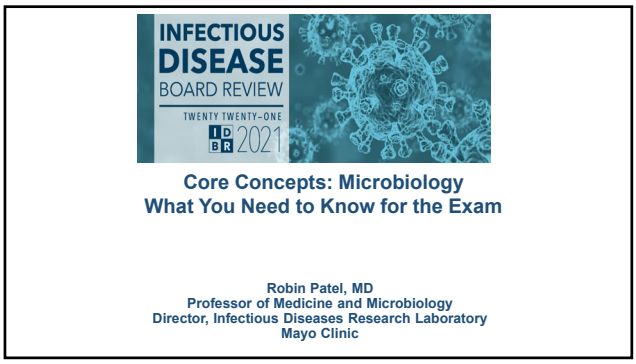
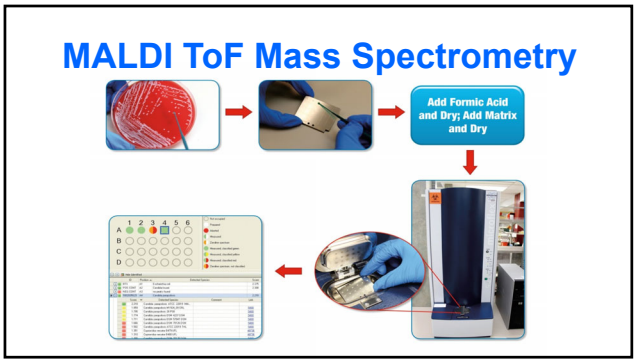
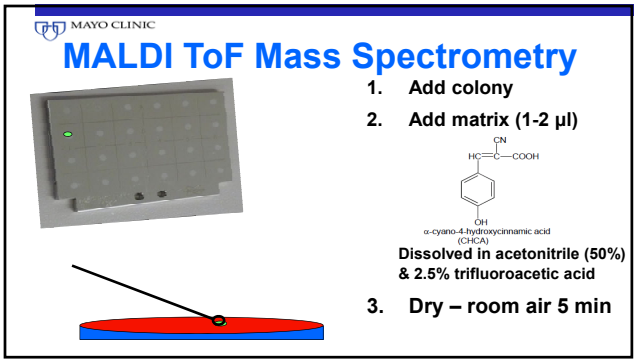
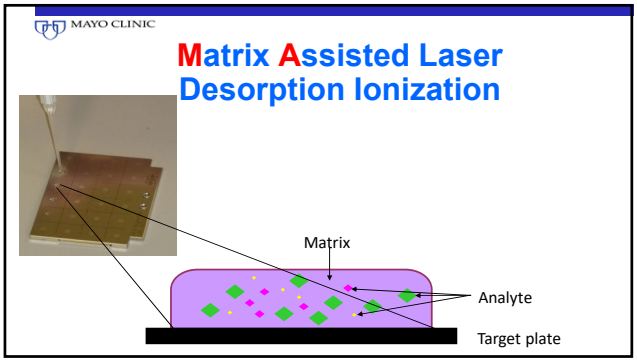
# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*

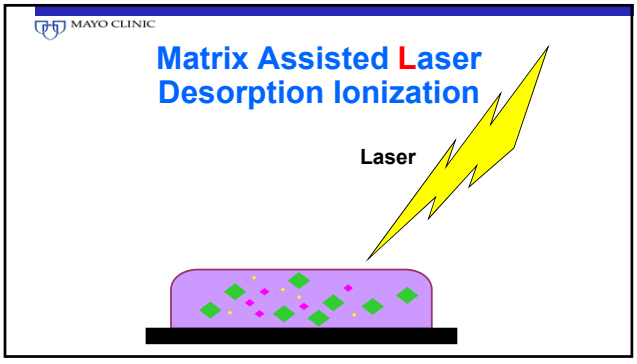






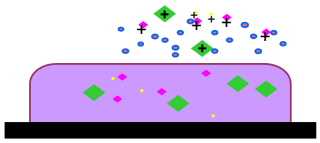



# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*

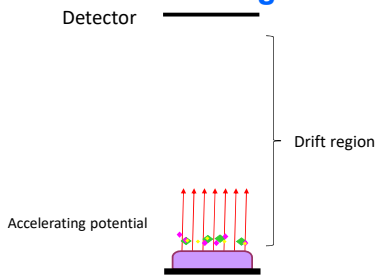
MAYO CLINIC

## Matrix Assisted Laser Desorption Ionization



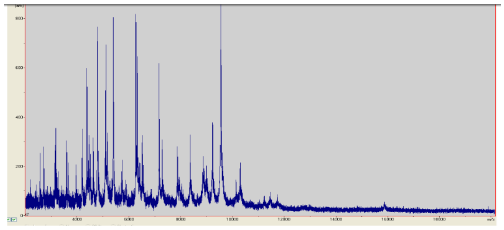
MAYO CLINIC

## Time of Flight




MAYO CLINIC

## Mass Spectrum Generated Compared with Library (Database)



## QUESTION #1

**Which of the following will not grow on sheep blood, chocolate and/or MacConkey agar?**

A. ***Granulicatella adiacens***
B. ***Bordetella pertussis***
C. ***Brucella melitensis***
D. ***Vibrio cholerae***
E. ***Abiotrophia defectiva***

## BACTERIA REQUIRING SPECIALIZED MEDIA

- *Bordetella pertussis*
- *Brucella* species (+/-)
- *Burkholdheria pseudomallei* (+/-)
- *Campylobacter* species
- *Francisella tularensis* (+/-)
- *Helicobacter pylori*
- *Legionella* species
- *Mycoplasma* species (+/-)
- *Ureaplasma* species

## QUESTION #2

**Which of the following bacteria may stain acid-fast positive?**

A. ***Rhodococcus* species**
B. ***Cutibacterium* species**
C. ***Finegoldia* species**
D. ***Microbacterium* species**
E. ***Wolbachia* species**

# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*

## ACID-FAST BACTERIA (MYCOLIC ACIDS)

- *Mycobacterium* species
- "Modified" acid fast stain positive
  - Weaker decolorizing agent (0.5-1% sulfuric acid in place of 3% acid-alcohol); do not stain well with Ziehl-Neelsen or Kinyoun stain
    - *Nocardia* species
    - *Rhodococcus* species
    - *Gordonia* species
    - *Tsukamurella* species
    - *Dietzia* species
- *Tatlockia* (*Legionella*) *micdadei* and some *Corynebacterium* species
  - [But not *Cutibacterium* (or *Propionibacterium*) species]

## QUESTION #3

**A laboratory technologist who has a longstanding history of diabetes mellitus inadvertently opens the lid of an agar plate growing an organism which is subsequently determined to be *Burkholdheria pseudomallei*.**

**You are asked to make a recommendation regarding postexposure prophylaxis.**

## QUESTION #3

**Which of the following would you recommend?**

**A. Trimethoprim-sulfamethoxazole**
**B. Amoxicillin**
**C. Streptomycin**
**D. Cephalexin**
**E. None**

## *Burkholderia pseudomallei* Laboratory Exposure

| | |
|---|---|
| Low risk Events | Inadvertent opening of the lid of an agar plate growing *B. pseudomallei* outside a biologic safety cabinet |
| | Inadvertent sniffing of agar plate growing *B. pseudomallei* in the absence of contact between worker and bacterium |
| | Splash event leading to visible contact of *B. pseudomallei* with gloved hand or protected body, in the absence of any evidence of aerosol |
| | Spillage of small volume of liquid culture (<1mL) within a functioning biologic safety cabinet |
| | Contamination of intact skin with culture |
| High risk Events | The presence of any predisposing condition without proper personal protective equipment (PPE): diabetes mellitus; chronic liver or kidney disease; alcohol abuse; long-term steroid use; hematologic malignancy; neutropenia or neutrophil dysfunction; chronic lung disease (including cystic fibrosis); thalassemia; any other form of immunosuppression |
| | Needlestick or other penetrating injury with implement contaminated with *B. pseudomallei* |
| | Bite or scratch by experimental animal infected with *B. pseudomallei* |
| | Splash event leading to contamination of mouth or eyes |
| | Generation of aerosol outside biologic safety cabinet (e.g., sonication, centrifuge incident) |

Peacock SJ et al. Emerg Infect Dis. 2008 Jul http://wwwnc.cdc.gov/eid/article/14/7/07-1501

## *Burkholderia pseudomallei* Postexposure Antimicrobial Drug Prophylaxis

| Antimicrobial Drug | Dosage | Frequency |
|---|---|---|
| Trimethoprim-sulfamethoxazole (TMP-SMX) | 2 × 160–800 mg (960 mg) tablets if >60 kg, 3 × 80–400 (480 mg) tablets if 40 kg-60 kg, and 1 × 160–800 mg (960 mg) or 2 × 80–400 (480 mg) tablets if adult <40 kg plus folate 5 mg/d | Every 12 h |
| Doxycycline | 2.5 mg/kg/dose up to 100 mg orally | Every 12 h |
| Amoxicillin–clavulanic acid | 20/5 mg/kg/dose. Equates to 3 × 500/125 tabs if >60 kg, and 2 × 500/125 tabs if ≤60kg | Every 8 h |

Peacock SJ et al. Emerg Infect Dis. 2008 Jul http://wwwnc.cdc.gov/eid/article/14/7/07-1501

## QUESTION #4

**Which of the following, if present in a clinical specimen, poses a hazard for laboratory personnel?**

***a. Entamoeba histolytica***
***b. Trichuris trichiura***
***c. Enterobius vermicularis***
***d. Strongyloides stercoralis***
***e. Babesia microti***

# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*

## *Strongyloides stercoralis*

- Larvae - two forms
  1. Rhabditiform (in stool)
  2. Filariform

  Infectious stage that develops in soil and occasionally in patient (leads to autoinfection and is hazardous to laboratory personnel)
- Larvae detected
  - Microscopically (top) or
  - By placing feces on plate and detecting migrating larvae where they leave a trail of bacterial colonies (bottom)

## LABORATORY- ACQUIRED BACTERIAL, FUNGAL AND PARASITIC INFECTIONS (SELECTED)

- *Bacillus anthracis*
- *Brucella* species
- *Burkholdheria pseudomallei*
  - (*Burkholdheria mallei*)
- *Coxiella burnetii*
- *Coccidioides immitis/posadasii* (*Blastomyces dermatitidis*, *Histoplasma capsulatum*)
- Dermatophytes
- Enteric pathogens
- *Francisella tularensis*
- *Mycobacterium tuberculosis*
- *Neisseria meningitidis*
- *Salmonella enterica* subsp. *enterica* serovar Typhi
- *Staphylococcus aureus*
- *Strongyloides stercoralis*
- *Yersinia pestis*

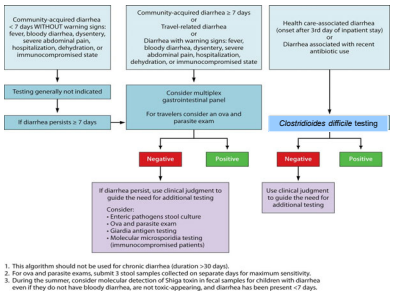
## ORGANISMS ABOUT WHICH THE LABORATORY SHOULD BE NOTIFIED IF SUSPECTED

- Avian influenza
- *Bacillus anthracis*
- *Brucella* species
- *Burkholdheria pseudomallei*
- *Burkholdheria mallei*
- *Clostridium botulinum*
- *Coxiella burnetii*
- *Coccidioides immitis/posadasii*
- Hemorrhagic fever viruses (e.g., Eboia, Marburg, Chapare, Crimean-Congo, Guanarito, Hanta, Junin, Kayasnur Forest Disease, Lassa fever, Lujo, Machupo, Omsk Hemorrhagic Fever, Sabia)
- *Francisella tularensis*
- Measles
- MERS, SARS-CoV
- Nipah virus, Hendra virus
- Smallpox
- *Yersinia pestis*

## FDA-APPROVED/CLEARED MULTIPLEX PANELS FOR GASTROINTESTINAL PATHOGENS IN STOOL

| | Verigene EP | Luminex GPP | BioFire GIP |
|---|---|---|---|
| Number of targets | 8 | 14 | 22 |
| *Campylobacter* species | ✓ | ✓ | ✓ |
| *Salmonella* species | ✓ | ✓ | ✓ |
| *Shigella* species/EnteroInvasive *E. coli* | ✓ | ✓ | ✓ |
| *Vibrio* species | ✓ | ✓ | ✓ |
| *Yersinia enterocolitica* | ✓ | | ✓ |
| *Escherichia coli* 0157 | | ✓ | ✓ |
| Enterotoxigenic *E. coli* | | ✓ | ✓ |
| Enteropathogenic *E. coli* | | | ✓ |
| Enteroaggregative *E. coli* | | | ✓ |
| *Plesiomonas shigelloides* | | | ✓ |
| Shiga toxin-producing *E. coli* | ✓ | ✓ | ✓ |
| *Clostridioides difficile* | | ✓ | ✓ |
| Norovirus GI/GII | ✓ | ✓ | ✓ |
| Rotavirus A | ✓ | ✓ | ✓ |
| Astrovirus | | | ✓ |
| Adenovirus 40/41 | | ✓ | ✓ |
| Sapovirus | | | ✓ |
| *Cryptosporidium* species | | ✓ | ✓ |
| *Entamoeba histolytica* | | ✓ | ✓ |
| *Giardia lamblia* | | ✓ | ✓ |
| *Cyclospora cayetanensis* | | | ✓ |

## TESTING ALGORITHM FOR ACUTE GASTROENTERITIS




1. This algorithm should not be used for chronic diarrhea (duration >30 days).
2. For ova and parasite exams, submit 3 stool samples collected on separate days for maximum sensitivity.
3. During the summer, consider molecular detection of Shiga toxin in fecal samples for children with diarrhea even if they do not have bloody diarrhea, are not toxic-appearing, and diarrhea has been present <7 days.

