Antibiotic Resistance Laboratory Network
Northeast Update and Expanded Drug Susceptibility Testing

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Wadsworth Center, NYSDOH
Updated AR Threats Report Fall anticipated 2019
AR Lab Network

- Provides gold standard methods and diagnostics in all 50 states, 5 large cities, and Puerto Rico, and includes 7 Regional labs with expanded testing capabilities
- Testing complements CDC’s Containment Strategy to help identify, respond and contain the urgent and serious threats and keep new threats from spreading

- **In 2018, the AR Lab Network tested 14,054 CRE and 10,279 CRPA.**
- **40% CRE and 3.5% CRPA were found to contain one of the 5 main carbapenemases.**
Core Testing:
- CRO Colonization testing
- CRE/CRPA Isolate Characterization
- Confirmation/Detection Discordant or Novel mechanisms
- Emerging threats (mcr genes and MDR Acinetobacter spp.)
- Pan-resistance
- *Candida* susceptibility (Mycology Laboratory)
- *Candida auris* identification and screening (Mycology Laboratory)
Acquired Carbapenemase genes:

- $\text{bla}_{\text{KPC}}$
- $\text{bla}_{\text{NDM}}$
- $\text{bla}_{\text{VIM}}$
- $\text{bla}_{\text{IMP}}$
- $\text{bla}_{\text{OXA-48}}$

$\text{(CP= carbapenemase producing)}$

**CRE**
Carbapenem-Resistant Enterobacteriaceae

**CRPA**
Carbapenem-Resistant Pseudomonas aeruginosa

**CP-CRE**
**CP-CRPA**
AR Lab Network Testing Approach

**Tiered Testing**

- **Network of participating clinical laboratories**
  - CT, MA, ME, NH, NJ, NY, NYC, RI, VT
  - AR isolates

- **State/Local testing**
  - Species identification
  - Confirmatory AST
  - Phenotypic screening
  - Molecular detection of AR genes

- **Regional lab testing**
  - ARLN
  - Confirmatory testing
  - Colonization screening
  - WGS
  - Targeted surveillance for AR pathogens

- **Isolates for further testing**

**Wadsworth Center Northeast**

**Confirmatory testing**

**WGS**

**Applied research**
Rapid Spread of Carbapenemase-Producing (CP) Enterobacteriaceae in the United States

KPC-CRE found in the US spread from 2 states in 2001 to 49 states, DC, and PR in 16 years

States with *Klebsiella pneumoniae* carbapenemase (KPC)-producing Carbapenem-resistant Enterobacteriaceae (CRE) confirmed by CDC
Prevalence Worldwide Carbapenemase Genes (KPC, NDM, OXA, VIM, IMP)

Logan et al., J Infect Dis. 2017. Feb 15
Regional Laboratory Testing Updates...

- CRAB - Detection of OXA-23, -24/40, -58 Multiplex in Isolates and Rectal Swabs – April 22, 2019

93 Alerts from 5 Jurisdictions
- OXA-23 (88)
- OXA-24/40 (5)
Regional Laboratory Testing Updates...

- Limitations with IMP variant detection by Cepheid Carba-R (IMP-1 only)
- Validation of CDC IMP assay for Isolates and Rectal Swabs
- Validation of CDC MCR-1/MCR-2 assay for Isolates and Rectal Swabs
- Validation of mSuperCARBA™ CHROMagar for efficient and enhanced isolate recovery from positive colonization screenings
- 84% isolate recovery
Regional Laboratory Testing Updates...

Whole-genome sequencing (WGS)

• Identify all AR genes in genome
  – Database includes over 5,500 unique genes
• Identify emerging and novel AR mechanisms
• Identify AR genes to the variant level (\(bla_{KPC-2}\) vs \(bla_{KPC-5}\))

• Outbreak investigations
  – Identify transmission events between patients
Antibiotic Resistance WGS at Wadsworth: Pipeline Overview

Illumina Sequencing Reads

\[ \downarrow \]

\textit{de novo} assembly & quality control

- Are the bacteria similar? (Multilocus Sequence Typing)
- What antibiotic resistance genes are present?
- Where are the antibiotic resistance genes located? Plasmid?

MLST

Gene Variant

Total analyzed with pipeline: >800
### Supplemental Reporting through LIMS

<table>
<thead>
<tr>
<th><strong>Antimicrobial Resistance Gene Analysis - Whole-Genome Sequencing</strong> *</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gene(s) Identified:</td>
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<tr>
<td><strong>blaNDM-1</strong></td>
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<table>
<thead>
<tr>
<th><strong>Multilocus Sequence Typing Analysis</strong> *</th>
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<tbody>
<tr>
<td><strong>MLST ST:</strong></td>
</tr>
<tr>
<td><strong>1399</strong></td>
</tr>
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</table>

* The performance characteristics of this test were determined by the Wadsworth Center. It has not been cleared or approved by the U.S. Food and Drug Administration.

END OF REPORT
A Look at the DATA.....
AR Lab Network – Northeast Regional Laboratory 2018 Testing

CRE Isolate Submission
- 834 Tested
- 542 identified as CP-CRE (65%)
  - 479 KPC
  - 46 NDM
  - 21 OXA-48 like
  - 1 KPC/NDM
  - 1 NDM/OXA-48

CRPA Isolate Submission
- 1418 Tested
- 55 identified as CP-CRPA (~4%)
  - 29 VIM
  - 15 KPC
  - 3 OXA-2/OXA50 like
  - 2 NDM
  - 2 IMP
  - 1 IMP/NDM

CRE/CRPA Colonization Screening
- 615 Rectal Swabs
- 4 States and NYC
- 70 Health care facilities
WGS-determined Carbapenemase Gene Variants

**bla\textsubscript{KPC} Variants Identified Using WGS**
- Total = 298
- Variants:
  - KPC-2: 63%
  - KPC-3: 30%
  - KPC-4: 6%
  - KPC-5: 1%

**bla\textsubscript{NDM} Variants Identified Using WGS**
- Total = 100
- Variants:
  - NDM-1: 46%
  - NDM-4: 3%
  - NDM-5: 44%
  - NDM-7: 7%

**bla\textsubscript{OXA-48} Variants Identified Using WGS**
- Total = 23
- Variants:
  - OXA-48: 26%
  - OXA-181: 22%
  - OXA-232: 52%

**bla\textsubscript{VIM} Variants Identified Using WGS**
- Total = 40
- Variants:
  - VIM-2: 75%
  - VIM-4: 17%
  - VIM-5: 5%
  - VIM-23: 3%

**bla\textsubscript{IMP} Variants Identified Using WGS**
- Total = 4
- Variants:
  - IMP-13: 25%
  - IMP-15: 25%
  - IMP-18: 25%
  - IMP-27: 25%

**bla\textsubscript{OXA-23} Variants Identified in AMR Pipeline Isolates**
- Total = 116
- Variants:
  - OXA-23: 2%
  - OXA-225: 98%
Northeast Region AR Lab Network CRE colonization screenings, 2017-2019

Cepheid Xpert® Carba-R PCR Results

Total: 1033 rectal swabs

2019 - >1300 rectal swabs tested
CDC's AR Lab Network closes the gap between local capabilities and the data needed to combat AR in healthcare and the community.

**CRE and CRPA Isolate Testing, 2018***

<table>
<thead>
<tr>
<th>Percentage of Isolates (%)</th>
<th>NY/NYC</th>
<th>NE Region</th>
<th>National</th>
</tr>
</thead>
<tbody>
<tr>
<td>CRE isolates tested NY and NYC: 1088</td>
<td>65.3%</td>
<td>50.40</td>
<td>37.90</td>
</tr>
<tr>
<td>% CP&amp;-CRE: 65.3</td>
<td>34.70</td>
<td>49.60</td>
<td>62.10</td>
</tr>
</tbody>
</table>

CRPA isolates tested NY and NYC: 1418
% CP&-CRPA: 3.5

*As of May 2019; data subject to change
*Graphs adjusted for number of isolates tested
& CP is defined as PCR-positive for at least one of the carbapenemase genes tested
Expanded Antimicrobial Susceptibility Testing (ExAST) for *Enterobacteriaceae* producing a metallo-beta-lactamase (MBL)

- NDM most prevalent MBL in *Enterobacteriaceae*
- Multidrug resistant due to additional AR genes
- Resistant to newer, more recently approved drugs for CRE:
  - Ceftazidime-avibactam
  - Meropenem-vaborbactam
  - Plazomycin
  - Imipenem-relebactam

(Mostly effective against KPC-producing CRE)
Expanded Antimicrobial Susceptibility Testing (ExAST) for Enterobacteriaceae producing a metallo-beta-lactamase (MBL)

New Antimicrobials Submitted to FDA or in Phase 3 Trials:
• Cefidericol
• Aztreonam-avibactam
• Cefepime-taniborbactam

(Show efficacy for treatment of NDM-producing CRE)

Recommended Treatment: Aztreonam-avibactam
(Aztreonam + Ceftazidime-Avibactam)
How is AST performed for Aztreonam-avibactam?

HP Printing technology repurposed for dispensing antibiotics instead of ink

- HP D300e allows for in-house creation of BMD panels with novel antibiotics
- The combinations are **not yet included in commercially available BMD panels**
  - Aztreonam
  - Ceftazidime-Avibactam
  - **Aztreonam-Avibactam**
  - Aztreonam-Ceftazidime-Avibactam
ExAST

- Multi-site Pilot Validation
- Northeast and Mid-Atlantic Regions
- ExAST Validation sign-off February 28, 2019
- Testing requests received March 2019
- 18 isolates tested to date
“What you need to know to Access Expanded Antimicrobial Susceptibility Testing (ExAST) for Hard-to-Treat Infections at Wadsworth Center”

What isolates can be submitted?

Enterobacteriaceae.

- Test non-susceptible to all beta-lactams including ceftazidime-avibactam or meropenem-vaborbactam
- OR
- Confirmed NDM, VIM or IMP by a molecular method
What is the testing process?

Isolate Confirmation

- *Enterobacteiraceaee* – MALDI-TOF MS
- Non-susceptible to beta-lactams – GNX2F
- Carbapenemase production – mCIM
- Carbapenemase gene-coded resistance – Real-time PCR

ExAST Panel

- Aztreonam
- Ceftazidime-Avibactam
- Aztreonam-Avibactam
- Aztreonam-Ceftazidime-Avibactam

Final Report

3 days
How is Testing Requested?

- Healthcare Providers, Clinical Laboratories, State Public Health Laboratories and Epidemiologists
  
- Pre-authorization/Approval Required

ARLNCORENY@health.ny.gov

- ExAST Fact Sheet
- Pre-authorization Approval Form
- NYS Specific Requisition
- Guidance for Isolate Submission
Pre-authorization Form

Section 1

Section 3: Submission Criteria (check all that apply)
A copy of initial AST (raw data preferred) and molecular test results (if available) must be sent with the isolate.

