



June 11, 2019 noon

Agenda

- Clinical Microbiology Lab in AS
- Case Discussions
- Open Discussion



Clinical Microbiology Lab in AS

- blood cx contamination
- gram stain
- susceptibility testing

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Does my patient have an infection?

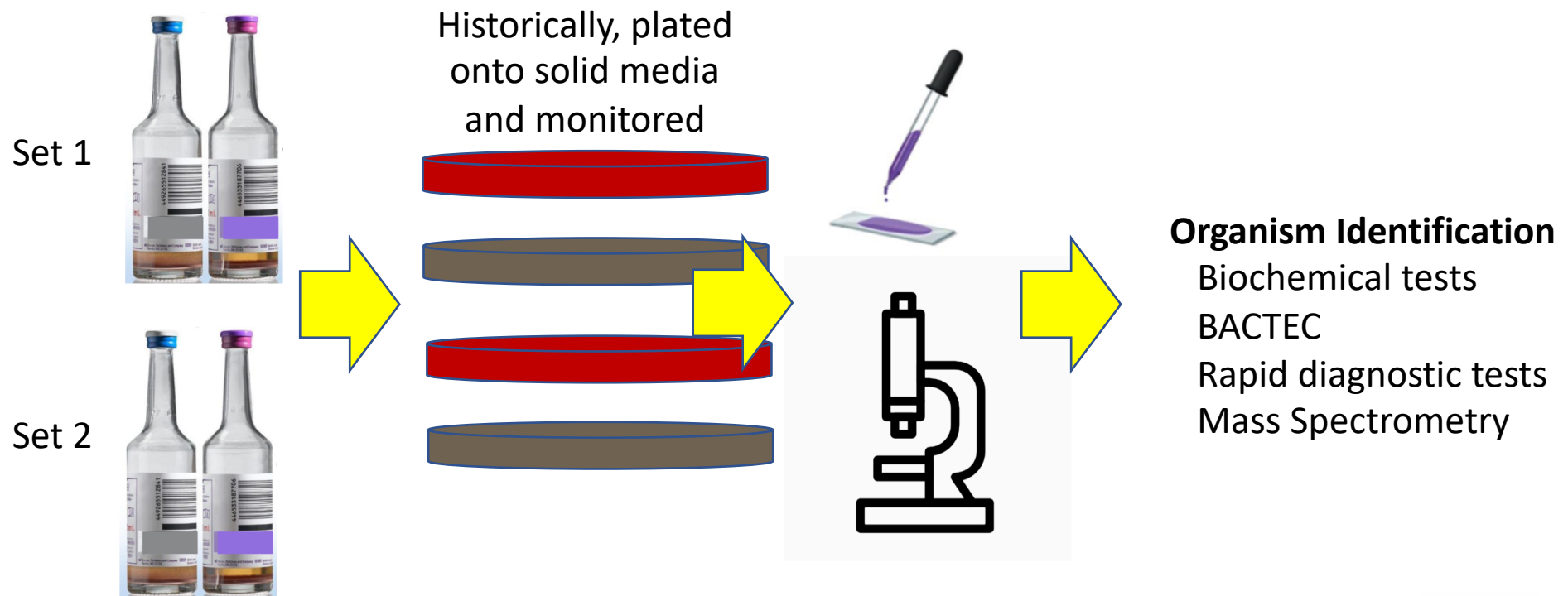
- Is my patient's illness caused by a microbe?
- If so, what is it?
- What is the susceptibility profile of the organism so therapy can be targeted?



Obtaining blood cultures in suspected infection

Adults: 2-4 blood culture sets per septic episode

- 20-30mL of blood per culture set injected into 2 bottles



CASE

54 yo F presents with fevers, chills, left flank pain and hypotension. Orders:

Blood cultures x 2

UA/urine culture

Empiric ceftriaxone

On day 2 of hospitalization, she is much improved.

- Blood cultures: 1/2 sets growing GPCs in clusters
- Urine Culture: 100,000 CFU E.coli



Question

What do these GPCs represent?

A) Same E.coli as the urine

B) Skin contaminant

C) Staph aureus

D) Do I need to care?



