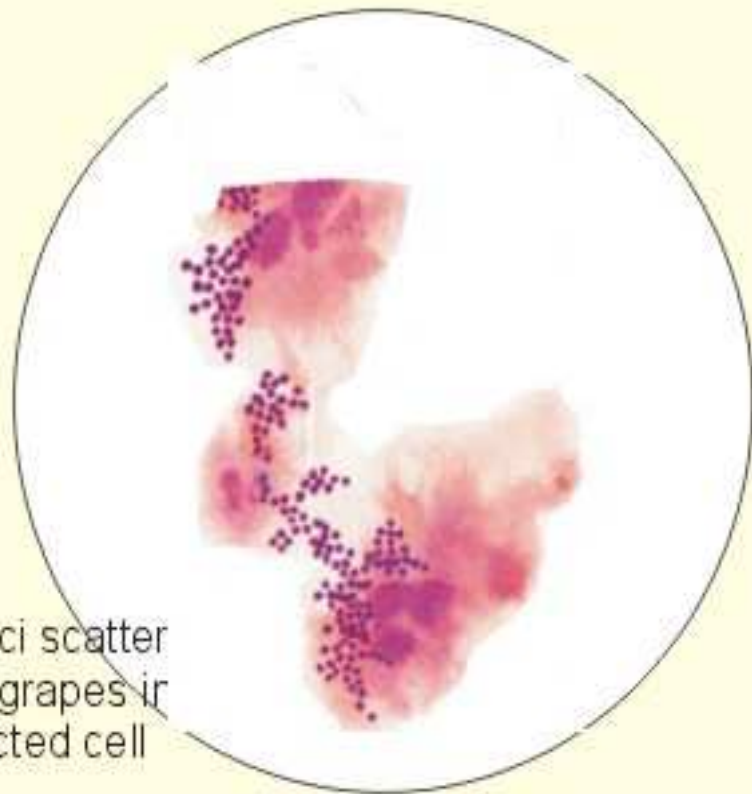


Staphylococcus

- Staphylococci, Micrococci and Stomatococcus belong to the family Micrococcaceae.
- They are gram positive spherical cocci, characteristic clumps of varying size, fancifully likened to bunches of grapes (Greek **Staphyle**, a bunch of grapes.)

