptococcus neoformans/C. gattii</td>
<td>50</td>
<td>26</td>
<td>26</td>
<td>0</td>
<td>24</td>
<td>24</td>
<td>52.0 (38.5, 65.2)</td>
<td>57.9 (43.3, 71.0)</td>
</tr>
</tbody>
</table>

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### Apparent Coinfections

<table>
<thead>
<tr>
<th>Case</th>
<th>Expected result from routine testing</th>
<th>Result(s) of BIOFIRE FILMARRAY Meningitis/Encephalitis panel</th>
<th>Result(s) following confirmatory testing</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Haemophilus influenzae</td>
<td>H. influenzae, Listeria monocytogenes</td>
<td>H. influenzae</td>
</tr>
<tr>
<td>2</td>
<td>Cryptococcus neoformans</td>
<td>C. neoformans/C. gattii, S. pneumoniae</td>
<td>C. neoformans/C. gattii</td>
</tr>
<tr>
<td>3</td>
<td>Neisseria meningitidis</td>
<td>N. meningitidis, S. pneumoniae</td>
<td>N. meningitidis</td>
</tr>
<tr>
<td>4</td>
<td>H. influenzae</td>
<td>H. influenzae, HHV-6</td>
<td>H. influenzae</td>
</tr>
<tr>
<td>5</td>
<td>C. neoformans</td>
<td>C. neoformans/C. gattii, HSV-1</td>
<td>C. neoformans/C. gattii</td>
</tr>
<tr>
<td>6</td>
<td>HSV-1</td>
<td>HSV-1, HHV-6</td>
<td>HSV-1, HHV-6</td>
</tr>
<tr>
<td>7</td>
<td>CMV</td>
<td>CMV, VZV</td>
<td>CMV, VZV</td>
</tr>
<tr>
<td>8</td>
<td>H. influenzae</td>
<td>S. pneumoniae, N. meningitidis</td>
<td>S. pneumoniae, N. meningitidis</td>
</tr>
</tbody>
</table>

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### Limitations

- HHV-6 & CMV unusual in immunocompetent; may be clinically insignificant
- HHV-6 detection may result from germline integration
- Off-panel organisms...
  - Staphylococcus epidermidis, Cutibacterium acnes, many Gram-negative bacilli, Mycobacterium tuberculosis, Borrelia burgdorferi, Histoplasma capsulatum, Coccidioides immitis/posadasii, Blastomyces species, West Nile virus, Powassan virus, Jamestown Canyon virus, California encephalitis virus, Eastern Equine encephalitis virus, St. Louis encephalitis virus, Western Equine encephalitis virus, Ehrlichia species, Naegleria fowleri, Balamuthia mandrillaris, Toxoplasma and Acanthamoeba species, etc.
Limitations, continued

- Potential false-positive *S. pneumoniae*
  - Possibly due to amplified DNA contamination or *S. pneumoniae* colonization in healthcare workers or laboratory staff handling samples
- Tuberculous meningitis diagnosis delayed because of false-positive HSV-1 result

Meningitis/Encephalitis Panel Algorithm

Recommended testing for patients with findings suggestive of acute <8 days of symptoms meningitis or encephalitis. Infectious Diseases and/or Neurology specialty consultation is recommended.
- Lumbar puncture to obtain CSF
- Perform the following tests and retain CSF for additional testing:
  - Opening pressure (may not be indicated in neonates/infants)
  - Cytology, cell count and differential
  - Protein and glucose
  - Gram stain
  - Bacterial culture (aerobic with antimicrobial susceptibility testing)
  - Cryptococcal antigen, if indicated per patient risk factors
  - Blood cultures

If CSF testing available on-site:
- Streptococcus pneumoniae antigen
- Herpes simplex virus (HSV) PCR
- Enterovirus PCR
- Varicella-zoster virus PCR

Pathogen-specific infection management

If CSF available for testing:

- CSF parameters do not suggest infection
- CSF parameters suggest infection

Immunocompromised patient or patient with recurrent/recent antibiotic treatment

Order: CSFME / Meningitis/Encephalitis Pathogen Panel, POC, Spinal Fluid

Testing not available

Pathogen-specific infection management

- CSF parameters suggest infection
- CSF parameters do not suggest infection

Reevaluate patient
Additional testing should be tailored based on clinical presentation, risk factors, CSF parameters, and exposure history.

- If not previously performed, consider:
  - CLFA / Cryptococcus Antigen Screen with Titer, Spinal Fluid
  - HSVC / Herpes Simplex Virus (HSV), Molecular Detection, PCR, Spinal Fluid
  - Syphilis testing
    - SyPhGR / Syphilis IgG Antibody with Reflex, Serum
    - VDSF / VDRL, Spinal Fluid
  - Fungal and mycobacterial testing
    - FGEN / Fungal Culture, Routine
    - FS / Fungal Smear
    - CTB / Mycobacteria and Nocardia Culture
    - SAFB / Acid-Fast Smear for Mycobacterium
    - MTBPR / Mycobacterium tuberculosis Complex, Molecular Detection, PCR
    - HBRP / Histoplasma capsulatum/Blastomyces species, Molecular Detection, PCR, Varies
    - HICBL / Histoplasma/Blastomyces Panel, Spinal Fluid
    - SHSTO / Histoplasma Antibody, Serum
    - FUNSF / Fungitell, CSF (1,3-beta-D-glucan)
    - BLAST / Blastomyces Antibody by EIA, Serum
    - COXIS / Coccioidielles Antibody with Reflex, Serum
    - COCO / Coccioidielles Antibody, Spinal Fluid
    - CIMP/R / Coccioidielles immittis/posadasii, Molecular Detection, PCR

- Vector-borne testing
  - Mosquito-borne pathogens
    - WNS / West Nile Virus Antibody, IgG and IgM, Serum
    - WNVC / West Nile Virus Antibody, IgG and IgM, Spinal Fluid
    - LCWNV / West Nile Virus, Molecular Detection, PCR, Spinal Fluid
    - ABOPC / Arbovirus Antibody Panel, IgG and IgM, Spinal Fluid
    - ARBOP / Arbovirus Antibody Panel, IgG and IgM, Serum
  - Tick-borne pathogens
    - LNBAB / Lyme Central Nervous System Infection IgG with Antibody Index Reflex, Serum and Spinal Fluid (see Lyme Neuroborreliosis Diagnostic Algorithm)
    - PBORR / Lyme Disease, Molecular Detection, PCR, Varies
    - For emerging vector-borne diseases (eg, Powassan virus, Jamestown Canyon virus) contact your local public health laboratory for more information
  - Neuroimmunology testing (consider if antibody prevalence in epilepsy and encephalopathy [APE2]
    score is ≥4)
    - ENS1 / Encephalopathy, Autoimmune Evaluation, Serum
    - ENC1 / Encephalopathy, Autoimmune Evaluation, Spinal Fluid

- Parasitic testing
  - FLARP / Free-Living Amebae, Molecular Detection, PCR, Spinal Fluid, Fresh and Paraffin Tissue
  - PTOX / Toxoplasma gondii, Molecular Detection, PCR
  - TXMGP / Toxoplasma gondii Antibody, IgM and IgG (Separate Determinations), Serum

- Other considerations
  - BRBPS / Broad Range Bacterial PCR and Sequencing, Varies
  - HIVOX / HIV-1 and HIV-2 Antigen and Antibody Diagnostic Evaluation, Plasma
  - LC/JC / JC Virus, Molecular Detection, PCR, Spinal Fluid
Thank You