



Basic Techniques in Microbiology

Dr. Aruna Poojary MD,DNB,D(ABMM) ,Dip (HIC),CIC

Consultant Microbiologist & HOD

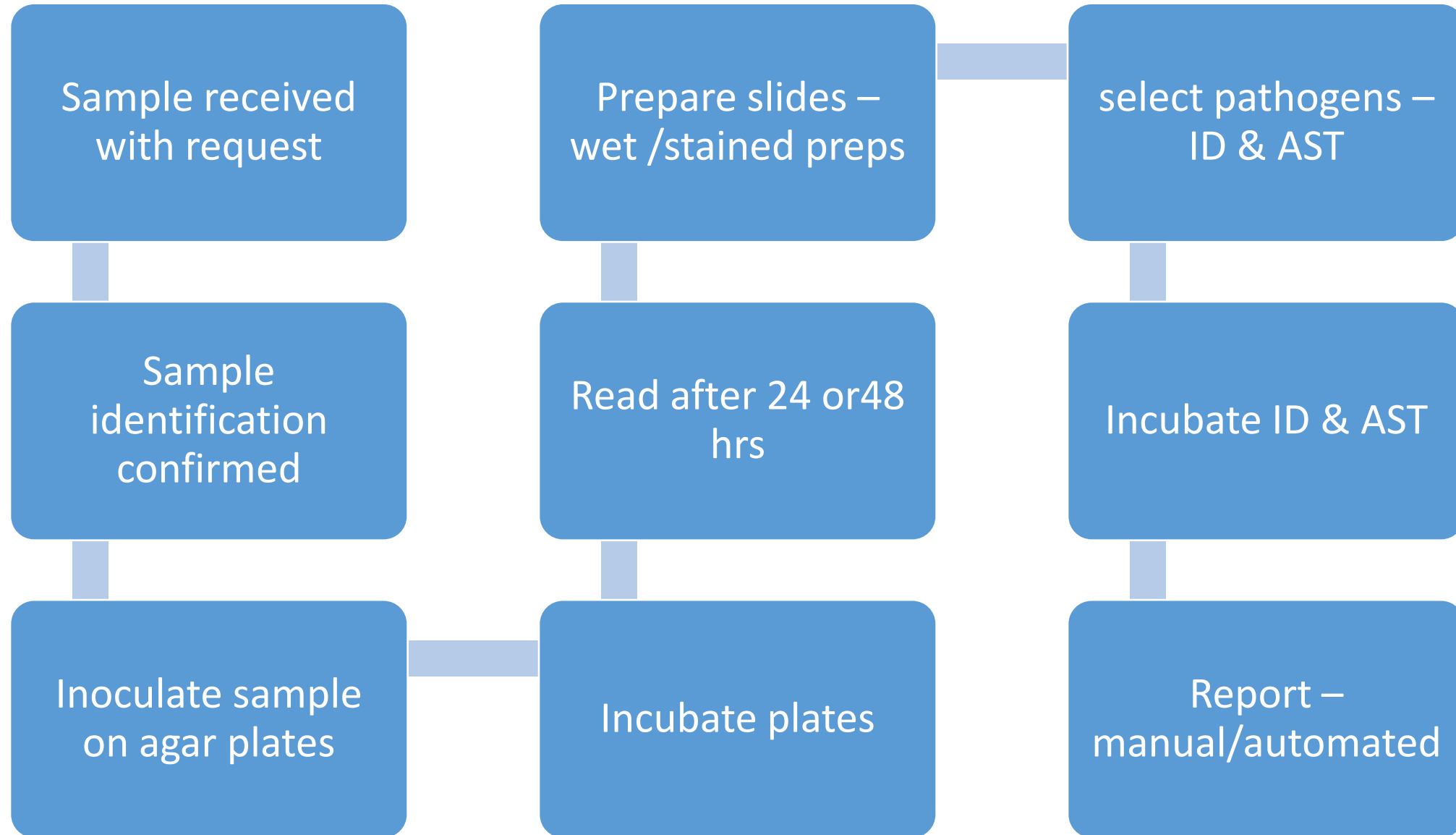
Dept. of Pathology & Microbiology

Breach Candy Hospital Trust

&

Freelance Infection Control Advisor

Workflow in a Micro lab - Culture



Biosafety precautions

- Use of appropriate PPE
- Use of appropriate biosafety devices
- No mouth pipetting
- Use of appropriate disinfectants
- Following biomedical waste practices