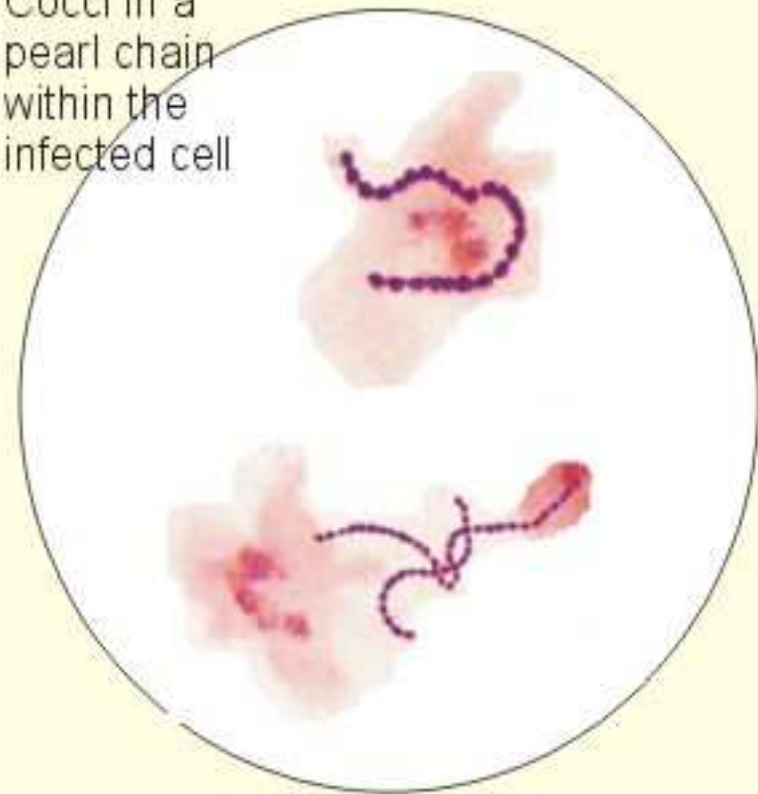
Microscopic Pictures Of Cocci



Cocci scatter
like grapes in
infected cell

Stafylococci in pus
Therapy: Penicillin

Cocci in a
pearl chain
within the
infected cell



Streptococci in pus
Therapy: Phenoxy-
methylpenicillin

History

- Von Recklinghausen(1871): first observed staphylococci in Pyogenic lesions.
- Pasteur(1880) : obtained liquid cultures from the cocci and produced abscesses by inoculating them into rabbits.
- Sir Alexander Ogston(1880) : established conclusively the causative role of coccus in various suppurative lesions.

Classification



Micrococcaceae

Planococcus

Non pathogenic

Found in marine

Environment

Stomatococcus

only one species

S.mucilogenus

(capsulated)

Staphylococcus

Micrococci

8 different

pathogenic species

Staphylococcus aureus

- Morphology and Cultural characteristics :
Spherical cocci, 1micrometer in diameter,
arranged in grape like clusters
- Non motile, non sporing.
- Grow readily on ordinary media with a temp.
range of 10-42⁰C, optimum being 37⁰C.
- They are aerobe and facultative anaerobe.

Cultural characteristics

- In liquid media, singles, pairs and short chains are also seen.
- On blood agar or nutrient agar, incubated for 24 hrs., it forms colonies 1-3 mm in diameter although dwarf colonies are not uncommon.
- Colonies are smooth, low convex, glistening, densely opaque and of butyrous consistency, surrounded by narrow zone of hemolysis on blood agar.
- Occasional strains are capsulated.

