Role of the Clinical Microbiology Laboratory in Infection Prevention

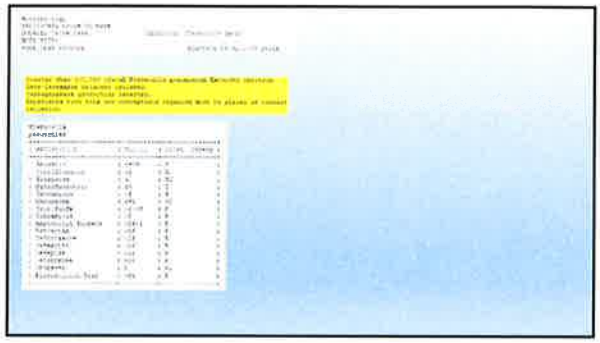
Rita M. Hollaway, PhD, ABMM
October 6, 2017

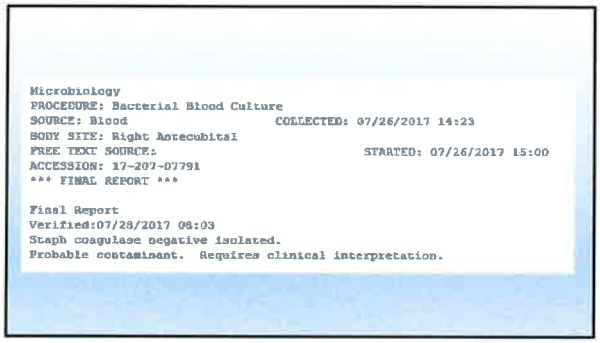
Contributions of Microbiology Laboratory

- Microbiology representative on the Infection Prevention Committee
- Aid in interpreting culture results and attaching isolation comments for multi-drug resistant organisms
- Recognition of new pathogens
- Using newer technology to decrease time to diagnosis of infections
- Monitoring resistant organisms and detection of new antibiotic resistance
- Surveillance efforts and outbreak investigations including strain typing

Microbiology Comments that Guide Interpretation of Results

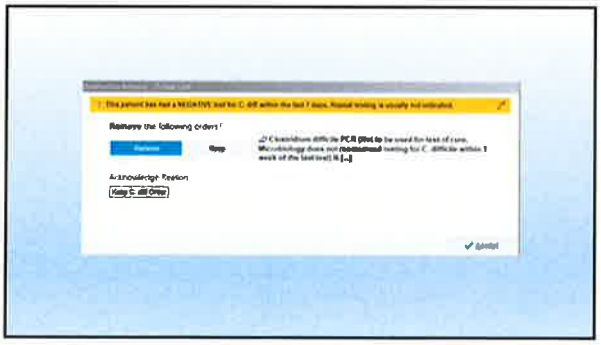
- Contact isolation for multi-drug resistant organisms
- Blood culture contamination
- *Clostridium difficile* testing





```
Microbiology
PROCEDURE: Bacterial Blood Culture
SOURCE: Blood                          COLLECTED: 07/26/2017 14:23
BODY SITE: Right Antecubital
FREE TEXT SOURCE:                                 STARTED: 07/26/2017 15:00
ACCESSION: 17-207-07791
^^^ FINAL REPORT ^^^

Final Report
Verified:07/28/2017 06:03
Staph coagulase negative isolated.
Probable contaminant.  Requires clinical interpretation.
```



Recognition of New Pathogens
(Example: *Candida auris*)

- *Candida auris* is emerging fungal pathogen reported on four continents
  - First described in Japan in 2009; recovered from patient's external ear discharge
  - Some isolates resistant to all three major classes of antifungals
- Problem for laboratory
  - Often misidentified as *C. haemuloni* and other yeasts by traditional biochemical methods and some MALDI-TOF databases
  - Laboratory solution to detection
    - Perform susceptibilities on all yeasts recovered from sterile sites
    - Identify resistant isolates by molecular sequencing of the D1-D2 region of 28S ribosomal DNA

Why Do We Care About *Candida auris*?