Basic procedures

- Labelling
- Inoculation : solid media /liquid media
- Preparing smears
- Staining
- Putting biochemical for identification
- Drug susceptibility : manual /automated
- Taking DST readings
- Agglutination tests

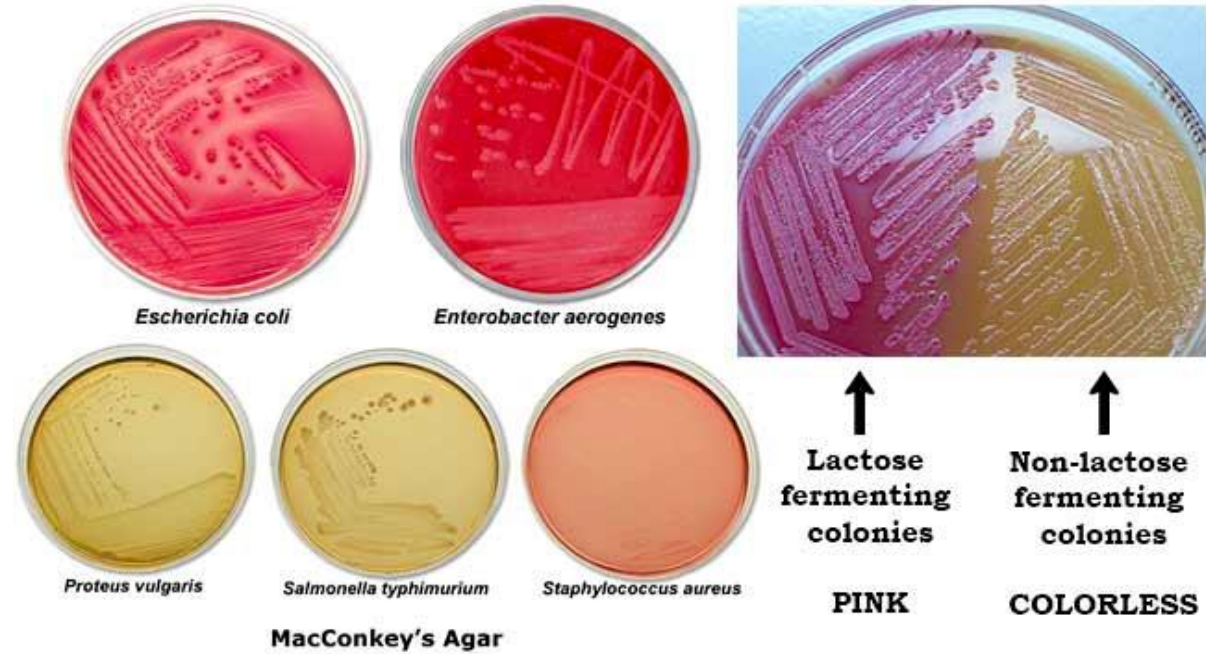
Labelling

- Barcode / name of pt
- Sample
- LIS nos
- date

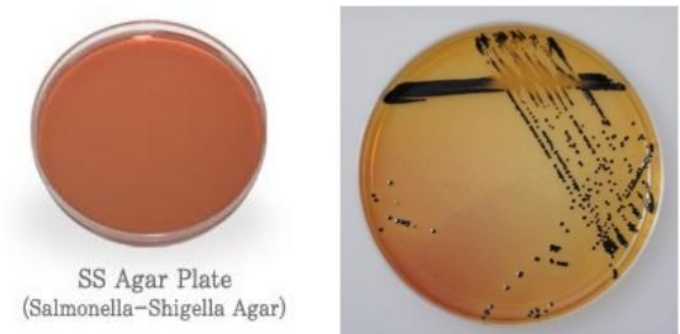


Inoculation : solid media /liquid media

- Choice of agar