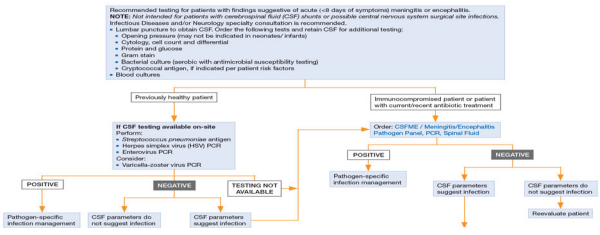
## BIOFIRE FILMARRAY MENINGITIS/ENCEPHALITIS PANEL

| Viruses | Bacteria | Fungi |
|---|---|---|
| Cytomegalovirus | *Escherichia coli* K1 | *Cryptococcus neoformans/gattii* |
| Enterovirus | *Haemophilus influenzae* | |
| Herpes simplex virus 1 | *Listeria monocytogenes* | |
| Herpes simplex virus 2 | *Neisseria meningitidis* | |
| Human herpes virus 6 | *Streptococcus agalactiae* | |
| Human parechovirus | *Streptococcus pneumoniae* | |
| Varicella zoster virus | | |

# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*

MAYO CLINIC

## Meningitis/Encephalitis Panel Algorithm




MAYO CLINIC

## FDA-Approved Multiplex Respiratory Panels

| Parameter | FilmArray | Verigene | NxTAG-RPP | eSensor RVP | ePlex |
|---|---|---|---|---|---|
| Analysis platform | FilmArray system or FilmArrayTorch | Verigene system | Luminex MAGPIX | eSensor | ePlex system |
| Acceptable specimens | NP swab | NP swab | NP swab | NP swab | NP swab |
| Number of targets | 20 | 16 | 20 | 14 | 18 |
| **Pathogens** | | | | | |
| **Viruses** | | | | | |
| Adenovirus | ✓ | ✓ | ✓ | ✓(differentiates B/E from C) | ✓ |
| Coronavirus (229E, HKU1, NL63, OC43) | | | | | ✓ |
| Coronavirus HKU1 | ✓ | | ✓ | | |
| Coronavirus NL63 | ✓ | | ✓ | | |
| Coronavirus 229E | ✓ | | ✓ | | |
| Coronavirus OC43 | ✓ | | ✓ | | |
| SARS-CoV-2 | ✓ | | | | ✓ |
| Human bocavirus | | | ✓ | | |
| Human metapneumovirus | ✓ | ✓ | ✓ | ✓ | ✓ |
| Influenza A | ✓ | ✓ | ✓ | ✓ | ✓ |
| Subtype H1 | ✓ | ✓ | ✓ | ✓ | ✓ |
| Subtype H3 | ✓ | ✓ | ✓ | ✓ | ✓ |
| Subtype 2009 H1N1 | ✓ | | | ✓ | ✓ |
| Influenza B | ✓ | ✓ | ✓ | ✓ | ✓ |
| Parainfluenza 1 | ✓ | ✓ | ✓ | ✓ | ✓ |
| Parainfluenza 2 | ✓ | ✓ | ✓ | ✓ | ✓ |
| Parainfluenza 3 | ✓ | ✓ | ✓ | ✓ | ✓ |
| Parainfluenza 4 | ✓ | ✓ | ✓ | | ✓ |
| Respiratory syncytial virus | ✓ | | | | |
| Respiratory syncytial virus A | | ✓ | ✓ | ✓ | ✓ |
| Respiratory syncytial virus B | | ✓ | ✓ | ✓ | ✓ |
| Rhinovirus/enterovirus | ✓ | ✓ | ✓ | ✓ | ✓ |
| **Bacteria** | | | | | |
| *Chlamydophila pneumoniae* | ✓ | | ✓ | | ✓ |
| *Mycoplasma pneumoniae* | ✓ | | ✓ | | ✓ |
| *Bordetella pertussis* | ✓ | ✓ | | | |
| *Bordetella parapertussis* | ✓ | | | | |
| *B. parapertussis/Bordetella bronchiseptica* | | ✓ | | | |
| *Bordetella holmesii* | | ✓ | | | |
| **Throughput** | Scalable | Low | High | Medium | Scalable |
| **Time to result** | 1 hour | ~2.3 hours | ~4 hours | ~6 hours | ~1.5 hours |

