

Practical Urinary Tract Infections



Microbiology Team – 430

Done By:

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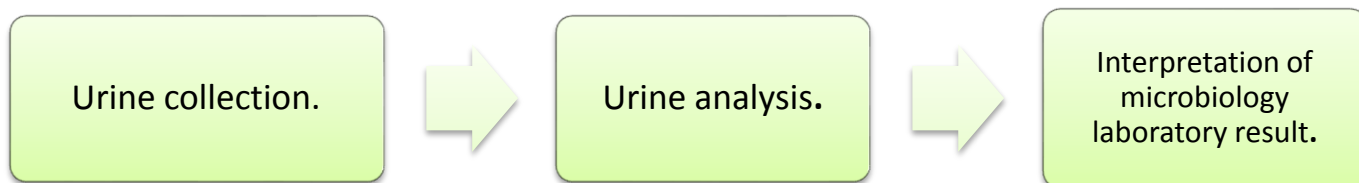
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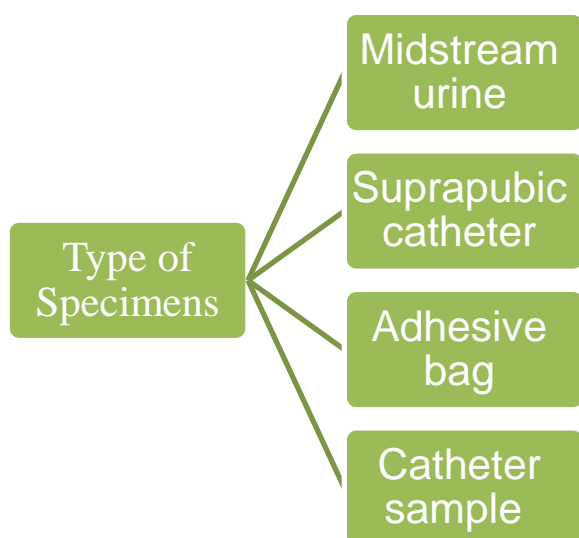
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Important aspects of Microbiologic Examination of UTI:



Urine collection:



The urinary catheter

Urine specimens for laboratory investigations from catheterized patients can be collected

Urine from the drainage bag should not be tested because it may have been standing for several hours.

Urine analysis:

- 1- Dip stick (leukocyte esterase, nitrate test).
- 2- Microscopic ex; cell-counting chamber (to count the number of WBCs).

Laboratory examination of urine: (Quantitative -Colony counts-)

- 1- Culture: (if we find $> 10^5$ organism/ml significantly there is an infection)
 - a. **Blood Agar:** culturing fastidious microorganism (needs more nutrients) and observed the hemolytic reaction (alpha, beta or gamma).
 - b. **MacConkey's agar:** observe lactose and non-lactose fermenting colonies.
 - i. **Lactose** fermenting colonies --> **pink**. (Then we do urease test).
 - ii. **Non-lactose** fermenting ones are colourless or appear same as the medium. (Then we do oxidase test).
 - c. **CLED agar (Cystine-Lactose-Electrolyte-Deficient -green in color-):**
Selective culture medium for detection and isolation Of Escherichia coli and coliform bacteria (bacteria in GIT) in urine.
- 2- Gram Stain:
 - a. **Blue** --> gram **+ve** bacteria.
 - b. **Red** --> gram **-ve** bacteria.



Causative agent of UTI:

Gram -ve	Gram +ve
<i>Escherichia coli</i>	<i>Enterococcus</i>
<i>Klebsiella</i>	<i>Staphylococcus saprophyticus</i>
<i>Proteus</i>	<i>Streptococcus agalactiae</i> (group B)
Other <i>Enterobacteriaceae</i> (<i>Enterobacter</i> , <i>Citrobacter</i>)	<i>Staphylococcus aureus</i> (Associated with staphylococemia)
<i>Pseudomonas aeruginosa</i>	

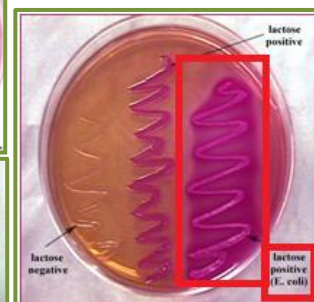
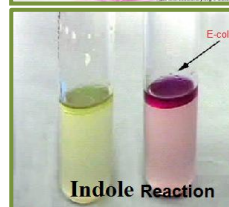
Other organism:

- *Candida*
- *Schistosoma haematobium*

Gram Negative Bacilli :

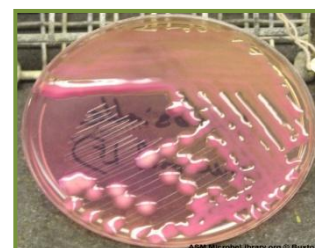
E-coli:

- Lactose fermentation (which means it appears pink in **MacConkey's agar**).
- Urease negative & citrate negative too.
- Have a **positive indole reaction**
- Treated with
 - Nitrofurantoin in cystitis.
 - ciprofloxacin & Co-trimoxazole and sometimes we can use amoxicillin in pyelonephritis.
 (Resistance to Ampicillin “ be careful”)



Klebsiella pneumoniae:

- Lactose fermentation.
- Urease +ve & citrate +ve too.
- Indole –ve.
- **Mucoid appearance** (because of the capsule).
- Treated with TMP/SMX.



Enterobacter aerogenes:

- Lactose fermentation.

Remember: *Pseudomonas aeruginosa* → +ve oxidase

- Urease +ve & citrate +ve too.

Proteus :

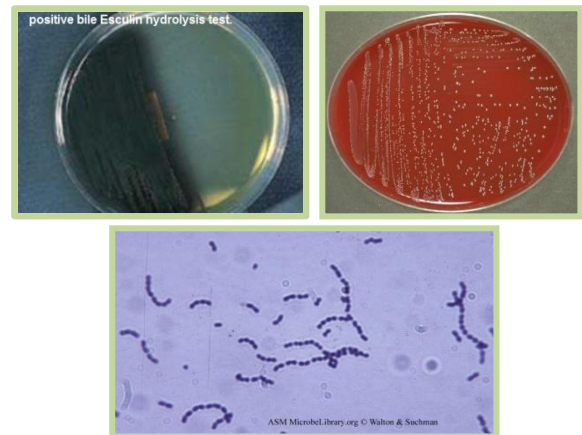
- Non-lactose fermentation.
- **Urease +ve.**
- Oxidase +ve
- Has **swarming growth in Blood agar** --> CLED media inhibit the proteus swarm (due to absence of electrolytes).
- Treated with Gentamicin, Piperacillin & Fluroquinolones.
- ↑ Ph of urine.
- **Alkaline urine** --> could lead to **stone formation**



Gram Positive Bacteria :

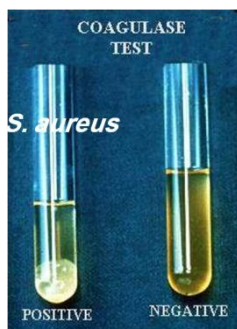
Enterococcus species: (e.g. E.faecalis)

- Cocci.
- Catalase –ve --> streptococcus.
- **positive bile Esculin hydrolysis test** --> **Gamma hemolytic (blackish colonies).**
- Treated with **ampicillin**



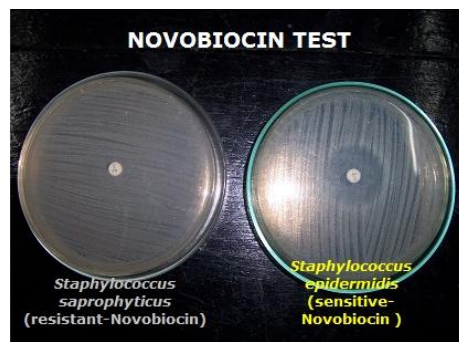
Staphylococcus aureus:

- Cocci.
- Catalase +ve (presence of bubble reaction) --> staphylococcus.
- Coagulase +ve--> aureus.



Staphylococcus saprophyticus:

- Cocci.
- Catalase +ve --> staphylococcus.
- Coagulase –ve --> saprophyticus or epidermis.
- **Novobiocin resistance** --> saprophyticus



To Summarize :

- Urine analysis should be in 1 hour after collecting, to avoid normal flora multiplication
- Dipslides is a transport media
- **MSU is the most common urine collection method**
- Other ways to collect urine: catheter and aspiration
- Lab investigation in urine analysis:
 - **Dip stick (leukocyte esterase, nitrate test).**
 - **Microscopic findings (10 WBC/hpf) pyuria**
 - Culture on agar (needs 18 hours)
- **On culture we find significant bacteruria (10^5 bacteria/ml)**
- In lab we don't use 1 ml of urine sample to culture on agar, we only use **(0.001ml)** sample, So significant bacteruria in lab is (100 bacteria/0.001ml)
- Pyelonephritis treatment for 10-14 days But Cystitis treatment for 3-7 days
- Ampicillin for enterococcus, any other drug for E.coli , and Any drug for proteus