- Possible association with health-care outbreaks
- U.S. cases in New York, New Jersey, Maryland and Illinois (7 cases)
- Several findings suggest transmission occurred within hospital
  - Similar molecular profiles for isolates recovered from patients at the same hospital
  - Colonized skin and other body sites weeks to months after initial infection.
  - Recovered *C. auris* from environmental sources in one patient's room.

Monitoring Resistant Organisms and Detection of New Antibiotic Resistance

Rapid Confirmation of Tuberculosis in Patients

Direct Amplification of *M. tuberculosis* complex in Clinical Samples

- Three FDA-approved tests for respiratory secretions
- Always order NAATs in conjunction with culture to recover organisms for further characterization and to perform drug susceptibility testing

Xpert ® MTB / RIF

- Nested real-time PCR test for detection of *M. tuberculosis* complex in raw sputum and concentrated sputum sediment
- Also detects rifampin – resistance associated mutations of *rpoB* gene
  - Does not provide confirmation of rifampin susceptibility
  - Other mechanisms of resistance may exist

Comparison of Sputum Results

	Raw	Concentrated
Smear-positive > 99% sensitivity		100%
Smear-negative > 79% sensitivity		60%
Specificity > 97%		99%

- Overall specificity varied depending on percentage of patients with AFB smear-positive tuberculosis in population tested

Xpert ® MTB / RIF

Two hundred thirty-nine (239) tests were reviewed over a 7 month period

MTB/Rif PCR Assay	AFB Smear	AFB Culture	Number of tests
MTB detected/Rif resistance not detected	AFB Seen	MTB isolated (Rif susceptible)	19
MTB detected/Rif resistance not detected	No AFB Seen	MTB isolated (Rif susceptible)	3
MTB NOT detected	No AFB Seen	No MTB isolated	159
MTB NOT detected	No AFB Seen	MTB isolated (Rif susceptible)	3
MTB NOT detected	No AFB Seen	MOTT isolated	55

72 yo F with diabetes and hypertension from Guatemala

- Presented to PCP with 3 mo hx of cough, fever and chills
- Chest X-ray showed RUL consolidation
- CT chest demonstrated cavitary disease
- Patient was caretaker for daughter diagnosed with pulmonary tuberculosis in previous year
- Admitted to Parkland for further evaluation
- Sputum collected on day of admission;
  - *AFB smear negative but MTB PCR positive*
- RIPE therapy started
- Patient returned home next day
- *M. tuberculosis* was not detected in culture until 22 days later

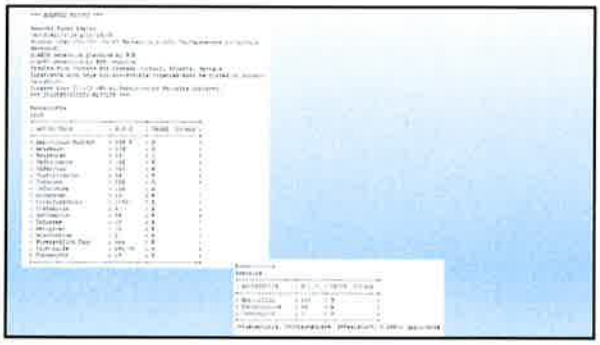
51 yo M with 2 mo hx of cough and weight loss

- Remote hx of exposure to tuberculosis; lives with uncle who had tuberculosis 40 to 50 years ago
- Incarceration and release in 2014
- Hx of tobacco and recreational drug use
- Three sputa collected over 2 days;
  - *AFB smears and MTB PCR positive*
- Started on RIPE therapy while in hospital
- *M. tuberculosis* recovered in culture after 10 days

Detection of Multi-drug Resistant Organisms

31 yo F presented with right flank pain and obstructive uropathy

- PMH included recent lap band gastrectomy in India
- Underwent ureteroscopy for right distal ureteral calculus
- Postoperative stent pain
- Collected urine for culture
- Complicated urinary tract infection with multi-drug resistant *Escherichia coli* and *Enterococcus faecalis*