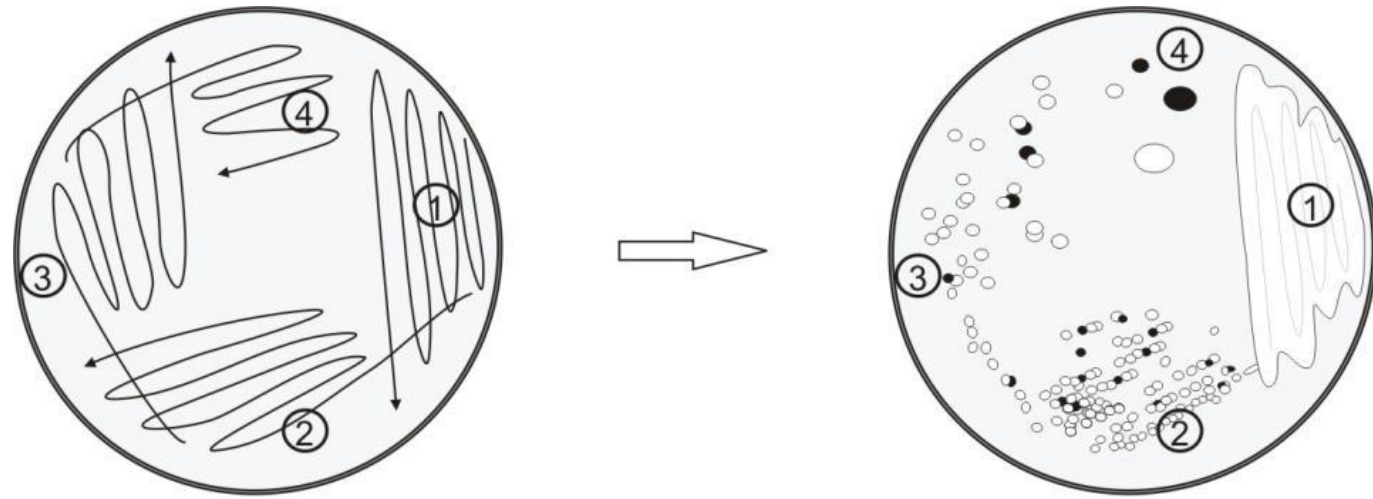
Salmonella Shigella agar



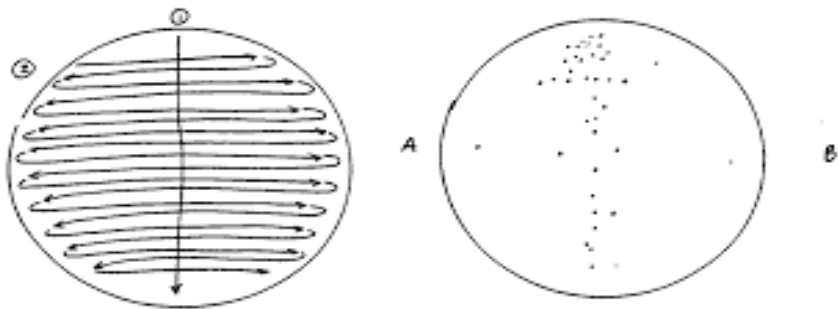
- For isolation and differential medium for pathogenic Gram-negative bacilli in particular, Salmonella and Shigella. Inhibitor for Coliforms.



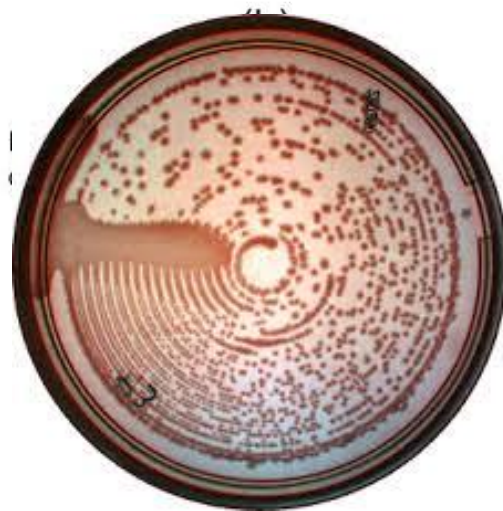
Methods of streaking



(a)



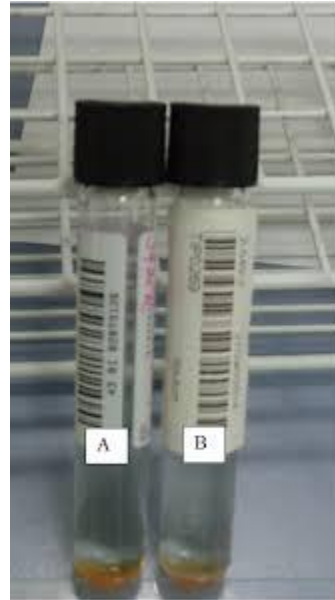
Plating a urine specimen. "A" shows the streaking pattern. "B" shows the growth after incubation.