Agar type	Findings
McConkey agar	Lactose fermenting colonies appear pink
Blood agar	Proteus swarming "motile" (growth in waves)
CLED agar	Inhibit Proteus swarming (growth in waves)

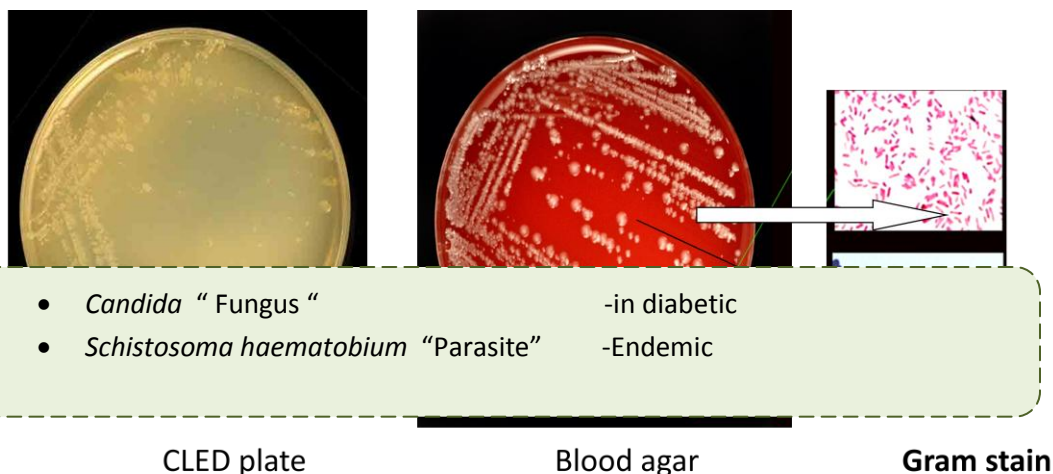
Gram -ve	Gram +ve
<i>Escherichia coli</i> <ul style="list-style-type: none"> ✓ Lactose fermentation ✓ Have a positive indole reaction 	<i>E.faecalis</i> <ul style="list-style-type: none"> ✓ Esculin test +ve (streptococcus Esculin test -ve) "<u>blackish colonies</u>" ✓ Old men with prostate enlargement ✓ Treated with ampicillin
<i>Klebsiella</i> <ul style="list-style-type: none"> ✓ Lactose fermentation. ✓ Urease +ve & citrate +ve too. <u>Indole -ve.</u> ✓ <u>Mucoid appearance</u> (because of the capsule). 	<i>Staphylococcus saprophyticus</i> <ul style="list-style-type: none"> ✓ Catalase test +ve ✓ Coagulase test -ve ✓ Causes honeymoon cystitis
<i>Proteus</i> <ul style="list-style-type: none"> ✓ Non-lactose fermentation. ✓ Urease +ve. ✓ Has swarming growth in Blood agar ✓ Split urea → alkaline urine → stone formation 	<i>S.aureus (Associated with staphylococemia)</i> <ul style="list-style-type: none"> ✓ Catalase test +ve ✓ Coagulase test +ve ✓ Hematogenous spread
Other <i>Enterobacteriaceae</i> (<i>Enterobacter, Citrobacter</i>)	<i>Streptococcus agalactiae</i> (group B)
<i>Pseudomonas aeruginosa</i> <ul style="list-style-type: none"> ✓ hospital acquired infection 	

Cases:

Case 1

The blood agar plate and CLED plate provided were inoculated with a sample of urine from a patient with a suspected urinary tract infection. Examine the plates and photographs provided.

- Identify the colonies on the blood agar plates and photographs?
 - **The photographs show the results of the Gram stain of each colony type.**
Large colonies are **Gram – ve** and small colonies are **Gram +ve**



Case 2

These Blood agar and CLED agar plates were inoculated with MSU from a 45 years old man suspected of having bladder stone and complaining of burning micturation.

Urine examination showed :

Moderate number of WBC and a PH of 8

A) What is the likely this pathogen?

➤ **proteus**

B) How would you confirm the identity of this pathogen?

➤ **Urease Test will be positive**

C) What is the role of this organism in forming stones?

➤ **Urease enzyme in proteus splits urea into ammonia and alkalizes the urine with production of crystals.**



CLED Agar

CLED Agar