Narrow zone of hemolysis on blood agar

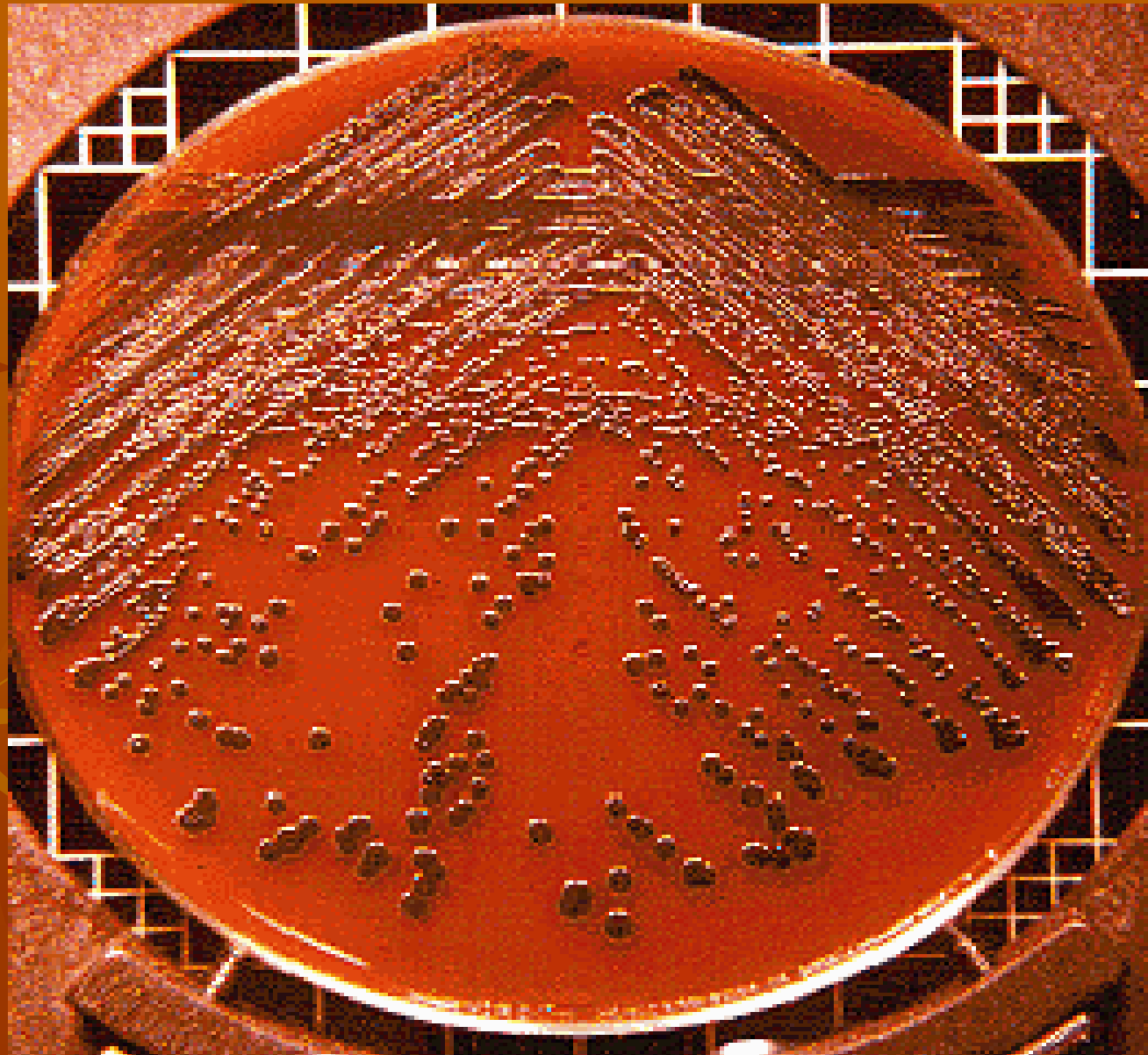


Cultural characteristics Cont..

- Most strains produce golden yellow pigment, though some may be white, orange or white. The pigment does not diffuse in to the medium. Pigment production is enhanced on fatty media such as Tween agar, or incorporated with milk or glycerol monoacetate.
- Selective media : Salt-milk agar, Salt cooked meat broth(8-10% nacl conc.), Lithium chloride and tellurite(Ludlam`s medium), Mannitol salt agar(7.5% salt conc.)

Mannitol salt agar





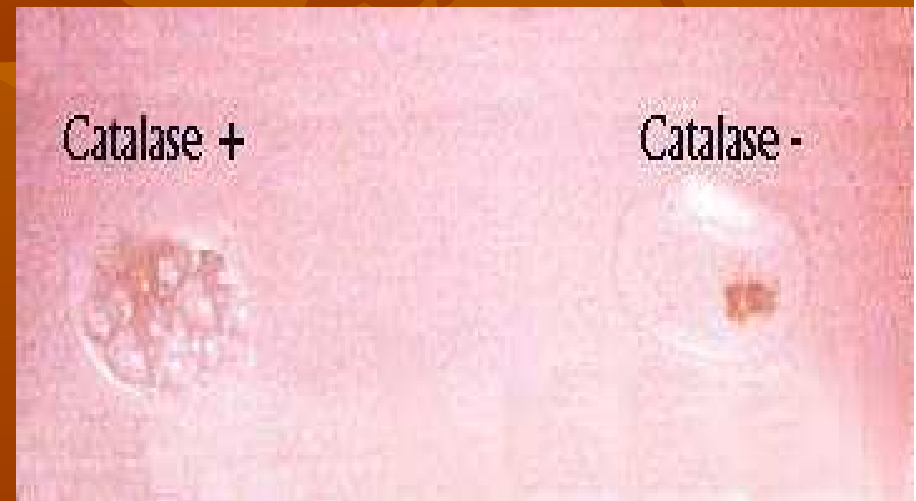
Black colonies of *staphylococcus aureus* on tellurite blood agar