Brief Gram Stain Interlude

GRAM-POSITIVE

GRAM-NEGATIVE



Fixation



Crystal Violet



Iodine Treatment



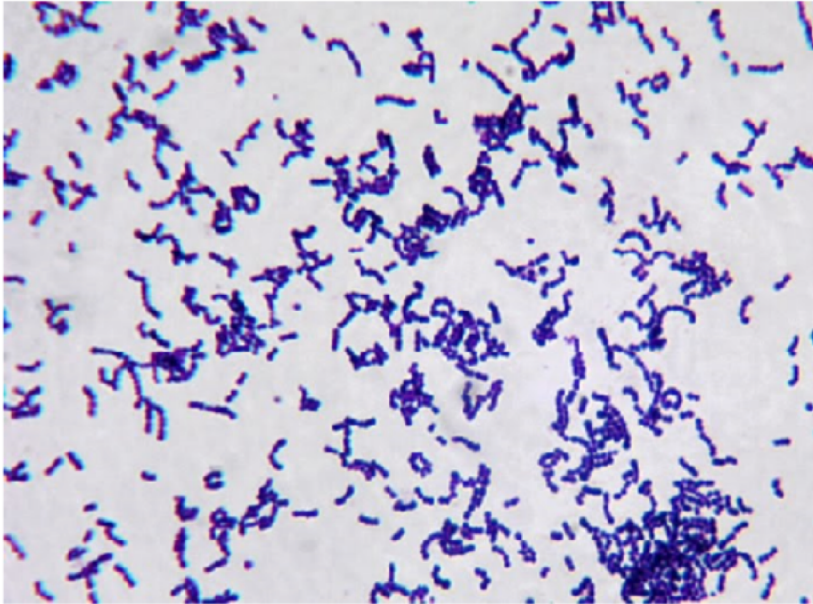
Decolorisation



Counter stain with
Safranin



Gram Stain



Gram Positive Bacteria

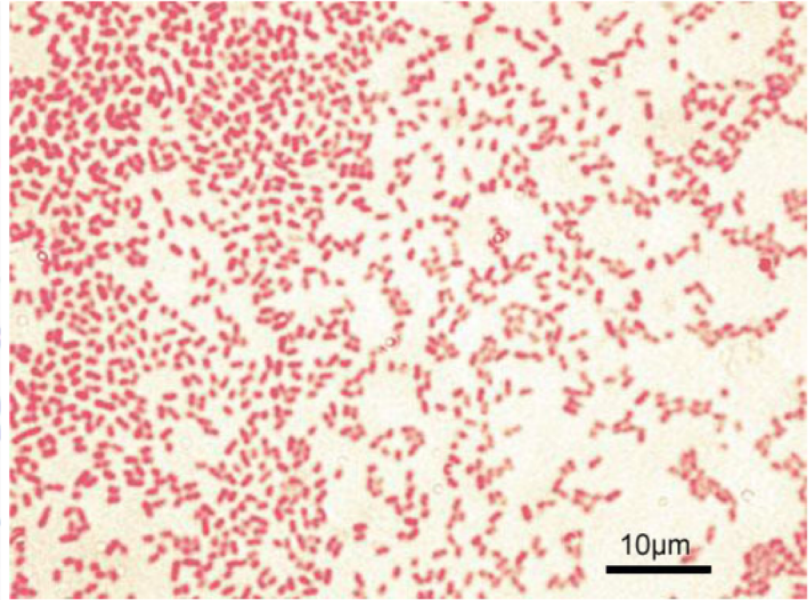
Staphylococcus

Streptococcus

Enterococcus

Clostridium

Corynebacterium, Propionibacterium, Bacillus



Gram Negative Bacteria

E. coli

Klebsiella

Enterobacter

Pseudomonas



back to our CASE

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On day 2 of hospitalization, she is much improved.

- **Blood cultures: 1/2 sets growing GPCs in clusters**
- **Urine Culture: 100,000 CFU E.coli**



Skin Contamination





























- Skin contamination of blood culture bottles
 - Very common
 - Very costly
 - Frequently confusing to clinicians
 - Common bacteria:
 - *Staphylococcus* spp (coagulase-negative staph)
 - *Streptococcus* spp
 - *Corynebacterium* spp
 - *Propionibacterium* spp
 - *Bacillus* spp
- Certain bacteria should never be considered a contaminant
 - *Staph aureus*
 - *Candida*






Contaminant vs Infection?

Common skin or environmental flora

Each set tests blood samples in an aerobic + anaerobic bottle

<u>Set 1</u>	<u>Set 2</u>	<u>Result</u>
 	 	True
 	 	True
 	 	True
 	 	Negative
 	 	Negative
 	 	True
 	 	Negative

Legend

-  Positive bottle
-  Negative bottle
-  Not tested



Susceptibility Testing



Blood culture: *E.coli*

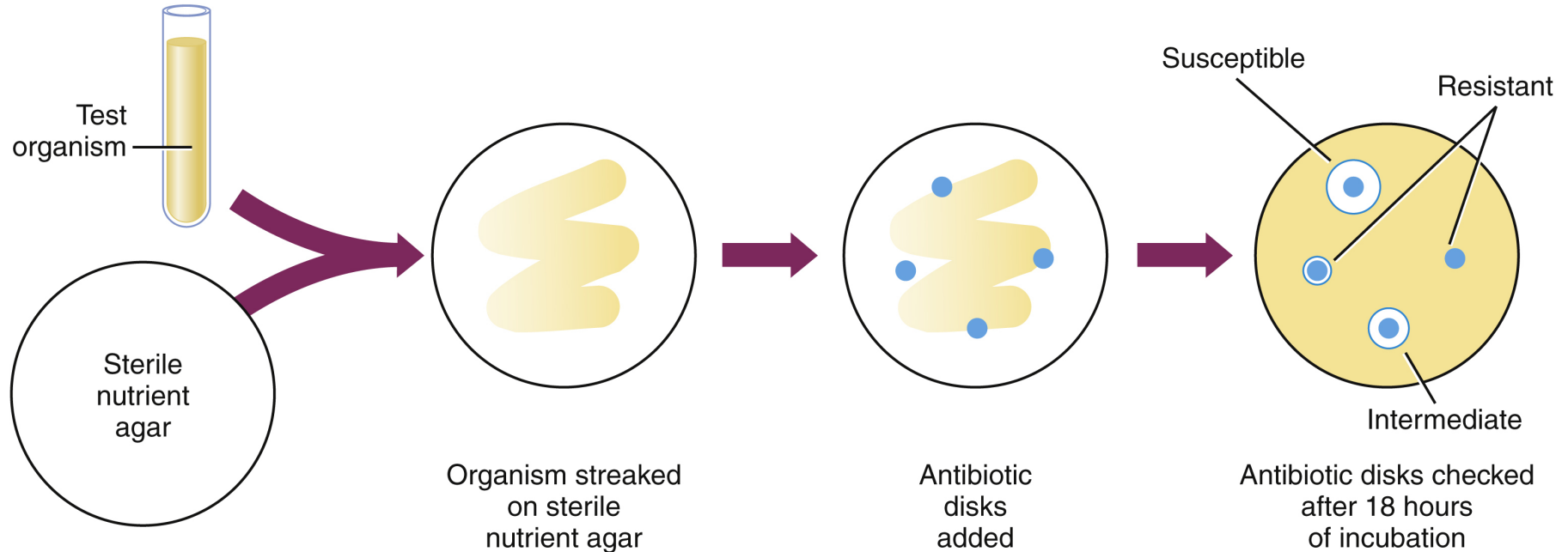
Drug	Interpretation	MIC value
Ceftriaxone	S	0.25
Cefazolin	S	0.5
Levofloxacin	R	8
Ertapenem	S	0.25
Gentamicin	S	2.0
Tobramycin	S	0.5
Piperacillin/tazobactam	S	4.0
Meropenem	S	0.5

- ✓ Use Interpretation column first to determine which antibiotic will be appropriate.
- ✓ If you don't see the antibiotic, don't assume susceptibility!



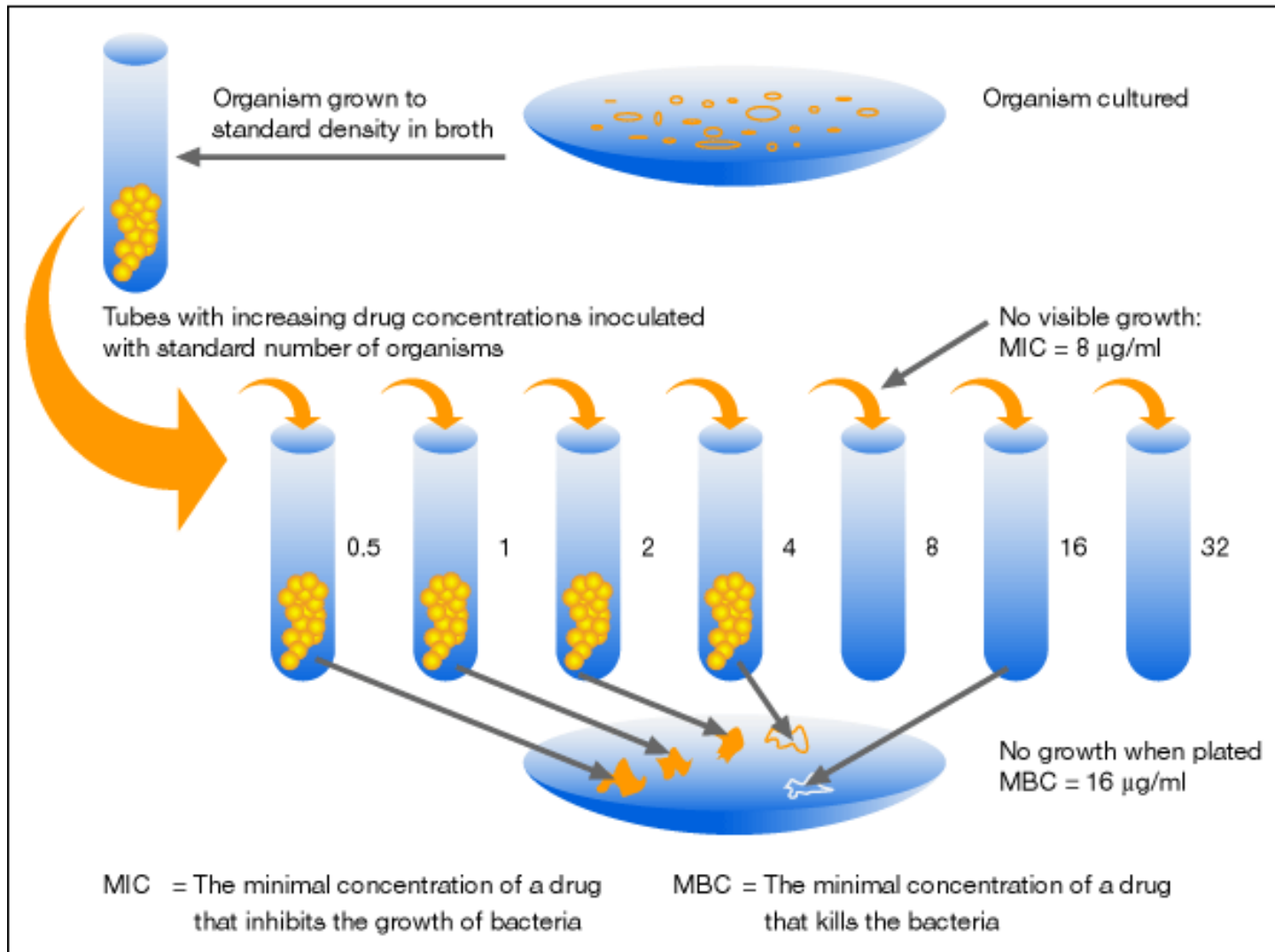
Disk Diffusion (Qualitative)

- “Kirby-Bauer-Turck” method.

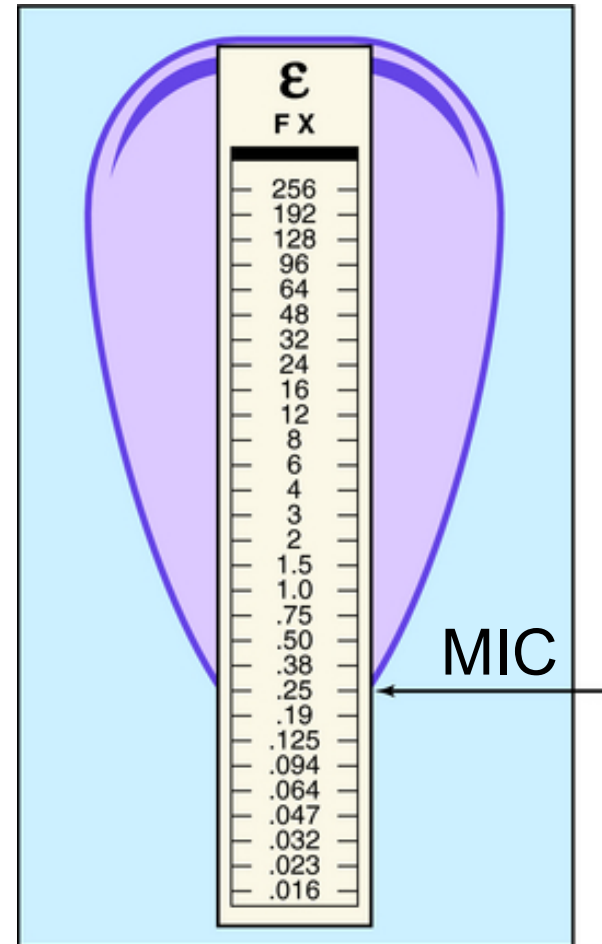


Broth Dilution (Quantitative)

Determination of MIC (here: broth ditution test)



E-test (Quantitative)



Susceptibility Testing

- MIC = Minimum Inhibitory Concentration
 - ✓ A lower MIC means smaller amounts of drug are necessary to inhibit growth
 - ✓ But... Lowest MIC does not necessarily mean that is the most effective drug *in vivo*
 - ✓ PK/PD properties of drug?
 - ✓ Drug toxicity... interactions... collateral damage... cost?



Summary

- Blood culture contamination
 - Lab QI
 - Unnecessary abx
- Susceptibility testing – MIC more to come
- Make friends with the Lab!