Detection of New Antibiotic Resistance It's Complicated

- Carbapenem – resistant Enterobacteriaceae (CRE) have two primary resistance methods
  - Overproduction of beta lactamases plus decreased outer membrane permeability OR
  - Production of carbapenemases – usually mediated by plasmids (larger concern for infection control)
- CRE bloodstream infections associated with significantly higher mortality rates than those with carbapenem-susceptible bacteria
- CREs include Ambler molecular class A (ex. KPC), Ambler class B (ex. New Delhi metallo-β-lactamase (NDM) and Ambler class D (ex. Oxacillinase-48)

Challenges to Accurately Detecting CRE in Laboratory

- CLSI (Clinical Laboratory Standards Institute) lowered carbapenem breakpoints in 2010
- However, manufacturers of automated test devices were slow to provide labs with tests whose performance has been validated against the new CLSI breakpoints
- Many labs rely on older, higher breakpoints combined with phenotypic tests to detect CRE (know what your lab uses)

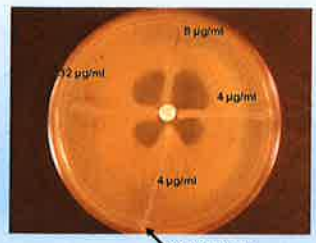
Methods for Carbapenemase Detection in Bacterial Colonies

Phenotypic methods
- Modified Hodge
- Carba NP Test

Genotypic methods
- Xpert® Carba-R

MODIFIED HODGE TEST

Hodge (Clover Leaf Test): KPC-producing Isolates With Varying Levels of Carbapenem Susceptibility

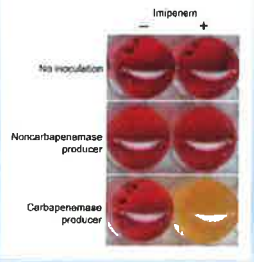


3 isolates of *K. pneumoniae* (9, 12, & 3 o'clock)

1 *Enterobacter* sp. (6.30 position)

Meropenem MICs shown beside streaks

Carba NP test



Xpert® Carba-R is a rapid PCR test designed to confirm the carbapenem-resistance genes encoding KPC, NDM, VIM, OXA-48 and IMP in 48 minutes.

NDM Strikes Fear in the Hearts of Healthcare Providers

- First and second Texas isolates recovered at Parkland
- May carry resistance genes to other antibiotics
- Carbapenemase not inhibited by beta lactamase inhibitors; zinc dependent enzyme
- Predominately in Indian Subcontinent and Middle East

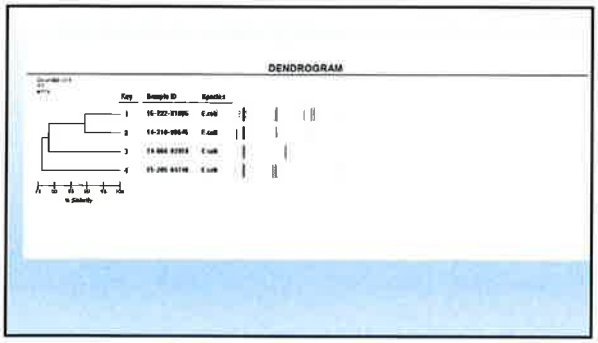
Surveillance Efforts and Outbreak Investigations including Strain Typing

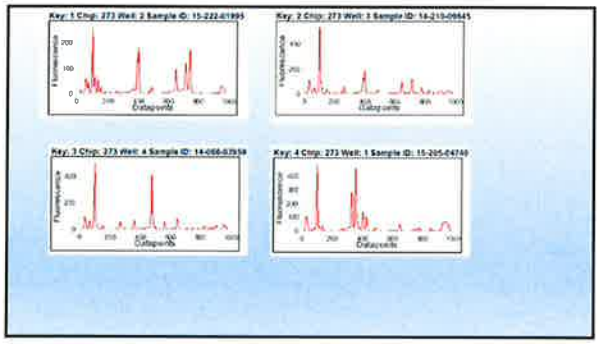
- Detection of NDM in urine prompted a survey of all CRE recovered in 12-month span at Parkland
- Performed Diversilab DNA fingerprinting on four CRE

