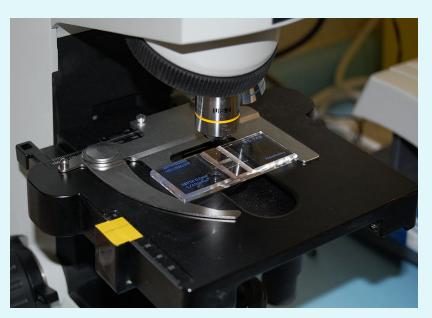
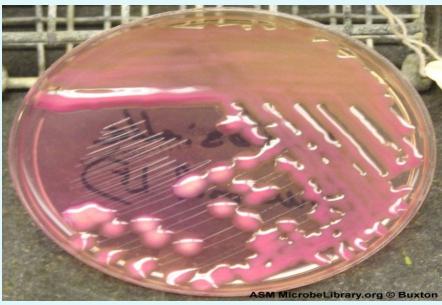
# Practical of Urinary Tract Infection





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**Department of Microbiology** 

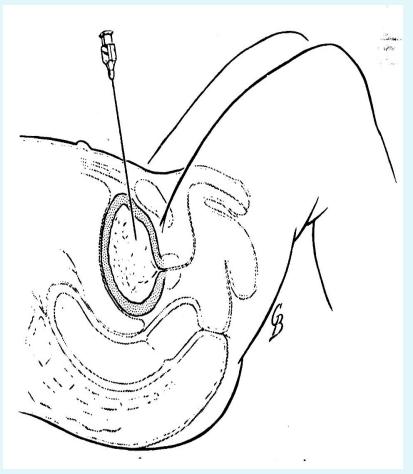
# Important aspects of Microbiologic Examination of UTI:

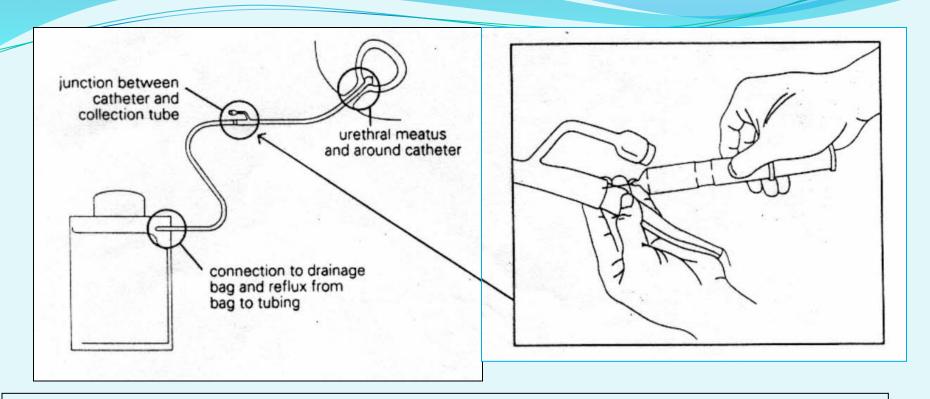
- Urine collection
- Urine analysis
- Urine culture
- Interpretation of microbiology laboratory result

# Type of Specimens

- ➤ Midstream urine (MSU)
- ➤ Adhesive bag
- ➤ Suprapubic aspiration
- ➤ Catheter sample





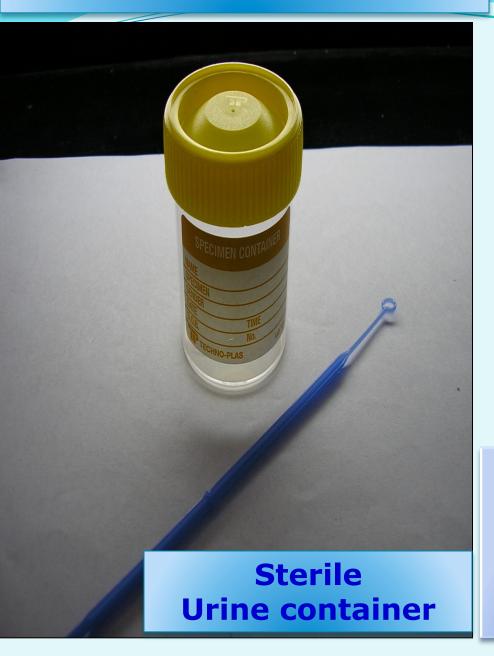


#### The urinary catheter

Urine specimens for laboratory investigations can be collected from catheterized patients as shown (left). The second port is for putting fluids into the bladder (right).

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

## TRANSPORT MEDIA



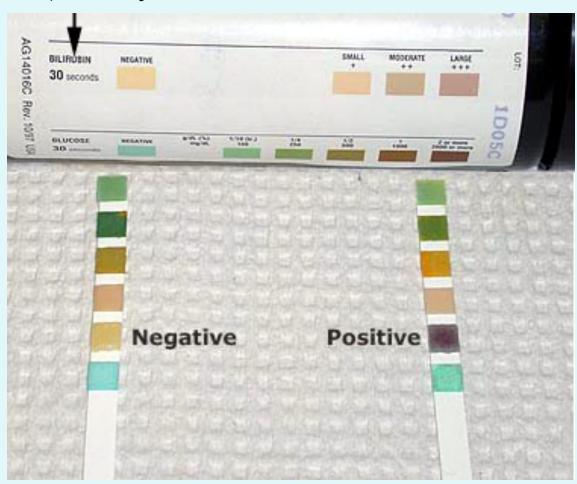


# dipslides

One side is CLED media, the other can be MacConkey (MAC) agar or blood agar.

# Urine analysis;

1- Dip stick (leukocyte esterase ,nitrate test)



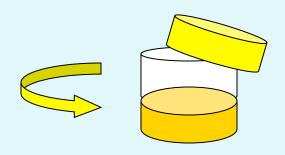
# Urine analysis;

- 1- Dip stick (leukocyte esterase ,nitrate test)
- 2-microscopic ex; cell-counting chamber

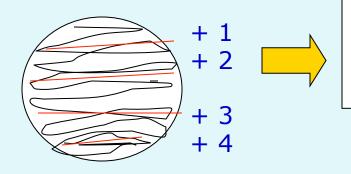




### Laboratory examination of urine



#### **Quantitative (Colony counts)**



a urine sample is streaked on surface of Blood Agar plate and CLED agar / Mc Conkey agar with a special loop calibrated to deliver a known volume.



Over night incubation

Isolation of colonies, Biochemical tests,
Drug susceptibility test,



Over night incubation

**RESULT** 

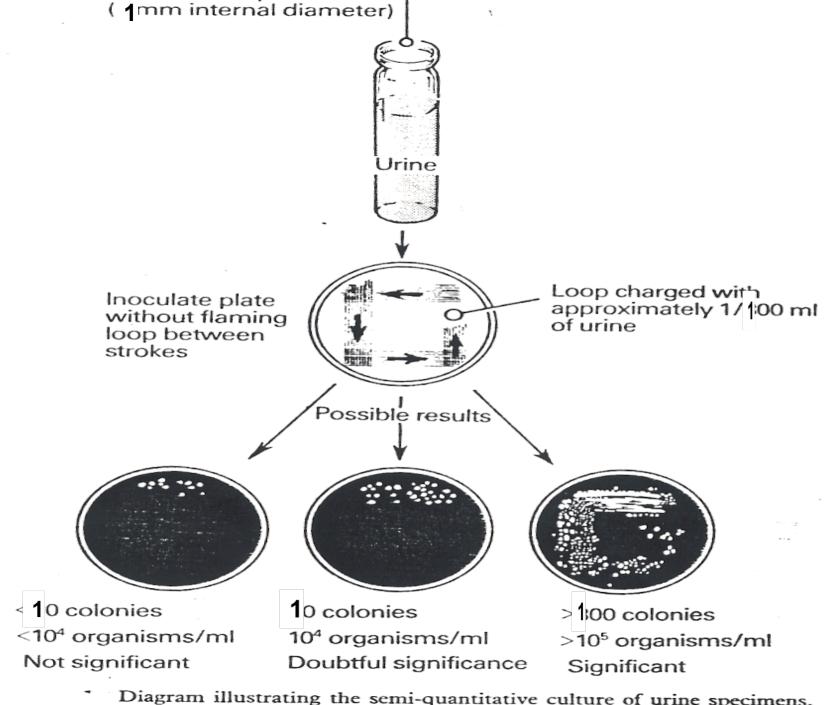


Diagram illustrating the semi-quantitative culture of urine specimens.

GRAM NEGATIVE	GRAM POSITIVE	
Escherichia coli	Enterococcus	
Klebsiella	Staphylococcus saprophyticus	
Proteus	Streptococcus agalactiae (group B)	
Other Enterobacteriaceae (Enterobacter, Citrobacter)	Staphylococcus aureus <sup>1</sup> (Associated with staphylococcemia <sup>(</sup>	
Pseudomonas aeruginosa		

# •Other organisms;

- •Candida
- •Schistosoma haematobium

Causes of UTI's	Outpatients	Inpatients
Escherichia coli	(%) 53-72	(%) 18-57
Coagulase negative Staphylococcus	2-8	2-13
Klebsiella	6-12	6-15
Proteus	4-6	4-8
Morganella	3-4	5-6
Enterococcus	2-12	7-16
Staphylococcus	2	2-4
aureus		
Staphylococcus saprophyticus	0-2	0.4
Pseudomonas	0-4	1-11
Candida	3-8	2-26

### culture media

blood agar



an enriched medium

MacConkey agar



a differential medium

**CLED** agar

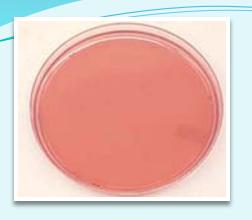


Selective medium

# **Blood agar**

An enriched medium, especially for culturing fastidious microorganism and observed the hemolytic reaction





MacConkey's agar showing both lactose and non-lactose fermenting colonies. Lactose fermenting colonies are pink whereas non-lactose fermenting ones are colourless or appear same as the medium.



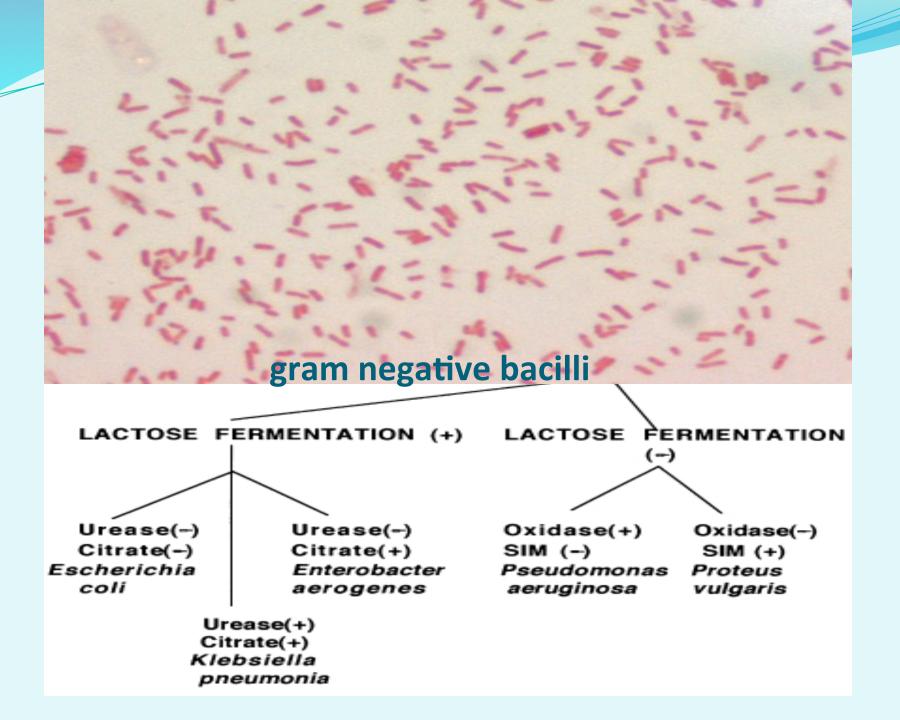


# **CLED** agar

Selective culture medium for detection and isolation Of *Escherichia coli* and *coliform* bacteria in urine



