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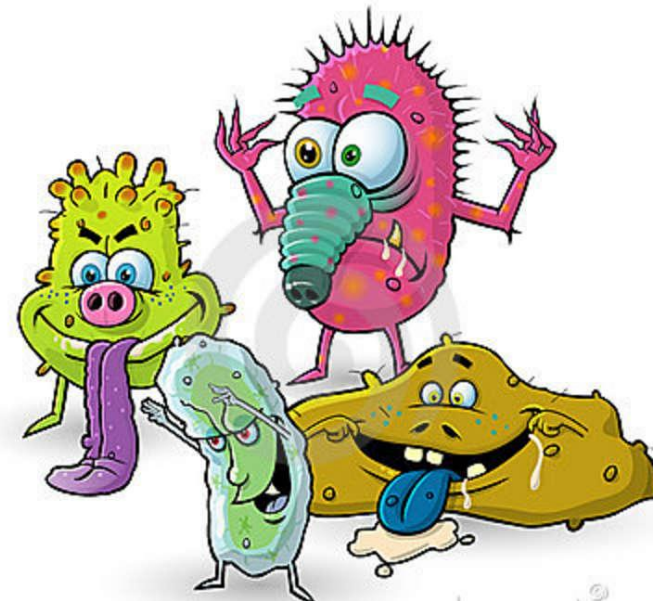


MTeam 432
Microbiology

PRACTICAL LECTURE ORGANISMS CAUSING UTI

Objectives:

Were not given.



Color
guide

- **Very important**

- male doctor's notes

- Additional information

- female doctor's notes

MICROBIOLOGIC EXAMINATIONS OF UTI:

1

Urine Collection:

1) collect urine sample:

- Mid Stream Urine
- suprapubic aspiration
- In and out catheter (intermittent catheter)

- Catheter
- adhesive bag

Urine from the bag of catheter should not be tested (standing for several hours) .So Catheter sample and adhesive bag are not prefer sample

2) Transport (quickly) by:

- sterile container
- dipslides: 1 is CLED media, the other is either MAC or blood agar.

2

Urine analysis:

1) Dip stick test to detect:

- leukocyte esterase
- Nitrate (It is not effective with gram +)
- pH of urine

2) Microscopic examination: cell counting chamber (detects bacteria, Pus cells , crystals or casts)

3) culture

cast (important) – which indicate Upper UTI and RBC

3

Laboratory tests:

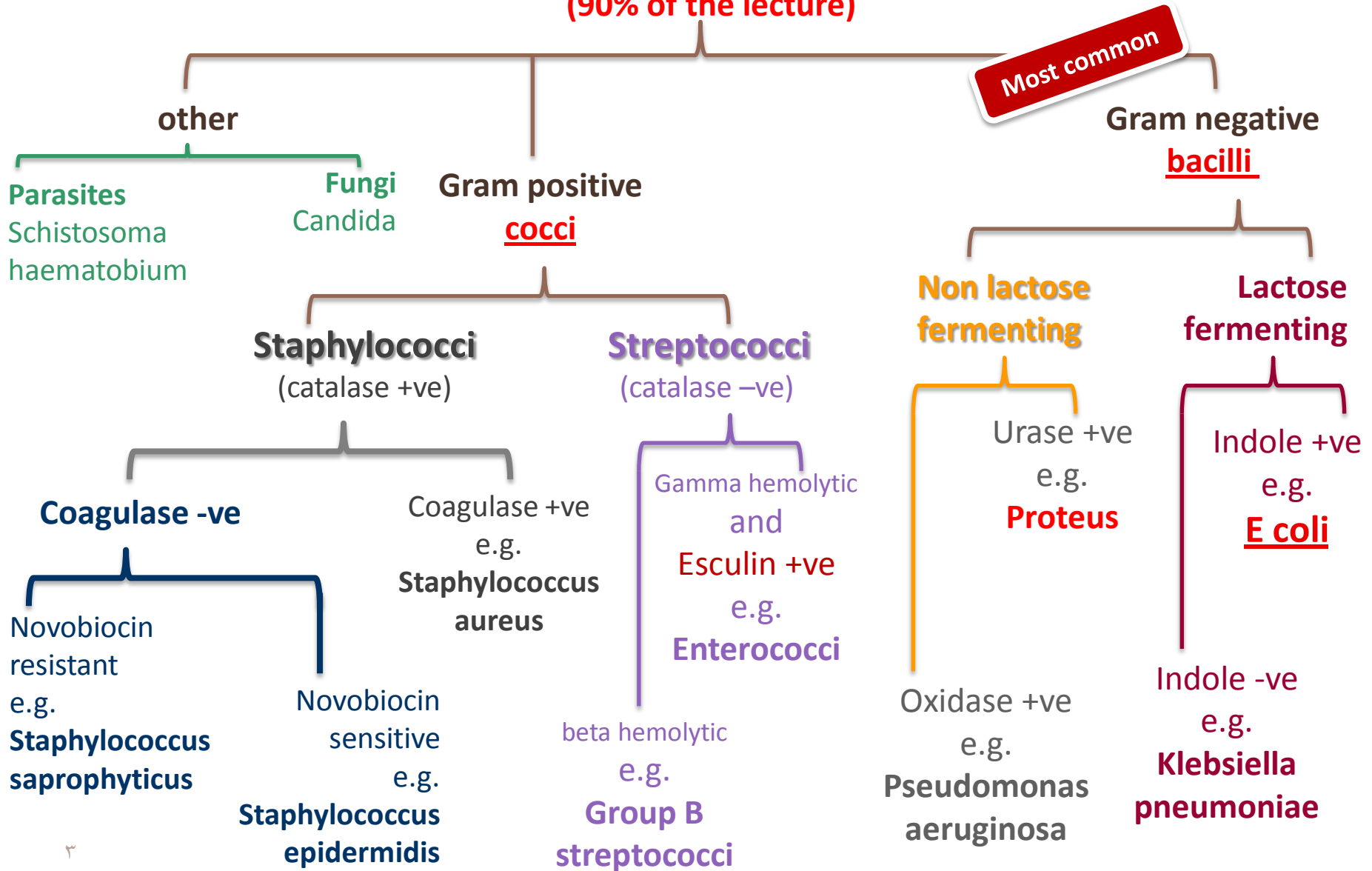
1) Quantitative: colony count by:

culturing → isolation of colonies → incubation

2) Semi-quantitative: detects significant bacteruria ($10^5/ml$)

Summary of the causes of UTI

(90% of the lecture)



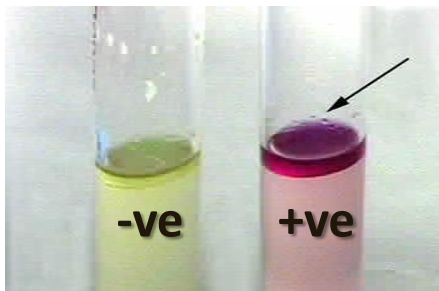
GRAM NEGATIVE BACTERIA

important

Gram
negative
Testing

| Material to be tested | Positive result | Negative result |
|--------------------------------------|---|---|
| 1) Lactose fermentation (next slide) | <ul style="list-style-type: none"> ▪ E coli ▪ Klebsiella pneumoniae | <ul style="list-style-type: none"> ▪ Proteus ▪ Pseudomonas aeruginosa |
| 2) Urease | Proteus | — |
| 3) Oxidase | Pseudomonas aeruginosa | <ul style="list-style-type: none"> ▪ E coli ▪ Klebsiella pneumoniae |
| 4) indole | E coli | Klebsiella pneumoniae |

Note: E coli and Klebsiella are enterobacteriaceae, they have common characteristics like:
+ve Nitrite & Lactose tests, – ve Oxidase tests



Indole test



Urase test



Oxidase test

GRAM NEGATIVE BACTERIA

Different media for culture

| Medium | Used for | positive result | negative result |
|---|---|------------------|-------------------|
| 1) Blood agar | An enriched medium, used for culturing fastidious microorganism and observed the hemolytic reaction | | |
| 2) MacConkey agar | Differential media. Differentiate b/w non-lactose fermenting colonies. | Pink color | Colorless |
| 3) CLED | | yellow | Colorless |
| Examples complete the missing names ;) | | E coli and | Proteus and |



Blood agar



MAC agar



CLED

It is Selective culture medium for detection and isolation Of E.coli and coliform bacteria in urine

GRAM NEGATIVE BACTERIA

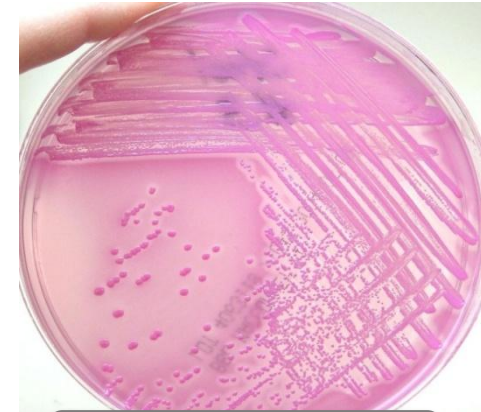
Further details about gram -ve bacilli

E coli

Gram negative bacilli

Identification:

- Pink on MAC
 - Yellow on CLED
 - Indole positive
- (same as klebsiella)
- (in contrast of klebsiella)



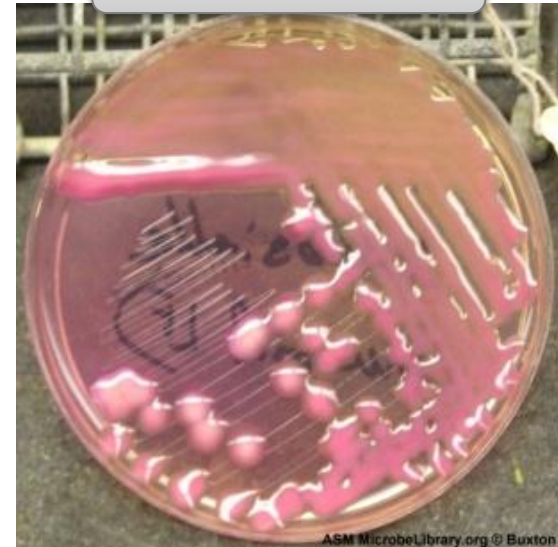
MAC agar

Klebsiella pneumoniae

Gram negative bacilli

Identification:

- Have mucoid appearance
- Pink on MAC
- Yellow on CLED
- Indole negative



ASM MicrobeLibrary.org © Buxton

GRAM NEGATIVE BACTERIA

Further details about gram -ve bacilli

Proteus spp: for upper and lower UTI. It produce:
1)urease
2) swarming in plate 3)stone 4) Recurrent infection

- ✓ No swarming in CLED → because it's Electrolyte-Deficient
- ✓ No swarming in MAC → because it contains bile

Swarming (circles) in blood agar



Proteus Gram negative bacilli

Identification:

- **Swarming:** gives waves when tested by blood agar, not in CLED or MAC
- colorless on MAC
- colorless (blue) on CLED
- **Urase positive:** split urea to Ammonia → alkaline urine → ↑ PH → Increase the potential of renal stones

Pseudomonas aeruginosa

Gram negative bacilli

Identification:

- Colorless on MAC
- colorless on CLED
- **oxidase positive**



GRAM POSITIVE BACTERIA

Enterococci

Gram positive cocci in chains

Identification:

- No hemolytic reaction on blood agar (gamma hemolytic)
- **Esculin positive**

Examples: enterococcus faecalis and enterococcus faecium



Blood agar



Bile esculin hydrolysis

Positive. *why?*
because it
produces enzymes
that hydrolysis the
bile.