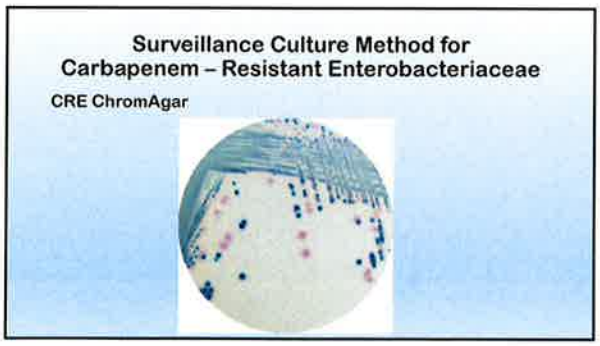
Parkland Health & Hospital System
Microbiology Laboratory
DiversiLab DNA Fingerprinting Results

MDRO or Pathogen: *Escherichia coli*, CRE
Date Requested: 8/17/2015

Culture Source	Culture Date	Location	PHHS Type
urine	8/10/2015	6W	G1P1 EC CRE
R foot wound	7/29/2014	7SS	G2P1 EC CRE
peritoneal fluid	8/7/2014	9N MICU	G3P1 EC CRE
urine	7/24/2015	Urology	G4P1 EC CRE

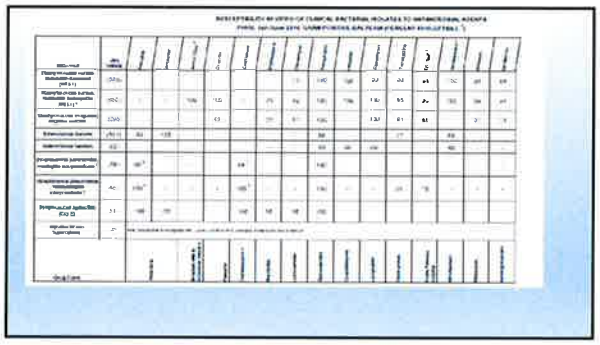

DENDROGRAM

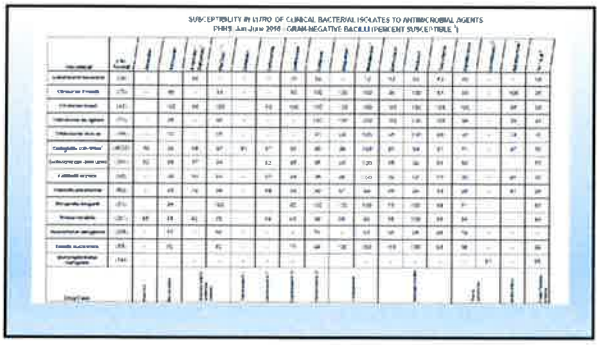



Surveillance Culture Method for
Carbapenem – Resistant Enterobacteriaceae
CRE ChromAgar

Generation of Antibiograms to Monitor Resistant Organisms

- Performed every six months at Parkland
- Generate Gram positive and Gram negative antibiograms with first isolates from patients





A Need for Integrated Infection Prevention Efforts Between Hospitals and Feeder Facilities

- Noted increase in *Acinetobacter* infections at university hospitals
- Initiated study to examine 487 clinical isolates from 212 hospitalized patients
- Patients admitted from outside healthcare facilities (nursing homes and long-term care facilities) accounted for majority of initial (59%) and total (63%) isolates
- Multi-drug resistant isolates more common among those who had resided at HCFs; >90% of patients for HCFs vs 62% of patients from home
- Prevention may need to focus on pre-hospital risk factors – reducing inappropriate use of antibiotics in community, avoiding unnecessary catheter placement and shortening hospital stays when possible

Thank you for your attention!