Inoculating liquid media – do's & don't's



Blood culture sets



MGIT tubes

Liquids like peptone

Incubation

- 25°C- 56°C
- Special environment – ambient, carbon dioxide, room temp



Anaerobic Jar

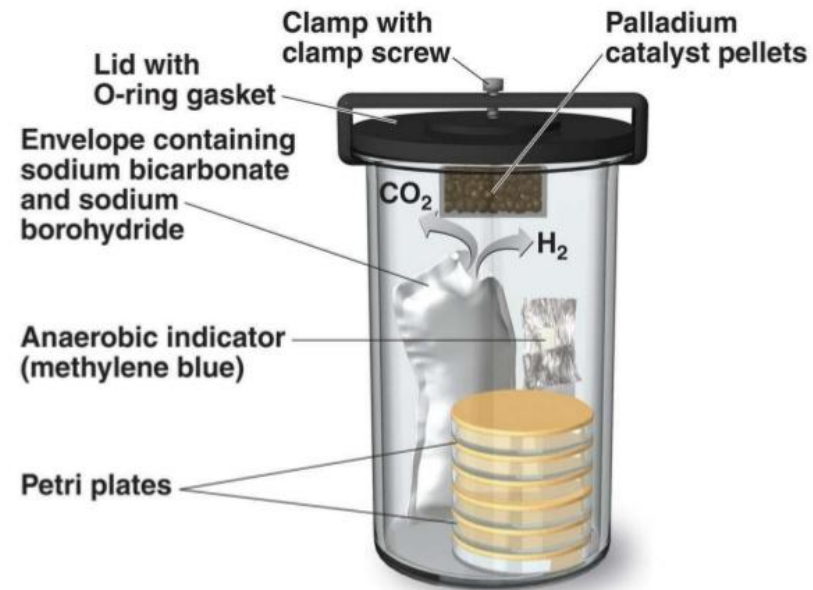


Figure 6.6

Preparing smears

- Size
- Fixation
- Drying
- Staining – Gram , Zeihl Neelson, Giemsa

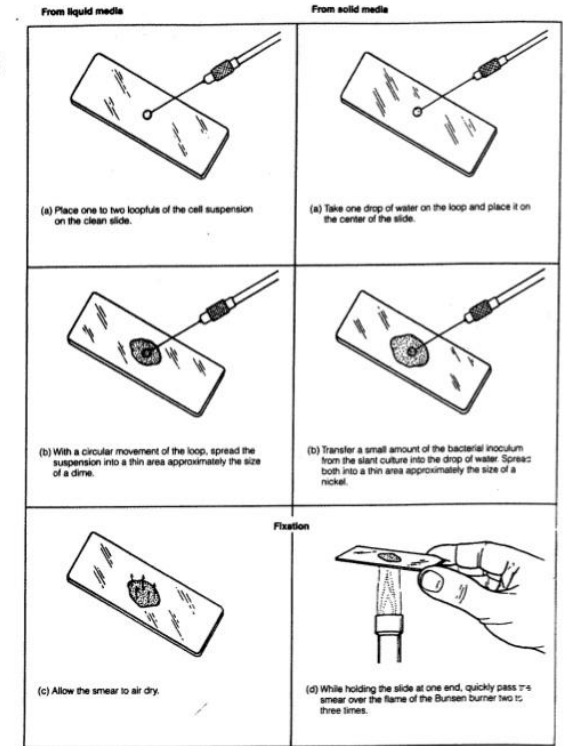


Figure 7.1 Bacterial smear preparation

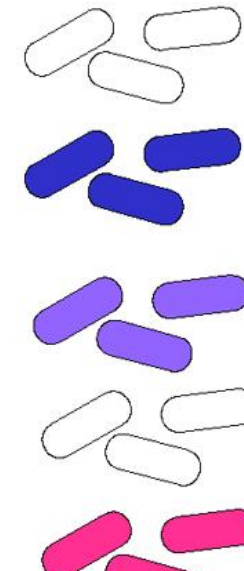
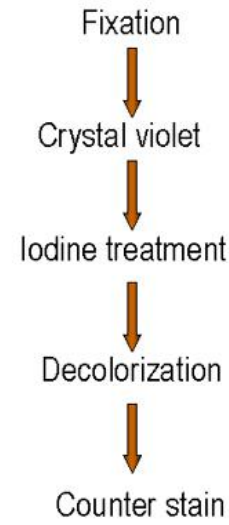
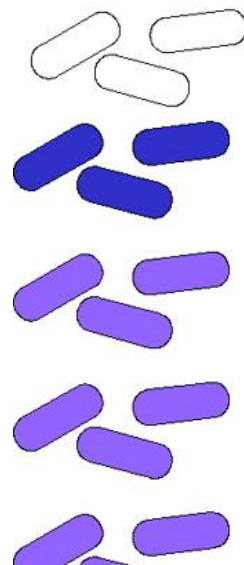