GRAM POSITIVE BACTERIA

S.aureus

Gram positive cocci in clusters

Identification:

- **Coagulase positive** (in contrast to s.epidermidis and s.saprophyticus)

s.epidermidis

Gram positive cocci in clusters

Identification:

- **Coagulase negative** (in contrast to s. aureus)
- **Novobiocin sensitive** (in contrast to s.saprophyticus)

s. saprophyticus

Gram positive cocci in clusters

Identification:

- **Coagulase negative** (in contrast to s. aureus)
- **Novobiocin resistance** (in contrast to s.epidermidis)

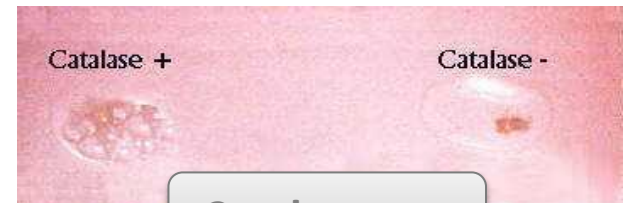
Coagulase: an enzyme that changes fibrinogen to fibrin

*Catalase test differentiates between staphylococci and streptococci. Catalase is produced by staph. So it will appear as bubbles gas if it positive

STAPHYLOCOCCI

Catalase for differentiate

All are ***catalase positive**
(in contrast to streptococci)

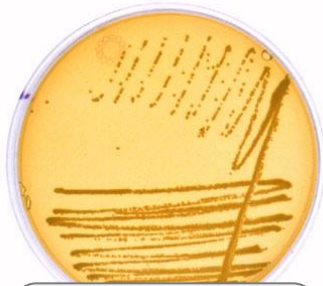


Catalase test

This slide is not important

FUNGI

Candida albicans



Sabouraud's
Dextrose Media



Blood agar



Chlamydospore



Germ tube

PARASITES

Schistosoma haematobium



Eggs of schistosoma
causes dysuria and
hematuria

important

| | | MAC agar | CLED | Blood agar |
|--------|-------------------|----------|---------------------|------------|
| Gram - | E.Coli | LF | √ | X |
| | Klebsiella Pneum. | LF (M) | √ | X |
| | ✦ Proteus | NF | Inhibition swarming | Swarming |
| | P.Aerogenosa | NF | X | X |

LF : Lactose fermentation
 NF : Non-Lactose fermentation
 (M) : Have mucoid appearance

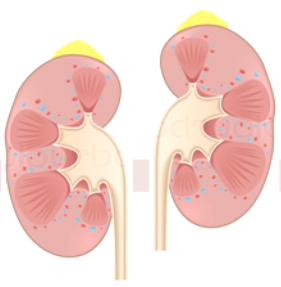
Important from 431

QUICK SUMMARY OF ORGANISMS

| | E.Coli | Klebsiella Pneum. | Proteus | P. Aerogenosa | Entero cocci | Staph Aureus | Staph Saprophyt. |
|----------------------|--------|-------------------|---------|---------------|--------------|--------------|------------------|
| Gram | -ve | -ve | -ve | -ve | +ve | +ve | +ve |
| Lactose fermentation | √ | √ | X | X | | | |
| ✦ Oxidase | -ve | -ve | -ve | +ve | | | |
| ✦ Urease | -ve | +ve | +ve | --- | | | |
| Indole | +ve | -ve | | | | | |
| Catalase | | | | | -ve | +ve | +ve |
| Coagulase | | | | | | +ve | -ve |
| Novobiocin | | | | | | Sensitive | resistant |

✦ Very important

Remember !
 Entrococci is
Esculin positive



Questions!

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

Urine examination showed :