☐ Infectious disease physician and/or facility’s infection control department have been notified of or consulted on test request

☐ Enterobacteriaceae tests non-susceptible to all beta-lactams, including ceftazidime-avibactam

☐ Enterobacteriaceae tests non-susceptible to all beta-lactams, including meropenem-vaborbactam

☐ Enterobacteriaceae confirmed as an metallo-β-lactmase (MBL) producer with laboratory test (e.g. eCIM, MBL Etest)

Enterobacteriaceae with: NDM gene ☐ IMP gene ☐ VIM gene ☐

State Public Health Laboratory: ____________________________________________________________

Point of Contact Name: ________________________________________________________________

Phone Number: ________________________________________________________________

Fax Number (results may be returned by secure fax): ________________________________

Email (results may be returned by encrypted email): ________________________________
# ExAST Specific Requisition

## Patient Demographics

<table>
<thead>
<tr>
<th>Last Name *</th>
<th>First Name *</th>
<th>MI</th>
<th>DOB *</th>
<th>Sex</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Street Address</th>
<th>City</th>
<th>State</th>
<th>Zip Code</th>
</tr>
</thead>
</table>

| NYS County of Residence * | NYS DOH Outbreak Number | CDESS Case Number | Submitter's Reference Number |

## Submitter (Laboratory report will be sent to)

<table>
<thead>
<tr>
<th>Name and Address *</th>
</tr>
</thead>
<tbody>
<tr>
<td>Name</td>
</tr>
<tr>
<td>Address</td>
</tr>
<tr>
<td>City</td>
</tr>
<tr>
<td>Telephone Number</td>
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</table>

## Specimen Information

<table>
<thead>
<tr>
<th>Specimen is:</th>
<th>Collection Date * MM / DD / YYYY</th>
</tr>
</thead>
<tbody>
<tr>
<td>Isolate</td>
<td></td>
</tr>
<tr>
<td>Primary Specimen</td>
<td></td>
</tr>
<tr>
<td>Autopsy Specimen</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Source / Specimen Type *</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Time Collected (if applicable for test)</th>
</tr>
</thead>
</table>

## Laboratory Examination Requested

- [ ] Bacterial
- [ ] Fungal
- [ ] Mycobacterial
- [ ] Parasitic
- [ ] Serology
- [ ] Viral

[www.wadsworth.org/IDtesting](http://www.wadsworth.org/IDtesting)

## Suspected Organism / Agent

- [ ] Identification / Confirmation
- [ ] TB Fast Track [www.wadsworth.org/mycobac/testtrack.htm](http://www.wadsworth.org/mycobac/testtrack.htm)
- [ ] Viral Encephalitis Panel [www.wadsworth.org/divisions/infdis/enceph/form.htm](http://www.wadsworth.org/divisions/infdis/enceph/form.htm)
- [ ] Other (specify) AMR-Expanded AST (Aztreonam-Avibactam)

## Submitting lab findings:

- [ ] Smear/Stain/Other results

## Comments

Provide copy of AST Results

<table>
<thead>
<tr>
<th>Specimen submitted on/in:</th>
<th>Media</th>
<th>Preservative</th>
<th>Tissue cell line</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>Relevant Exposure:</th>
<th>Contact known case</th>
<th>Food/water</th>
<th>Nosocomial</th>
</tr>
</thead>
<tbody>
<tr>
<td>Travel</td>
<td>Location &amp; Dates</td>
<td>Animal</td>
<td>Arthropod</td>
</tr>
<tr>
<td>Type</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
ExAST Testing

Example # 1:

• Physician request via State Epidemiologist
  *Escherichia coli*, NDM from Urine

  Resistant to all drugs tested by Clinical Laboratory

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**Expanded Broth Microdilution Susceptibility**

<table>
<thead>
<tr>
<th>Antibiotic Combination</th>
<th>MIC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ceftazidime/avibactam</td>
<td>&gt;64/4 ug/ml</td>
</tr>
<tr>
<td>Aztreonam</td>
<td>64 ug/ml</td>
</tr>
<tr>
<td>Aztreonam/avibactam</td>
<td>Not interpretable; 8/4 ug/ml</td>
</tr>
</tbody>
</table>

[Note]: For aztreonam-avibactam, a minimum inhibitory concentration (MIC) is reported without an interpretation because clinical breakpoints for this drug combination have not been established. This drug demonstrates in vitro activity against metallo-β-lactamase (MBL) producing Enterobacteriaceae. Its clinical efficacy is under evaluation in clinical trials. Surveillance data indicate that MICs of MBL-producing Enterobacteriaceae (n=580) range from <=0.015/4 to 8/4 ug/ml.
Example # 2:

- Clinical laboratory request via State Epidemiologist 
  *Klebsiella pneumoniae*, nearly Pan-resistant from Tracheal Aspirate

  - Resistant to all drugs tested by Clinical Laboratory including 
    ceftazidime-avibactam and meropenem-vaborbactam 
    (Cefepime susceptible)

  - Carbapenemase-production and molecular testing for 
    carbapenemase genes not performed by Clinical laboratory
Example # 2:

- Isolate forwarded for CP-CRE testing at Regional Laboratory

*Klebsiella pneumoniae*

NDM/OXA-48

**Expanded Broth Microdilution Susceptibility**

<table>
<thead>
<tr>
<th>Antibiotic Combination</th>
<th>MIC Requirement</th>
</tr>
</thead>
<tbody>
<tr>
<td>Ceftazidine/avibactam</td>
<td>Resistant; &gt;64/4 µg/ml</td>
</tr>
<tr>
<td>Aztreonam MIC</td>
<td>Resistant; &gt;64 µg/ml</td>
</tr>
<tr>
<td>Aztreonam/avibactam MIC</td>
<td>Not interpretable; 1/4 µg/ml</td>
</tr>
</tbody>
</table>

**NOTES:**

[1] For aztreonam-avibactam, a MIC is reported without an interpretation because clinical breakpoints for the drug combination have not been established. This drug demonstrates in vitro activity against metallo-β-lactamase (MBL) producing Enterobacteriaceae. In a large surveillance study, the MICs of 580 MBL-producing Enterobacteriaceae ranged from ≤0.015/4 to 8/4 µg/mL with a mode of 0.125/4 µg/mL. The clinical efficacy of this drug combination for treating infections is unknown and under evaluation in a clinical trial. Because of limited treatment options, the 2018 Sanford Guide recommends the combination of ceftazidime-avibactam + aztreonam as a last resort for the treatment of serious infections caused by MBL-producing Enterobacteriaceae (Note: the agent in this regimen that is active against MBL-producing bacteria is aztreonam-avibactam). Please see the Sanford Guide for additional information if the ceftazidime-avibactam + aztreonam drug combination is considered for treatment.
### Escherichia coli

<table>
<thead>
<tr>
<th>Source</th>
<th>Carbapenemase</th>
<th>WGS</th>
<th>AZT/AVI MIC</th>
</tr>
</thead>
<tbody>
<tr>
<td>Blood</td>
<td>NDM</td>
<td>NDM-5 CTX-M-15</td>
<td>0.06/4</td>
</tr>
<tr>
<td>Urine</td>
<td>NDM</td>
<td>NDM-5 TEM-1</td>
<td>8/4</td>
</tr>
<tr>
<td>Urine</td>
<td>NDM/OXA-48</td>
<td>NDM-5 OXA-48</td>
<td>8/4</td>
</tr>
<tr>
<td>Sputum</td>
<td>NDM/OXA-48</td>
<td>NDM-5 OXA-181</td>
<td>2/4</td>
</tr>
<tr>
<td>Blood</td>
<td>NDM</td>
<td>NDM-5 CTX-M-15 TEM-1</td>
<td>4/4</td>
</tr>
<tr>
<td>Perirectal</td>
<td>NDM</td>
<td>NDM-1 TEM-1</td>
<td>0.12/4</td>
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<tr>
<td>Perirectal</td>
<td>NDM</td>
<td>Pending</td>
<td>8/4</td>
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</table>
## Klebsiella pneumoniae