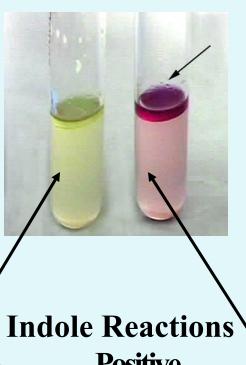




# E coli



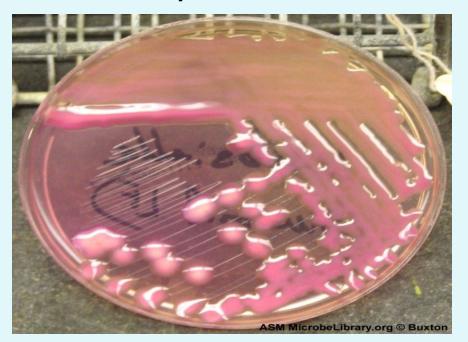




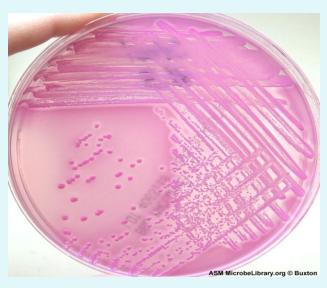
Negative

**Positive** 

#### Klebsiella pneumoniae



E coli





Klebsiella

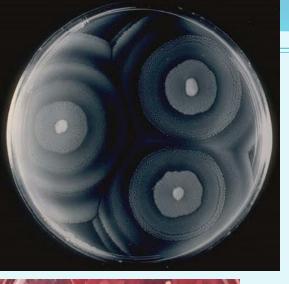
# Proteus growth: Swarming







CLED [(Cystine-Lactose-Electrolyte-Deficient) inhibits the proteus swarm proteus is Urease positive
Urease splits urea into
ammonia; and alkalinizes
the urine with production of
crystals