EXPERIMENT 7

Gram Positive

Gram Negative

What happens in staining process!!!

	Acid Fast	Non Acid Fast
Carbol fuchsin		
Acid alcohol decolorizer		
Counterstain		
methylene Blue		



Putting biochemical for identification

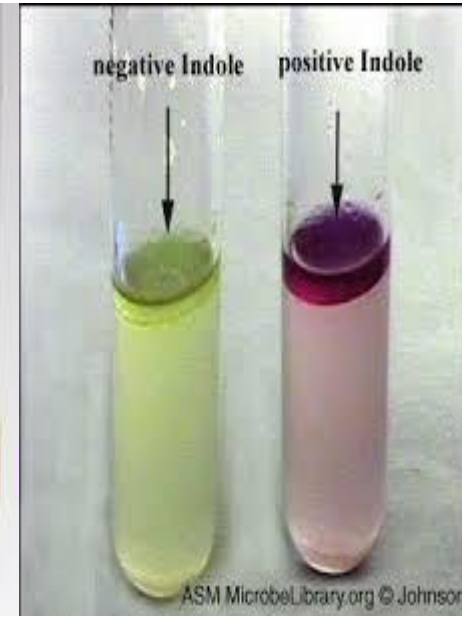
- Significance
- Sequence



Citrate



Triple sugar iron agar



Indole test

Drug susceptibility : manual /automated

- Disc diffusion/E test

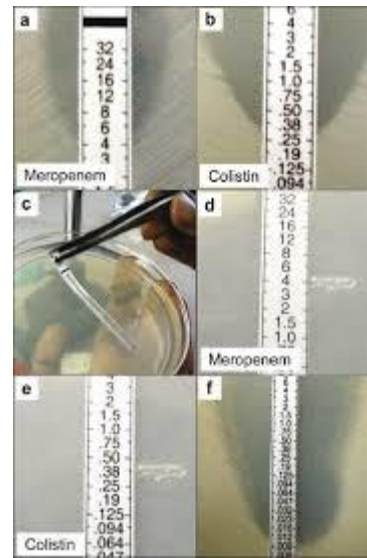
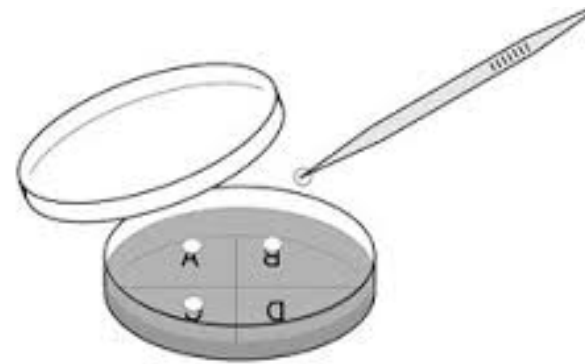
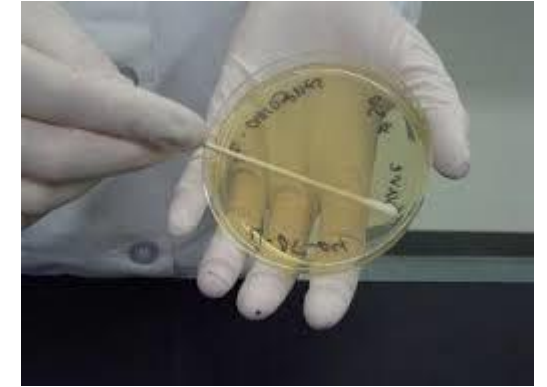
-Media