Moderate number of WBC and a PH of 8

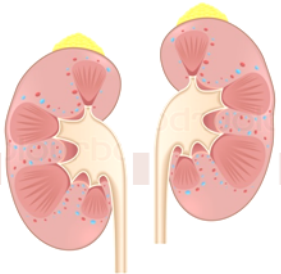


CLED



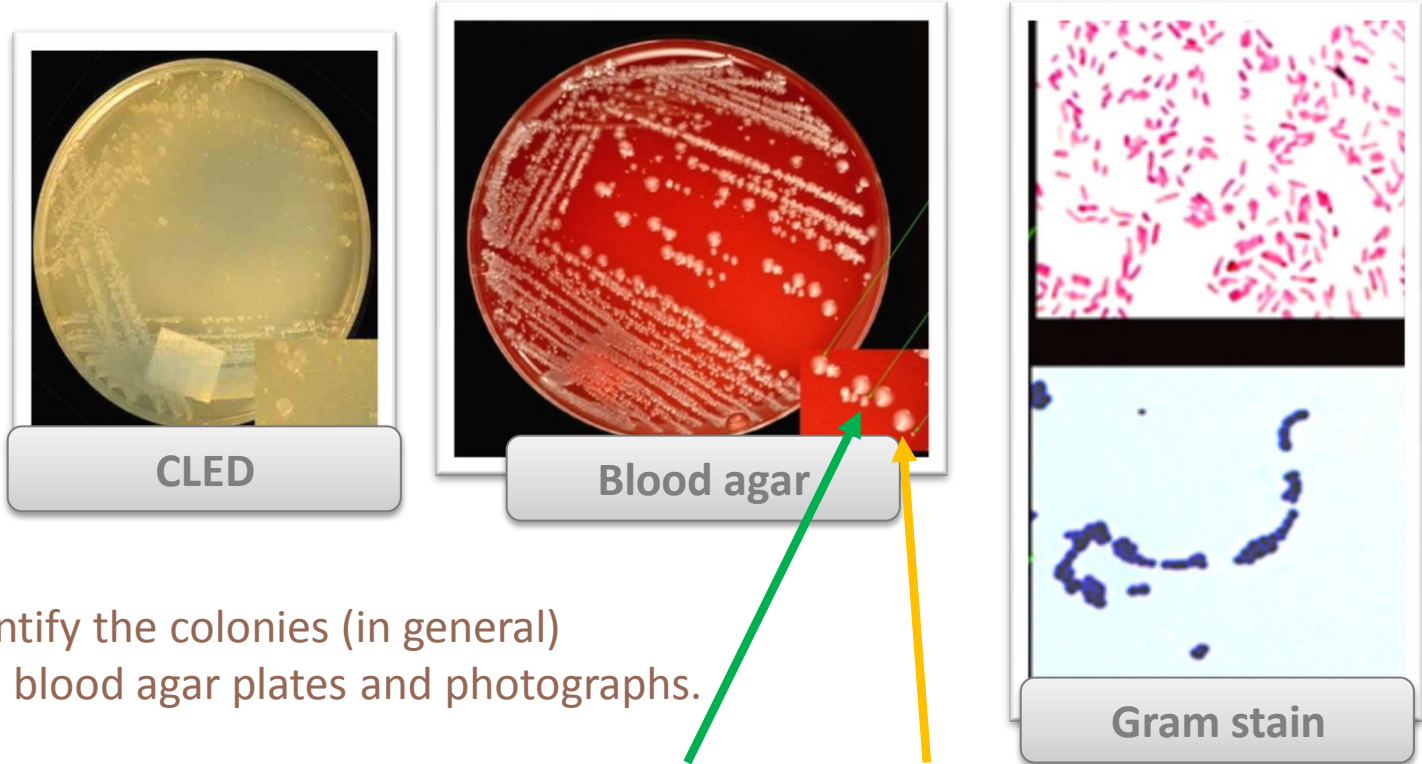
Blood agar

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



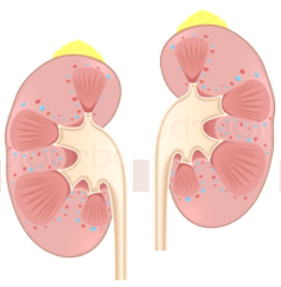
Questions!

The blood agar plate and CLED plate provided were inoculated with a sample of urine from a patient with a suspected urinary tract infection. After examining the plates and gram stain, two colonies, large and small, were found.



A. Identify the colonies (in general) on the blood agar plates and photographs.

Small colony **Large colony**



Questions!

*Good
luck
432!*

answers

Question 1:

A) Proteus

B) - Swarming on blood agar. - Urase test would be positive
- colorless (takes the color of the media which is blue) on CLED
- pH=8 (proteus causes alkaline urine)

C) it makes the urine alkaline. Alkalinization of urine could lead to the formation of stones

Question 2:

Large colonies: E coli. Because: - gram stain shows gram -ve bacilli
- CLED medium is yellow (i.e. lactose fermenting)

•It could also be klebsiella, but E coli is more likely to cause of UTI.

Small colonies: gram stain shows gram +ve cocci in clusters. So it could be one of the 3 staphylococci (further tests are needed to confirm which one)