<table>
<thead>
<tr>
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<th>Carbapenemase</th>
<th>WGS</th>
<th>AZT/AVI MIC</th>
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</thead>
<tbody>
<tr>
<td>Tracheal Aspirate</td>
<td>NDM/OXA-48</td>
<td>NDM-5, OXA-232, CTX-M-15, SHV-11, TEM-1</td>
<td>1/4</td>
</tr>
<tr>
<td>Blood</td>
<td>NDM</td>
<td>NDM-1, CTX-M-15, SHV-11, TEM-1</td>
<td>0.25/4</td>
</tr>
<tr>
<td>Urine</td>
<td>NDM</td>
<td>NDM-9, CTX-M-15, SHV-11, TEM-1</td>
<td>0.25/4</td>
</tr>
<tr>
<td>Perirectal</td>
<td>NDM</td>
<td>NDM-5, CTX-M-15, SHV-11, TEM-1</td>
<td>0.25/4</td>
</tr>
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</table>
### Enterobacter cloacae

<table>
<thead>
<tr>
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<th>AZT/AVI MIC</th>
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</thead>
<tbody>
<tr>
<td>Urine</td>
<td>NDM</td>
<td>NDM-1</td>
<td>0.06/4</td>
</tr>
<tr>
<td>Blood</td>
<td>NDM</td>
<td>NDM-1</td>
<td>0.5/4</td>
</tr>
<tr>
<td></td>
<td></td>
<td>CTX-M-15</td>
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<td>TEM-1</td>
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</tr>
<tr>
<td>Urine</td>
<td>NDM</td>
<td>NDM-1</td>
<td>1/4</td>
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<tr>
<td></td>
<td></td>
<td>TEM-1</td>
<td></td>
</tr>
<tr>
<td>Blood</td>
<td>NDM</td>
<td>Pending</td>
<td>2/4</td>
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### Morganella morganii

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<th>AZT/AVI MIC</th>
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<tbody>
<tr>
<td>Tracheal Aspirate</td>
<td>NDM/KPC</td>
<td>NDM-7</td>
<td>0.5/4</td>
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<tr>
<td></td>
<td></td>
<td>KPC-2</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>TEM-1</td>
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</tr>
</tbody>
</table>
ARLNCORENY@health.ny.gov

https://wadsworth.org/antimicrobial-resistance-laboratory-network
Acknowledgements

Wadsworth Center AR Team
AR Fellows
Wadsworth Center Core Facilities

NYS Bhai, NYS MARO, BCDC Epidemiology

Center for Disease Control and Prevention: ARX, DHQP

Association of Public Health Labs: Nikki Marchan, Kelly Wroblewski

Clinical Laboratories
Sentinel Laboratories
State/City Public Health Labs
State/City HAI Epi/Teams
# Therapeutic Options for CRE Infections

<table>
<thead>
<tr>
<th>Drug Status</th>
<th>Drug</th>
<th>Activity for MDROs</th>
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<tbody>
<tr>
<td></td>
<td></td>
<td>KPC</td>
</tr>
<tr>
<td>Drugs of Last Resort</td>
<td>Colistin</td>
<td>?</td>
</tr>
<tr>
<td></td>
<td>Tigecycline</td>
<td>✔</td>
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<tr>
<td>Recently Approved</td>
<td>Ceftazidime-avibactam</td>
<td>✔</td>
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<tr>
<td></td>
<td>Meropenem-vaborbactam</td>
<td>✔</td>
</tr>
<tr>
<td></td>
<td>Eravacycline</td>
<td>✔</td>
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<tr>
<td></td>
<td>Plazomycin</td>
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<tr>
<td></td>
<td>Imipenem-relebactam</td>
<td>✔</td>
</tr>
<tr>
<td>Recently Submitted to FDA or Phase 3</td>
<td>Cefidericol</td>
<td>✔</td>
</tr>
<tr>
<td></td>
<td>Aztreonam-avibactam</td>
<td>✔</td>
</tr>
<tr>
<td></td>
<td>Cefepime-taniborbactam</td>
<td>✔</td>
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</table>