I. CURRENT DCLS LABORATORY TESTING

<table>
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<th>Test Type</th>
<th>Method</th>
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<tr>
<td>Identification Confirmation</td>
<td>MALDI-TOF (Bruker Biotyper)</td>
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<tr>
<td>Antimicrobial Susceptibility Testing (AST)*</td>
<td>Broth Microdilution/Sensititre</td>
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<tr>
<td>Phenotypic testing - Carbapenemase Production</td>
<td>Modified Carbapenem Inactivation Method (mCIM)</td>
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<tr>
<td>Resistance Mechanism Identification</td>
<td>Real-Time PCR</td>
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* Submitting laboratories are REQUIRED to provide AST results when submitting suspected CRE/CRPA isolates to DCLS for testing. These AST results are assessed upon receipt to determine potential pan-resistance.

TESTING METHODS
- **mCIM** – Phenotypic testing for carbapenemase production will be performed for all CRE and CRPA isolates.
- **AST** – The following drug Minimum Inhibitory Concentration (MIC) values will be determined for all CRE isolates and mCIM-positive CRPA isolates:
  1. Amikacin
  2. Aztreonam*
  3. Cefepime*
  4. Cefotaxime* (CRE only)
  5. Ceftazidime*  
  6. Ciprofloxacin
  7. Colistin*  
  8. Ertapenem (CRE only)  
  9. Imipenem*
  10. Levofloxacin
  11. Meropenem*
  12. Piperacillin/Tazobactam**
  13. Polymyxin B
  14. Tobramycin
  15. Trimethoprim/Sulfamethoxazole (CRE only)

* Testing required by the CDC’s Antibiotic Resistance Laboratory Network (ARLN) for CRE and CRPA isolates.
** Testing required by the CDC’s ARLN for Pseudomonas aeruginosa isolates.
- **Real-time PCR** - Analyses will be performed on the following resistance marker genes for all CRE isolates and mCIM-positive CRPA isolates:
  1. KPC
  2. NDM
  3. OXA-48-like
  4. VIM
  5. IMP
  6. mcr-1 and mcr-2

II. SPECIMEN TYPES
- Pure suspect CRE isolates on slant or plate media.
  1. Member of the Enterobacteriaceae family
  2. Resistance to at least 1 carbapenem:
     - imipenem, meropenem or doripenem = MIC of \( \geq 4 \) µg/mL
     - ertapenem = MIC of \( \geq 2 \) µg/mL

Note: *Morganella spp.*, *Proteus spp.*, and *Providencia spp.* have intrinsic elevated MICs to imipenem. MIC results for meropenem, doripenem, and/or ertapenem must be used to determine if these organisms should be forwarded to DCLS for testing.
- Pure suspect CRPA isolates on slant or plate media.
  1. Isolate identified as *Pseudomonas aeruginosa*
  2. Resistance to at least 1 carbapenem:
     - imipenem, meropenem or doripenem = MIC of \( \geq 8 \) µg/mL
  3. Mucoid *Pseudomonas aeruginosa* isolates will NOT be tested

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III. SPECIMEN TRANSPORT

- Ship isolates at room temperature.
- Submit a completed DCLS Clinical Microbiology/Virology Test Request form with isolate, to include the following:
  1) Complete patient information (including address)
  2) Complete submitter information
  3) Date of specimen collection
  4) Specimen source
  5) Organism identification, denoting “suspected carbapenem resistance”
  6) Outbreak ID number (if applicable)
- Include copy of laboratory’s AST results in specimen package
- **Shipping Address:**
  Division of Consolidated Laboratory Services
  Attn: CRE Team
  600 North 5th Street
  Richmond, Virginia 23219

IV. RESULTS REPORTING

- Turnaround time for reporting results will be within 6 business days of specimen receipt.
- Alert value results (results requiring additional reference laboratory testing) will be verbally reported within 1 working day of results. An alert email notification will be sent to the CDC as well.
- Non-alert-value, positive results (results NOT requiring additional reference laboratory testing) will be verbally reported to the submitting laboratory and VDH within 2 working days of results.
- Negative results will not be verbally reported.
- A hard copy report of final test results will be provided to the submitter by mail.
  - CRE final results will include: identification, AST, phenotypic, and molecular
  - CRPA final results will include: if mCIM negative, identification and phenotypic only; if mCIM positive, identification, AST, phenotypic, and molecular
- VDH will receive electronic reporting of results.
- Reference laboratory results for additional characterization testing will be provided to the submitting laboratory and VDH within 2 working days of receipt from the reference laboratory.

V. CONTACT INFORMATION

- For questions regarding testing, specimen collection or shipment, please contact one of the following:
  - Dr. LaToya Griffin-Thomas (804-648-4480, ext. 281)
  - Microbial Reference Laboratory (804-648-4480, ext. 345 or 251)
  - Molecular Detection and Characterization Laboratory (804-648-4480, ext. 298)
  - DCLS 24/7 Emergency Mobile Number (804-335-4617)

VI. RESOURCES

Updated information on CRE/CRPA testing can be found on the DCLS website, under the “Hot Topics” link:

Link to DCLS Clinical Microbiology/Virology Test Request Form: