Lec(1) Microbiology 26/5/2021

Staphylococci

Species :

Staphylococcus aureus

Staphylococcus intermedius

Staphylococcus hyicus

Staphylococcus saprophyticus

Staphylococcus chromogenes

Staphylococcus epidermidis

Morphology and staining :-

1- **Gram +ve, cocci**
2- The diameter varies from (1.5-0.8)μ
3- Arrangement: **bunches clusters (grape like cluster)** observed in smears taken from solid culture growth.
4- They are **Non-motile, non-spore forming**
5-**Some strains are capsulated when isolated from pathogenic specimen** or fresh culture

Cultural characteristics:-

1- aerobic or facultative anaerobic
2- grows on simple media at temperature ranged between 15-40C°.optimum temperature for **growth is 37 C°**
3- the **important media** are:

a- Nutrient agar:-the colonies are pigmented with (golden yellow or white color), circular, 1-2μ,convex, opaque, glistering with enteic edges.

b- **Blood agar:** pathogenic strains produce wide zone of β-hemolysis (clear zone)while other strains do not.

c- **Milk agar:** the colonies are similar to these on nutrient agar this medium stimulates the endopigmentation produced by some strains golden yellow color produced by *Staphylococcus aureus* ,lemon yellow color produced by *Staphylococcus citrus,* white color produced by *S. albus*

d-Manitol salt agar (MSA): because of *Staphylococci* tolerance to high salt concentration, the MSA acts as highly selective media for them. It contains 7.5-10% of sodium chloride.(this medium is used for isolation of *Staphylococci*from pathogenic specimens (pus, feces) . on this medium *Staphylococcus aureus* and some types of coagulase negative staphylococci CNS are positive for manitol fermentation appear as yellow colonies while other CNS appear as small orange or pink colonies.

Biochemical test :-

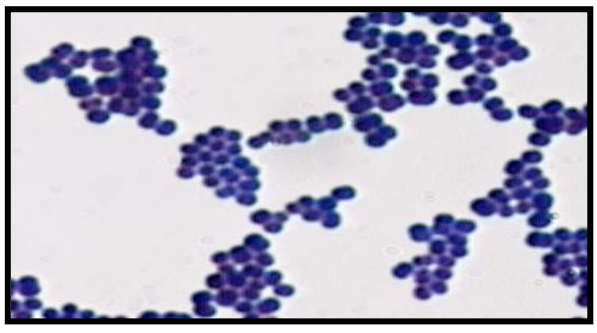
1-Catalase, Urease , DNase ,Coagulase and phosphatase positive.
2-Gives negative reaction for oxidase and indol test.
3-Ferment glucose , produce acid without gas
4-Utilize nitrate to nitrite.

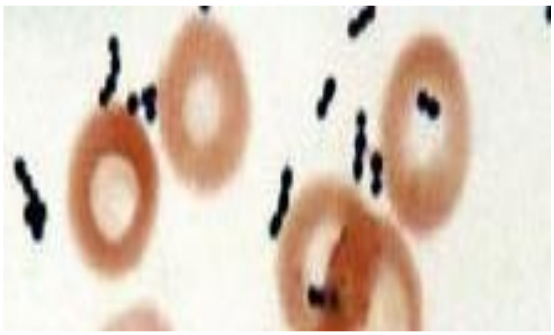
Table(1) Biochemical tests uses in the differential between types belong to *Staphylococci*

Species	Coagulase	B-haemolysis	maltose	mannitol	DNASE	Vp	Hyaluronidase
S. *aureus*	+(4hr)	+	+	+	+	+	+
s. *intermedius*	+(2hr)	+	+	v	-	-	-
s. *hyicus*	v	-	-	-	+	-	+
s. *saprophyticus*	-	-	+	v	-	-	v
s. *epidermidis*	-	-	+	-	-	-	-

Diagnosis :-

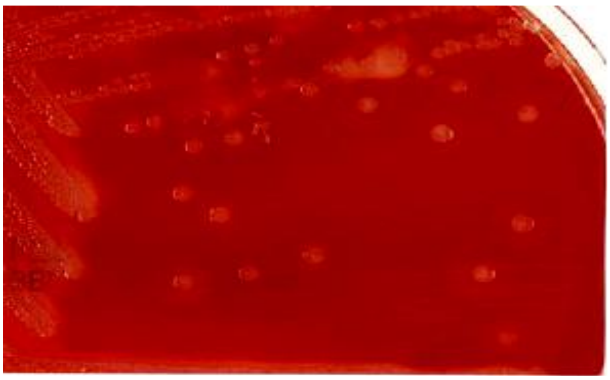
The specimens which is taken from the infection depends on the lesion
1-Gram stain smear
2-Culture on sheep and human blood agar to notice the hemolysis types
3-Using the selective media for isolation like MSA
4-Performing the coagulase test
5-Performing the biochemical test.



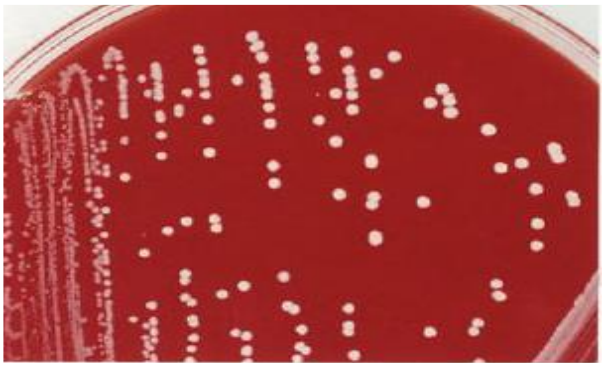


Staphylococci in bunches clusters (grape like cluster)

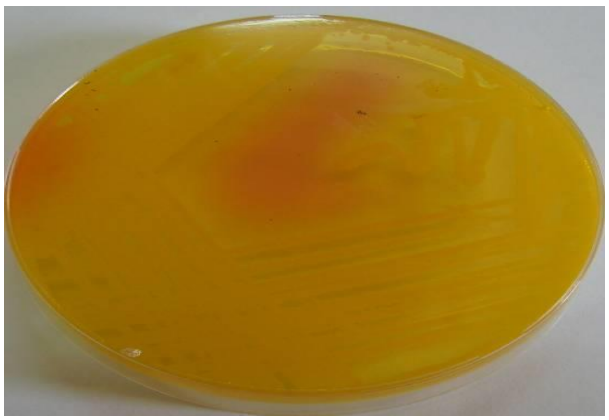
Staphylococci in pairs,short and long chains



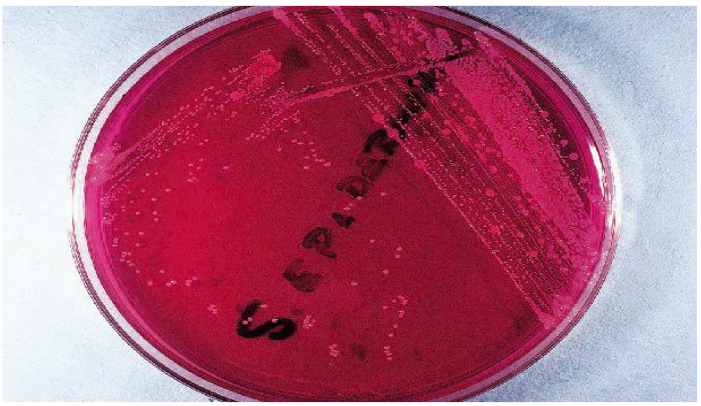
Colonies of staphylococcus aureus on5% sheep blood agar



colonies of staphylococcus epidermidis on blood agar



Colonies of staphylococcus aureus on **Mannitol Salt Agar**



colonies of of staphylococcus epidermidis on **Mannitol Salt Agar**

Lec(13) Microbiology /10/2011

Streptococci

Species :

Streptococcus agalactiae
S. dysagalactiae subsp. dysagalactiae
S.equi subsp. equi
S.equi subsp. zooepidemicus
S. uberis
S.bovis
S. pyogenes
Enterococcus faecalis

Morphology and staining :-

1-*Streptococci* and *Enterococc*i are **Gram+ positive cocci**.
2-They occur in **pairs or chains of varying length**.
3-**Non motile with the exception of some of the enterococci, non spore forming**.

Cultural characteristics:-

1-Most types of *Streptococci* are **aerobes or facultative anaerobes**.
2-They are **fastidious and require the addition of blood or serum or glucose to cultural media .**
3-**Optimum temperature for growth is 37c**.
4-the important media which are uses to cultivate the streptococci are.

a-**Blood Agar:-**Regarded the important media for cultivated these bacteria, because is stimulated streptococcal growing and we can observing the haemolysis types. The colonies are small spherical pin-head shape 1 µm in diameter, white to grayish in color ,on horse blood agar produce B-haemolysis(complete haemolysis).

b-**Edwards medium**:- it is selective media for *Streptococci* especially these which causing mastitis .this medium contains esculain and crystals violate which inhibiting other contaminates bacteria .

c-**MacConkey Agar:-** using for cultivating of enterococci which tolerate the bile salts on this medium and appear as small pin-head colonies.

d-**Brain heart infusion Agar:-**use to cultivated of streptococci .in this medium the characteristic chains arrangement seen more obviously in microscopic examination.

CAMP test:-this test is use to distinguished group B *Streptococcus agalactiae*Which cause chronic mastitis. demonstrating the arrow head- shaped enhancement heamolysis that occurs when the beta-toxin produced by *s. aureus* (the microorganism streaked horizontally across the sheep blood agar plate acts synergistically with the CAMP

facter protein produced by group *Streptococc*i (streaked perpendicular to the staphylococcus but not quite touching).