Proteus spp,



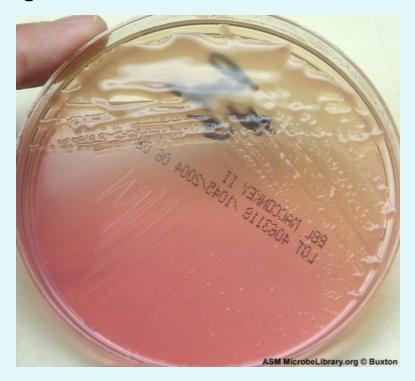


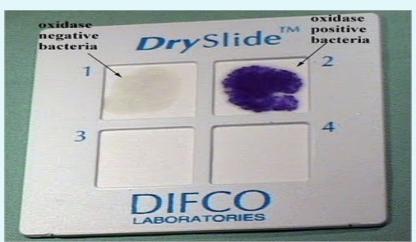




#### Pseudomonas aeruginosa







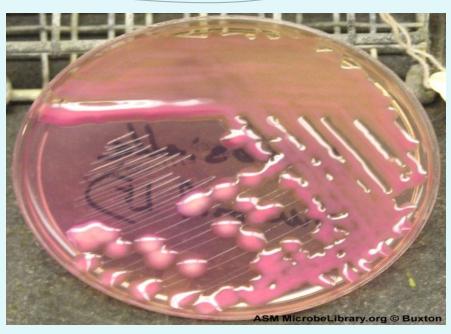
#### E coli



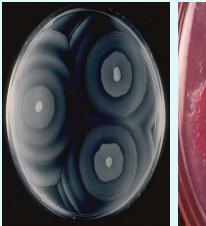
#### Pseudomonas aeruginosa



#### Klebsiella pneumoniae



Proteus spp,





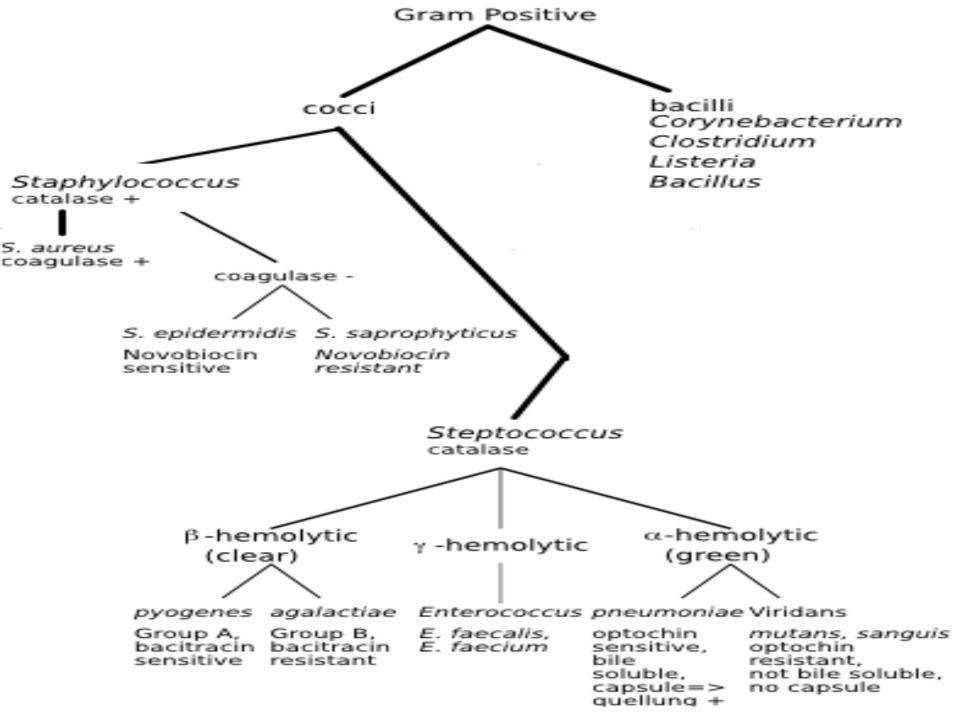




#### Three API 20E strips:

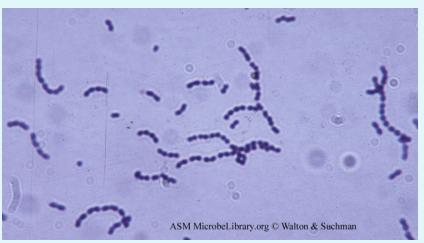
- a. Immediately after inoculation
- b. After 24 hours incubation
- c. That in (b) after addition of reagents to certain wells.

The organisms here is *Escherichia coli*. Here the first carbohydrate well (glucose) is also used for the nitrate reduction test



### Enterococcus species





### **Biochemical Identification**

Bile Esculin hydrolysis



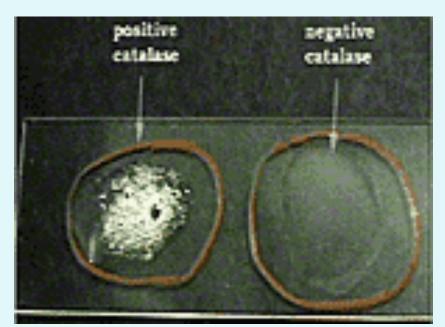
Both Group D streptococci and enterococci produce a positive (left) bile Esculin hydrolysis test.

# Staphylococcus spp



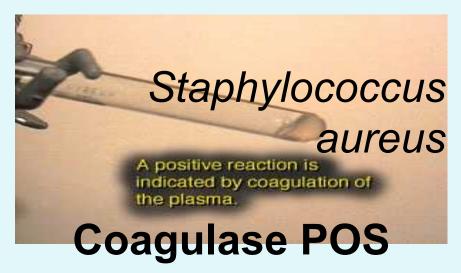
# **Differential Characteristics**

Catalase  $2H_2O_2 \rightarrow O_2 + 2H_2O$ Streptococci vs. Staphylococci



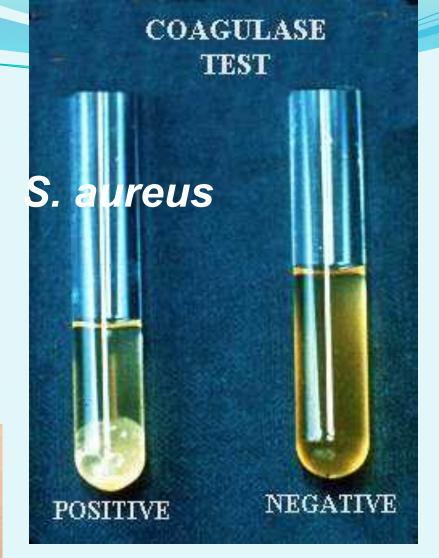


# Differential Characteristics



If the cells did not produce coagulase, the plasma remains fluid.

Coagulase NEG

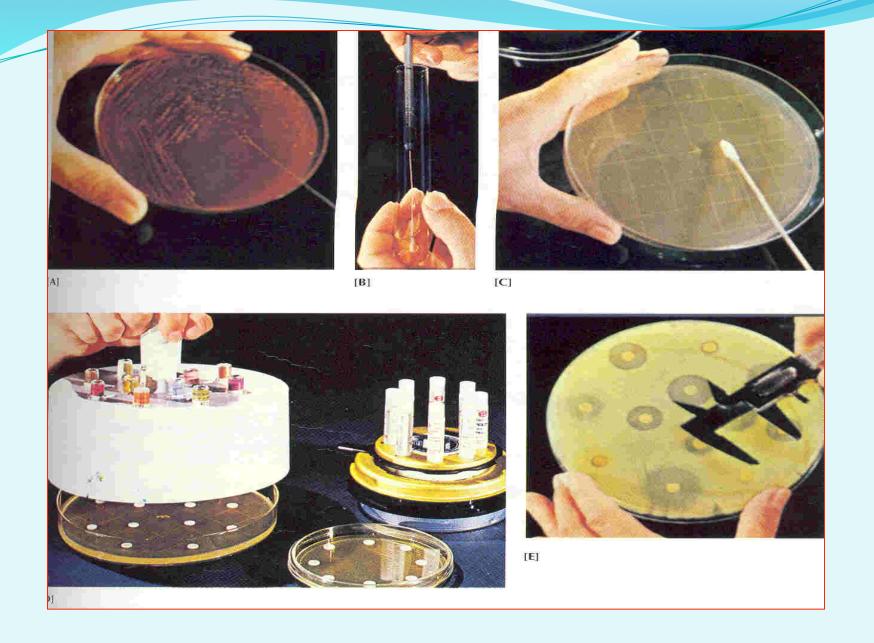


## **NOVOBIOCIN TEST**



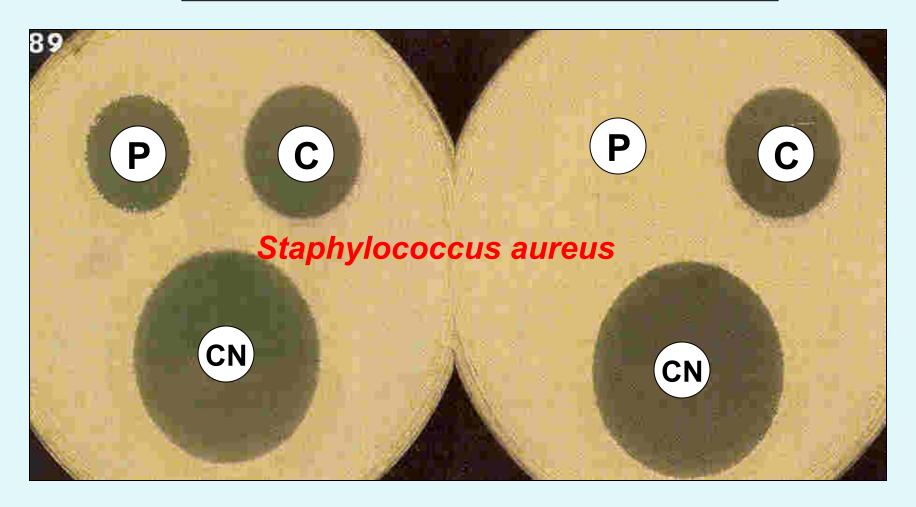
Staphylococcus saprophyticus (resistant-Novobiocin)