Ramanan et al. Clin Microbiol Rev 2018;31:1-28

MAYO CLINIC

## Lower Respiratory Tract Panels

| | Curetis Unyvero | BioFire |
|---|---|---|
| **Bacteria** | | |
| *Acinetobacter* spp. | ✓ | |
| *Acinetobacter calcoaceticus-baumannii* complex | | ✓ |
| *Chlamydia pneumoniae* | ✓ | ✓ |
| *Citrobacter freundii* | ✓ | |
| *Enterobacter aerogenes* | | ✓ |
| *Enterobacter cloacae* | | ✓ |
| *Enterobacter cloacae* complex | ✓ | |
| *Escherichia coli* | ✓ | ✓ |
| *Haemophilus influenzae* | ✓ | ✓ |
| *Klebsiella oxytoca* | ✓ | ✓ |
| *Klebsiella pneumoniae* | ✓ | |
| *Klebsiella pneumoniae* group | | ✓ |
| *Klebsiella variicola* | ✓ | |
| *Legionella pneumophila* | ✓ | ✓ |
| *Moraxella catarrhalis* | ✓ | ✓ |
| *Morganella morganii* | ✓ | |
| *Mycoplasma pneumoniae* | ✓ | ✓ |
| *Proteus* spp. | ✓ | ✓ |
| *Pseudomonas aeruginosa* | ✓ | ✓ |
| *Serratia marcescens* | ✓ | ✓ |
| *Staphylococcus aureus* | ✓ | ✓ |
| *Stenotrophomonas maltophilia* | ✓ | |
| *Streptococcus agalactiae* | | ✓ |
| *Streptococcus pneumoniae* | ✓ | ✓ |
| *Streptococcus pyogenes* | | ✓ |

| | Curetis Unyvero | BioFire |
|---|---|---|
| **Viruses** | | |
| Influenza A | | ✓ |
| Influenza B | | ✓ |
| Respiratory Syncytial Virus | | ✓ |
| Human Rhinovirus/Enterovirus | | ✓ |
| Human Metapneumovirus | | ✓ |
| Parainfluenza virus | | ✓ |
| Adenovirus | | ✓ |
| Coronavirus (non-SARS-CoV) | | ✓ |
| SARS-CoV-2 | | ✓ |
| **Resistance genes** | | |
| $bla_{KPC}$ | ✓ | ✓ |
| $bla_{NDM}$ | ✓ | ✓ |
| $bla_{IMP}$ | | ✓ |
| $bla_{OXA-23}$ | ✓ | |
| $bla_{OXA-48}$ | ✓ | |
| $bla_{OXA58}$ | ✓ | |
| $bla_{OXA58-like}$ | | ✓ |
| $bla_{VIM}$ | ✓ | ✓ |
| $bla_{CTX-M}$ | ✓ | ✓ |
| $bla_{TEM}$ | ✓ | |
| *mecA* | ✓ | |
| *mecA/C* and *MREJ* | | ✓ |

## QUESTION #5

- You are asked to see a 62 year old man with a positive blood culture to advise on management.
- Gram stain of the positive blood culture bottle shows Gram positive cocci in clusters.
- A rapid PCR panel performed on the positive blood culture bottle contents detects *Staphylococcus aureus*, *Staphylococcus epidermidis* as well as *mecA/C* but not *mecA/C* and MREJ.

## QUESTION #5

Which of the following is the interpretation of this finding?

A. Methicillin-susceptible *S. aureus* and methicillin-resistant *S. epidermidis*
B. Methicillin-susceptible *S. aureus* and methicillin-susceptible *S. epidermidis*
C. Methicillin-resistant *S. aureus* and methicillin-resistant *S. epidermidis*
D. Methicillin-resistant *S. aureus* and methicillin-susceptible *S. epidermidis*

MAYO CLINIC

## FDA-Approved Multiplex Panels for Detection of Select Organisms and Resistance Genes in Positive Blood Cultures

# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*

MAYO CLINIC

## FDA-Approved Multiplex Panels for Detection of Select Organisms and Resistance Genes in Positive Blood Cultures, continued

| | | FilmArray BCID2 | VERIGENE® Gram-Positive Blood Culture Test | VERIGENE® Gram-Negative Blood Culture Test | GenMark Dx ePlex BCID-GP Panel | GenMark Dx ePlex BCID-FP Panel | GenMark Dx ePlex BCID-GN Panel |
|---|---|---|---|---|---|---|---|
| Yeasts | *Candida albicans* | ✓ | | | | ✓ | |
| | *Candida auris* | ✓ | | | | ✓ | |
| | *Candida dubliniensis* | | | | | ✓ | |
| | *Candida famata* | | | | | ✓ | |
| | *Candida glabrata* | ✓ | | | | ✓ | |
| | *Candida guilliermondii* | | | | | ✓ | |
| | *Candida kefyr* | | | | | ✓ | |
| | *Candida krusei* | ✓ | | | | ✓ | |
| | *Candida lusitaniae* | | | | | ✓ | |
| | *Candida parapsilosis* | ✓ | | | | ✓ | |
| | *Candida tropicalis* | ✓ | | | | ✓ | |
| | *Cryptococcus gattii* | | | | | ✓ | |
| | *Cryptococcus neoformans* | | | | | ✓ | |
| | *C. neoformans/gattii* | ✓ | | | | | |
| | *Fusarium species* | | | | | ✓ | |
| | *Rhodotorula species* | | | | | ✓ | |
| | Pan *Candida* | | | | ✓ | | ✓ |
| Resistance genes | *mecA* | | ✓ | | ✓ | | |
| | *mecC* | | | | ✓ | | |
| | *mecA/C* | ✓ | | | | | |
| | *mecA/C* and MREJ | ✓ | | | | | |
| | *vanA* | ✓ | ✓ | | ✓ | | |
| | *vanB* | ✓ | ✓ | | ✓ | | |
| | $bla_{KPC}$ | ✓ | | ✓ | | | ✓ |
| | $bla_{NDM}$ | ✓ | | ✓ | | | ✓ |
| | $bla_{VIM}$ | ✓ | | ✓ | | | ✓ |
| | $bla_{IMP}$ | ✓ | | ✓ | | | ✓ |
| | $bla_{OXA-48}$ | ✓ | | ✓ | | | ✓ |
| | *mcr-1* | ✓ | | | | | |