Lance field groups:-is the serological method to classification of streptococci based on the group specific C- substance (polysaccharide) in the bacterial cell wall, which precipitated with anti'sera. therefore distinguish streptococcus to groups(A,B,C,D,E,G,…ect

Procedure method of the test:-

1- cultural the *Streptococcus* species in nutrient broth for 24hr.then precipitated it by centrifuge.
2- Add 2ml from diluted HCL 200:1 to bacterial precipitate,then putting in water bath at 100c for10 minutes. Then filtration (the filter consider polysaccharide).
3- Putting the Antigen extract (polysaccharide) in test tubes and add antisera of different specificities. incubater in temperature room.
4- Positive result formation precipitate line during 30 minutes

Biochemical test :-

1-catalase Negative and oxidase positive.
2-haemolysis patterns.

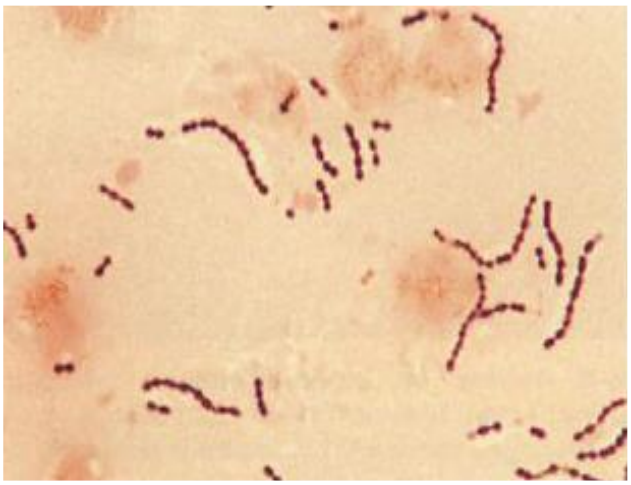
Table(2) Biochemical tests uses in the differential between types belong to *Streptococc*i

Species	Lancefield group	B-hemolysis	inulin	lactose	raffinose	salicin	sorbitol	trehalose
S. pyogenes	A	B	-	+	-	+	-	+
Streptococcus agalactiae	B	B(α,Y)	-	+	-	+	-	+
S. dysagalactiae subsp . dysagalactiae	C	α(B,y)	-	+	-	-	-	+
S.equi subsp. equi	C	B	-	-	-	+	+	-
S. uberis	none	α(Y)	+	+	-	+	+	+
S.bovis	D	α	+	+	+	+	-	V
*S.*pneumoniae	none	α	+	+	+	V	-	+
Enterococcus faecalis	D	α (B,y)	-	+	-	+	+	+

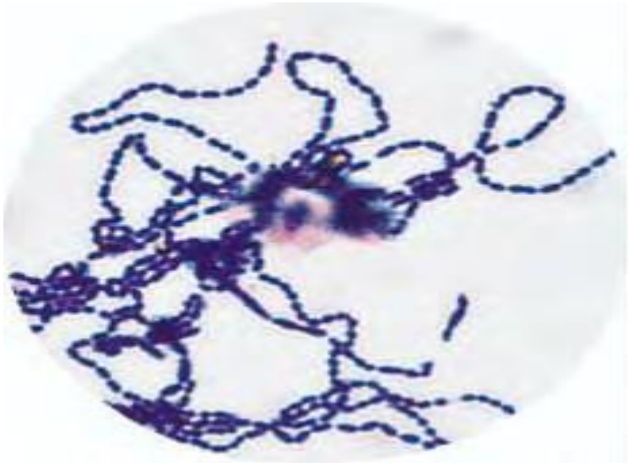
Diagnosis :-

1- depending on the pathological condition of the cause and animal types.
2- Smears from pus, milk samples can be fixed and stained by the gram stain and seen the chain arrangrment.
3- Inoculating the specimen on the blood agar and selective media for streptococci.

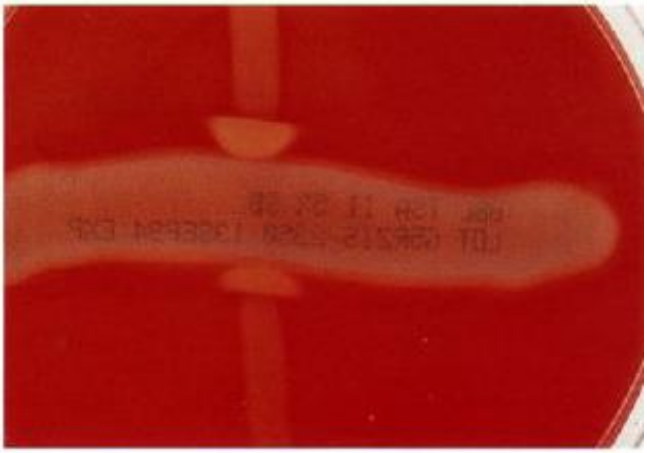
4- Recognized it by lancefield groups.
5- Identification by using biochemical tests.



Gram stain of streptococci in broth culture



Gram stain of streptococci in broth culture



CAMP test

colonies of streptococci on blood agar