-method

Applying discs

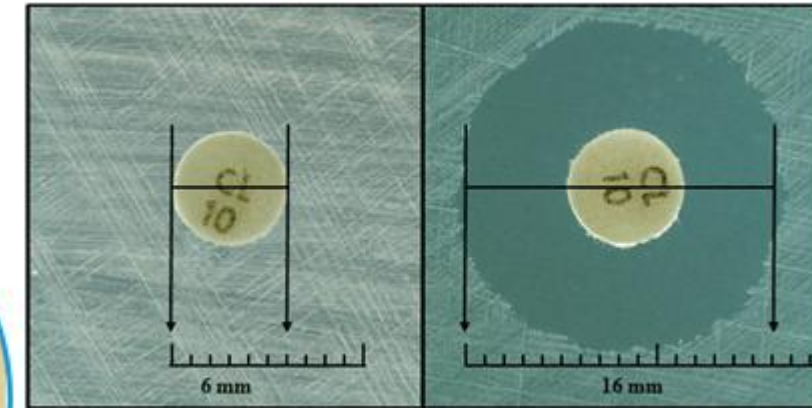
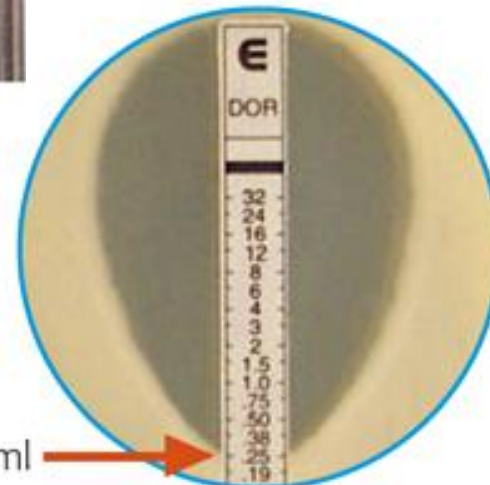
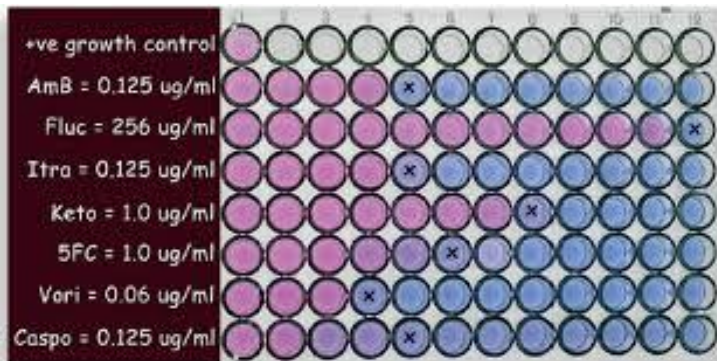
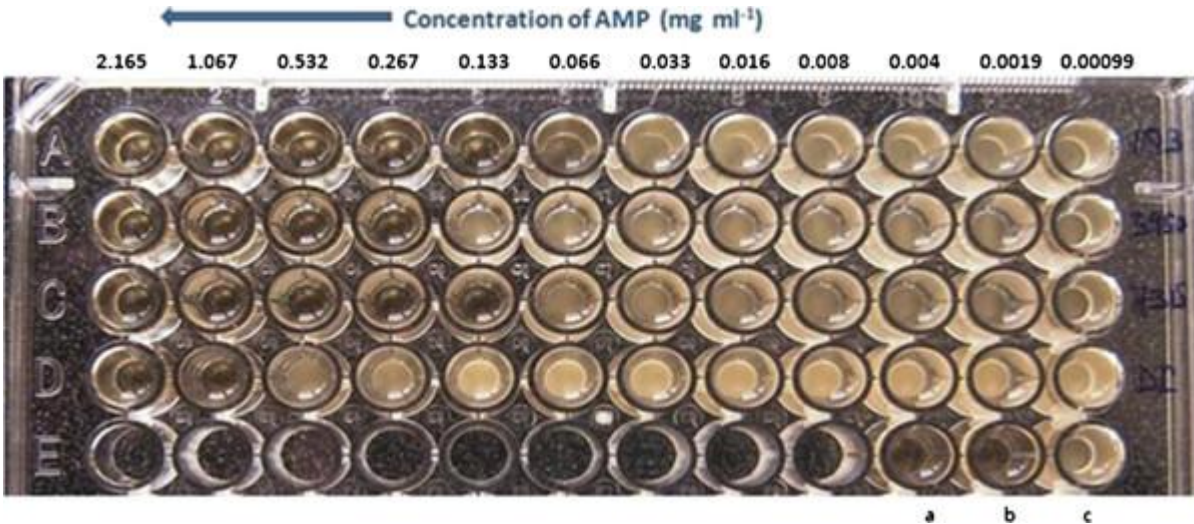
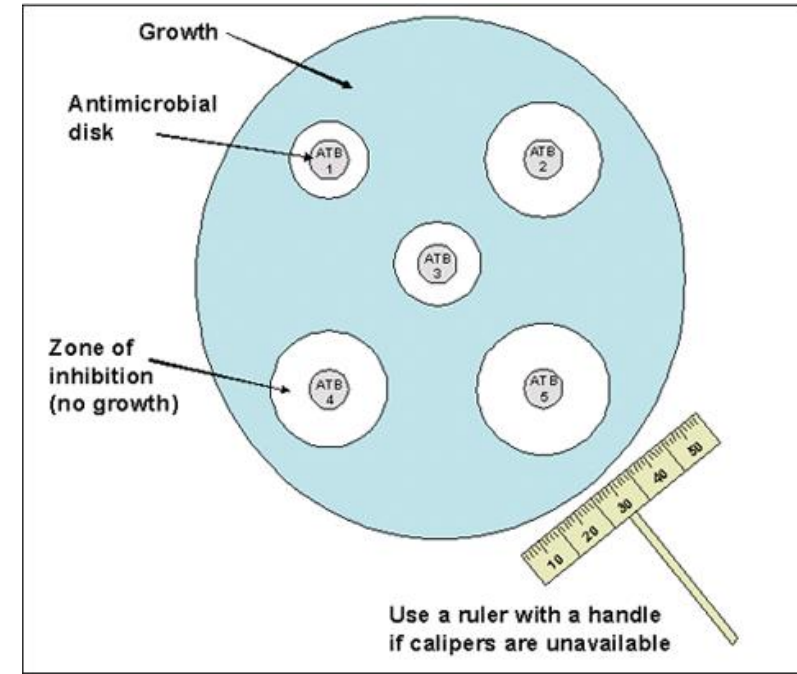
-incubation