## STAPHYLOCOCCI
## METHICILLIN RESISTANCE

- Methicilllin resistance mediated by *mecA* (or rarely *mecC*) gene products
  - Penicillin binding protein (PBP) target altered (PBP2a)
    - Confers resistance to all available β-lactams (except ceftaroline)
    - Even if staphylococci that are methicillin-resistant *appear* susceptible to these other β-lactams, they are not effective
- Oxacillin or cefoxitin tested
- *mecA/C* and MREJ specific for *Staphylococcus aureus*
- For serious infections, susceptibility to oxacillin confirmed using PBP2a testing or nucleic acid amplification test (NAAT) to detect *mecA* (and *mecC*)

MAYO CLINIC

## T2Direct Diagnostics
## Direct from Blood

- **Multiplex PCR and T2 magnetic resonance, average turnaround time 4.3 hours**
- **T2Candida Panel**
  - ***Candida albicans***
  - ***Candida tropicalis***
  - ***Candida krusei***
  - ***Candida glabrata***
  - ***Candida parapsilosis***
- **T2Bacteria Panel**
  - ***Enterococcus faecium***
  - ***Staphylococcus aureus***
  - ***Klebsiella pneumoniae***
  - ***Pseudomonas aeruginosa***
  - ***Escherichia coli***

## QUESTION #6

- **A 52 year old woman receives a liver transplant (CMV $D^+/R^-$) at your medical center.**
- **Seven months later (after she has completed a course of valganciclovir), she develops fever and diarrhea and is found to have a CMV viral load of 20,000 IU/ml.**
- **In addition to treating the patient with intravenous ganciclovir and performing a colonoscopy to assess for CMV colitis, you recommend follow-up CMV viral load testing.**

## QUESTION #6

**How often should this test be performed?**

**A. Daily**
**B. Twice a week**
**C. Weekly**
**D. Every two weeks**
**E. Monthly**

## OPTIMAL FREQUENCY CMV VIRAL LOAD TESTING

- **Weekly viral load testing sufficient to document antiviral response, antiviral resistance emergence**
  - **$T_{1/2}$ virus ~5-8 days**
  - **May rise 1st few days on therapy**
  - **Obtain baseline viral load day therapy started**
- **Treatment**
  - **Until viral clearance, symptom resolution and 2 week minimum**
- **Changes >3-fold (>0.5 log)**
  - **Biologically important changes in viral replication**
- **Preemptive treatment → weekly viral load testing**

# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*

## QUESTION #7

**You are consulted to advise on the course of action for a 57 year old female liver transplant recipient (transplant for alcoholic steatohepatitis; CMV $D^+/R^+$) who has a whole blood HHV-6 viral load of $3.6x10^6$ copies/ml at three months post-transplant. The test was performed because of a report of subjective fever of four days' duration. She has no other new symptoms. The patient received one month of acyclovir prophylaxis post-transplant and is currently receiving mycophenolate mofetil, prednisone and trimethoprim-sulfamethoxazole. Her post-transplant course was complicated by one episode of treated rejection on day 30 post transplant. Physical examination is unremarkable and she is afebrile.**

## QUESTION #7

**Which of the following would you recommend?**

**A. Intravenous ganciclovir**
**B. Oral valganciclovir**
**C. Oral acyclovir**
**D. Intravenous foscarnet**
**E. No antiviral therapy is indicated**

## CHROMOSOMALLY INTEGRATED HUMAN HERPESVIRUS-6

- High HHV-6 levels in whole blood
  - (>5.5 $\log_{10}$ copies/ml)
  - Suggest chromosomally integrated HHV-6
- 1:1 ratio of viral to human genomes

Pellett et al. Rev Med Virol 2012;22:144-55

## QUESTION #8

A 65 year old man has multiple blood cultures positive for *Pseudomonas aeruginosa* resistant to amikacin, gentamicin, tobramycin, aztreonam, cefepime, ceftazidime, meropenem, piperacillin-tazobactam, ciprofloxacin, and levofloxacin. You call the clinical microbiology laboratory to request susceptibility testing of an additional antimicrobial.

Which of the following is most appropriate?