Staphylococcus epidermidis (sensitive-Novobiocin)

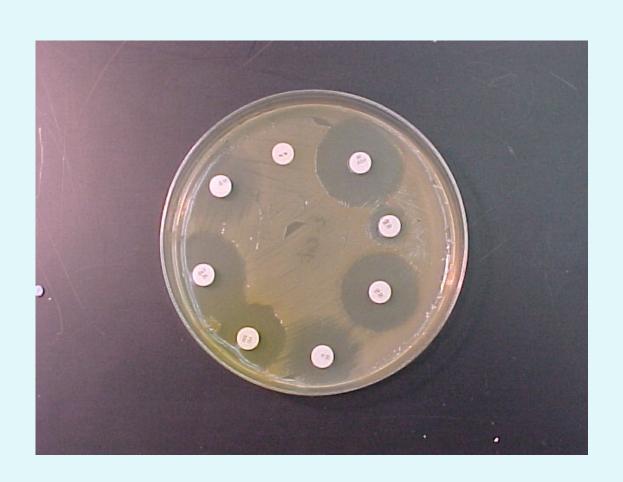


Gentamisin (CN) : 12 - 15 Chloramphenicol (C) : 12 - 18 Penicilin (P) : 28 - 29

R (Resistant); S (Sensitive)



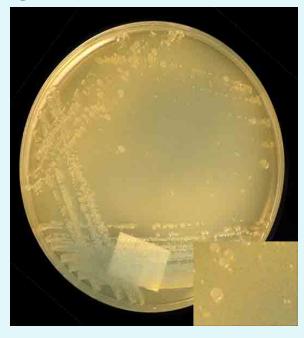
### Antibiotic sensitivity test: Agar diffusion method

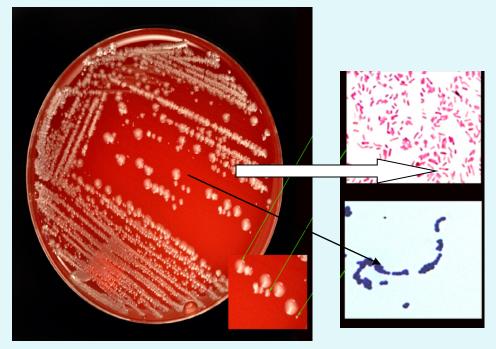


## Case 1

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......and small colonies are Gram......





CLED plate Blood agar Gram stain

## 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





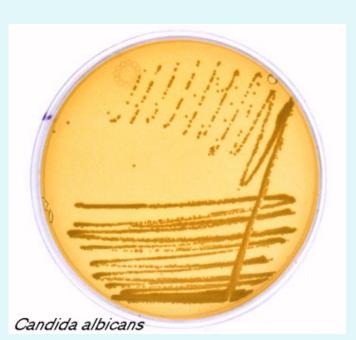
**CLED** 

**Blood agar** 

- A) What is the likely this pathogen?
- B) How would you confirm the identity of this pathogen?
- C) What is the role of this organism in forming stones?

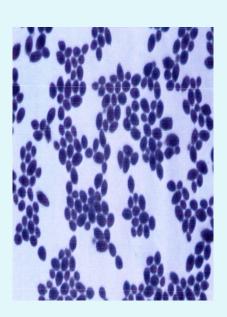
### Candida albicans

# Growth on Sabouraud's Dextrose Media



# Candida albicans on blood agar;





# Candida albicans: identification tests

#### Chlamydospore



#### Germ tube test



## Schistosoma haematobium



Schistosoma
haematobium
(urine; eggs 115-170 x
45-65 micrometers)
(primates)

### discussion

Mention one organism from each of the following which may cause urinary tract infection

- A) Bacteria .....
- B) Parasites .....
  - C) Fungi .....