- Automated methods



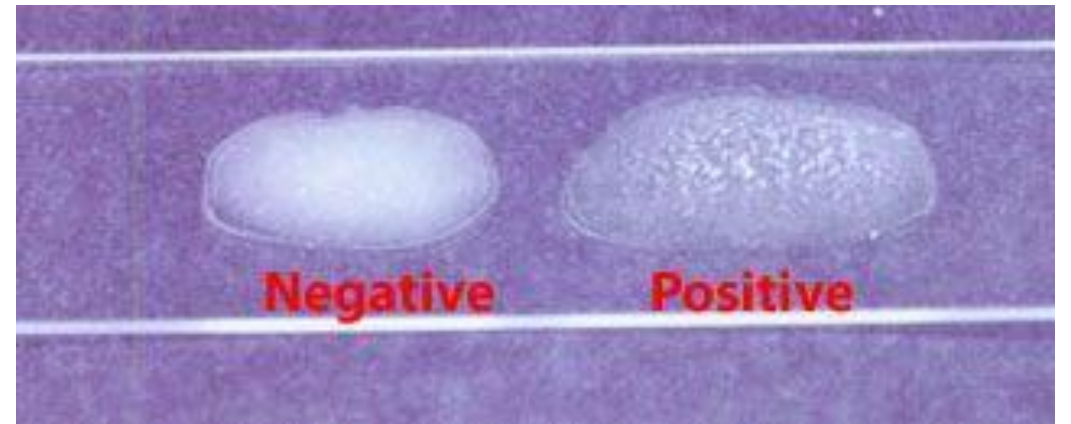
Taking DST readings

- Light
- Measurement of zone of inhibition
- Interpretation



Agglutination tests

- E.g
- Procedure
- Significance



Quality control

- Verification/Validation
- Internal
- External
- Frequency
- Corrective actions

THANK YOU!