A. Dalbavancin
B. Tedizolid
C. Ceftolozane/tazobactam
D. Oritavancin

## QUESTION #9

You are asked to see a 43 year old woman to advise on management of a positive blood culture.

- Gram stain of her blood culture bottle shows Gram-negative bacilli.
- A rapid PCR panel performed on the positive blood culture bottle contents detects *Enterobacteriaceae* and $bla_{KPC}$.

## QUESTION #9

**The $bla_{KPC}$ gene product would be expected to confer resistance to which of the following?**

**A. Cefepime**
**B. Plazomicin**
**C. Colistin**
**D. Ceftazidime/avibactam**

# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*

## TYPICAL SUSCEPTIBILITY OF A KPC-PRODUCER

***Klebsiella pneumoniae* carbapenemase producer**

| | | | | | |
|---|---|---|---|---|---|
| Ampicillin | >16 R | Ampicillin/Sulbactam | >16/8 R | Piperacillin/Tazobactam | 64/4 R |
| Cefazolin | >16 R | Oral cephalosporins | R | Cefepime | >16 R |
| Ceftazidime | >16 R | Ceftriaxone | >32 R | Ertapenem | >1 R |
| Meropenem | >8 R | Aztreonam | >16 R | Ciprofloxacin | >2 R |
| Levofloxacin | 4 I | Amikacin | >32 R | Gentamicin | >8 R |
| Tobramycin | 4 S | Tigecycline | 2 S | TMP/SMX | >2/38 R |

## TYPICAL SUSCEPTIBILITY OF AN ESBL-PRODUCER

***Escherichia coli***

– Extended spectrum beta-lactamase producer

| | | | | | |
|---|---|---|---|---|---|
| Ampicillin | >16 R | Ampicillin/Sulbactam | >16/8 R | Piperacillin/Tazobactam | 16/4 S |
| Cefazolin | >16 R | Oral cephalosporins | R | Cefepime | >16 R |
| Ceftazidime | >16 R | Ceftriaxone | >32 R | Ertapenem | ≤0.5 S |
| Meropenem | ≤1 S | Aztreonam | >16 R | Ciprofloxacin | ≤1 S |
| Levofloxacin | ≤2 S | Amikacin | ≤8 S | Gentamicin | ≤1 S |
| Tobramycin | 4 S | Tigecycline | 2 S | TMP/SMX | >2/38 R |

## QUESTION #10

Which of the following susceptibility patterns would be typical for an *Escherichia coli* isolate carrying a New Delhi metallo-β-lactamase (NDM)?

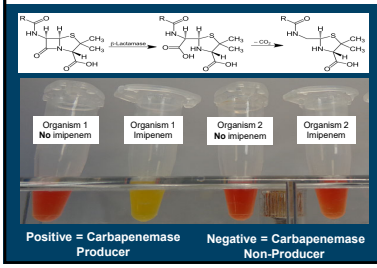
| | Cefazolin | Cefotaxime | Ceftazidime | Piperacillin/ tazobactam | Imipenem | Aztreonam |
|---|---|---|---|---|---|---|
| a) | R | S | S | S | S | S |
| b) | R | R | R | S | S | R |
| c) | R | R | R | R | S | R |
| d) | R | R | R | R | R | R |

## QUESTION #11

Which of the following tests for carbapenemase production?

A. PBP2a test
B. D-test
C. Carba NP test
D. Polymerase chain reaction assay

## CARBAPENEMASE PRODUCTION TEST
### Carba NP TEST

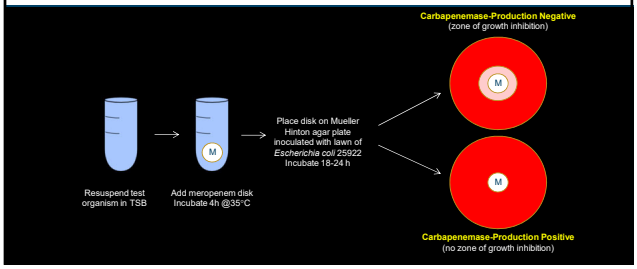



Positive = Carbapenemase Producer

Negative = Carbapenemase Non-Producer

- β-lactam ring hydrolyzed by carbapenemase
- ↓pH (detected by indicator dye color change red → yellow)
- Rapid (2 hours)

## CARBAPENEMASE PRODUCTION TEST
## MODIFIED CARBAPENEM INACTIVATION

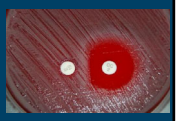


# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*

## QUESTION #12

The image shows *Staphylococcus aureus* grown with an erythromycin disc (left) and a clindamycin disc (right).

Which of the following is the correct interpretation of these results?



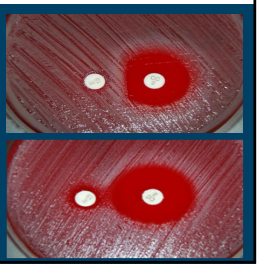
A. **Erythromycin susceptibility, inducible clindamycin resistance**
B. **Erythromycin resistance, constitutive clindamycin resistance**
C. **Erythromycin resistance, inducible clindamycin resistance**
D. **Erythromycin susceptibility, constitutive clindamycin resistance**

## INDUCIBLE CLINDAMYCIN RESISTANCE (D-TEST)

- Macrolide resistance from alteration in ribosomal target → co-resistance to clindamycin; constitutive or inducible
  - Constitutive, erythromycin & clindamycin test resistant
  - Inducible, erythromycin tests resistant but clindamycin tests falsely susceptible
- (Macrolide resistance due to efflux → no effect on clindamycin)

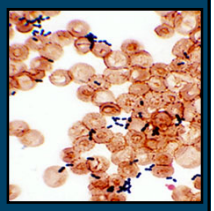
## INDUCIBLE CLINDAMYCIN RESISTANCE (D-TEST)

- Erythromycin & clindamycin disks incubated on plate
  - Flattening of zone of inhibited growth between disks = inducible clindamycin resistance (top)
  - If erythromycin does not influence zone around clindamycin disk, clindamycin susceptible (bottom)



## QUESTION #13

- **You are asked to see a 95 year old woman who is a resident of a long-term care facility to advise on therapy for bacteremia associated with a urinary tract infection.**
- **She has had two sets of blood cultures collected, both of which signaled positive after 17 hours of incubation.**
- **Gram stain of the bottles is shown.**
- **A rapid PCR panel performed on the positive blood culture bottle detects *Enterococcus* species as well as *vanA/vanB*.**



## QUESTION #13

**Which of the following is the most likely identity of the blood culture isolate?**

A. ***Enterococcus gallinarum***
B. ***Enterococcus faecium***
C. ***Enterococcus faecalis***
D. ***Enterococcus casseliflavus***
E. ***Enterococcus avium***

## ENTEROCOCCI
### VANCOMYCIN SUSCEPTIBILITY TESTING

- **Vancomycin MICs ≥32 µg/ml**
  - **Typically VanA or VanB mediated resistance**
  - **Typically *E. faecium***
  - **Epidemiologically significant**
- **Vancomycin MICs, 8-16 µg/ml (intermediate)**
  - **VanC**
  - ***E. gallinarum* or *E. casseliflavus/flavescens***
  - **Not epidemiologically significant**

# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*

## QUESTION #14

A 44 year old man who underwent bilateral lung transplantation for pulmonary hypertension develops a sternal wound infection with sternal dehiscence 15 days post-transplant.
Blood cultures are negative. He undergoes sternal debridement with the finding of purulence and negative Gram and KOH stains.
After three days of incubation, pinpoint, clear colonies are visualized on cultures on sheep blood agar, however Gram stain of these colonies is negative.

## QUESTION #14

Which of the following is the most appropriate empiric antibiotic to treat this patient?

a) Cefepime
b) Ceftriaxone
c) Trimethoprim-sulfamethoxazole
d) Azithromycin
e) Doxycycline

## *Mycoplasma hominis*

- Post-cardiothoracic transplant
  - Pleuritis, surgical site infection and/or mediastinitis
- Treatment
  - Inactive
    - Cell wall active antibiotics
    - Trimethoprim/sulfamethoxazole
    - Aminoglycosides
    - Erythromycin and azithromycin
  - Active
    - Tetracyclines (doxycycline preferred)
    - Fluoroquinolones
    - Clindamycin

Sampath, R., et al. EBioMedicine (2017), http://dx.doi.org/10.1016/j.ebiom.2017.04.026

## QUESTION #15

A transplant hepatologist calls to inquire about ganciclovir resistance testing on a liver transplant patient with CMV colitis and the following CMV viral loads:

7/01/16: 26,000 IU/ml (day of diagnosis)
7/11/16: 25,000 IU/ml
7/20/16: 22,000 IU/ml
7/31/16: 27,000 IU/ml

- The patient is CMV $D^+/R^-$, received 3 months of valganciclovir prophylaxis, and now has CMV disease after discontinuing valganciclovir.
- He has been receiving full dose intravenous ganciclovir since July 1st and his diarrhea is unchanged.

## QUESTION #15

A plasma test for mutations in which of the following genes is most appropriate?

A. UL51
B. UL54
C. UL89
D. UL97
E. Testing is unlikely to be helpful given the patient's viral load

## QUESTION #16

Results of testing show a M460V UL97 mutation. This mutation would be expected to confer resistance to:

A. Cidofovir
B. Foscarnet
C. Ganciclovir
D. Ganciclovir and foscarnet
E. Ganciclovir and cidofovir

# 04 – Core Concepts: Microbiology
# 05 – Microbiology: What You Need to Know for The Exam
*Speaker: Robin Patel, MD*