Biochemical Reactions

- Catalase positive ,Coagulase positive
- Ferment Mannitol
- Urease positive, nitrate reduction positive
- Liquefy gelatin, MR and VP +ve, Indole –ve
- Produce Phosphatase
- Reduce tellurite to form black colonies on potassium tellurite medium

Catalase

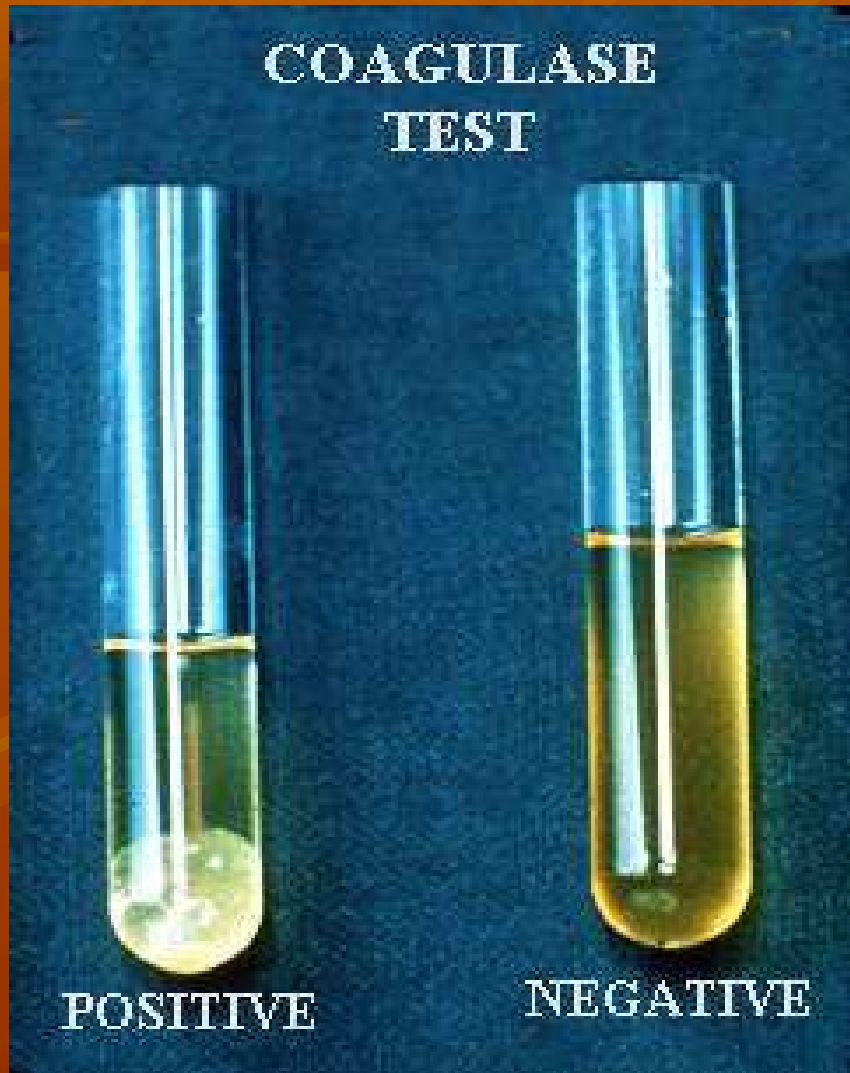
- The catalase test involves adding hydrogen peroxide to a culture sample or agar slant. If the bacteria in question produce catalase, they will convert the hydrogen peroxide and oxygen gas will be evolved. The evolution of gas causes bubbles to form and is indicative of a positive test.



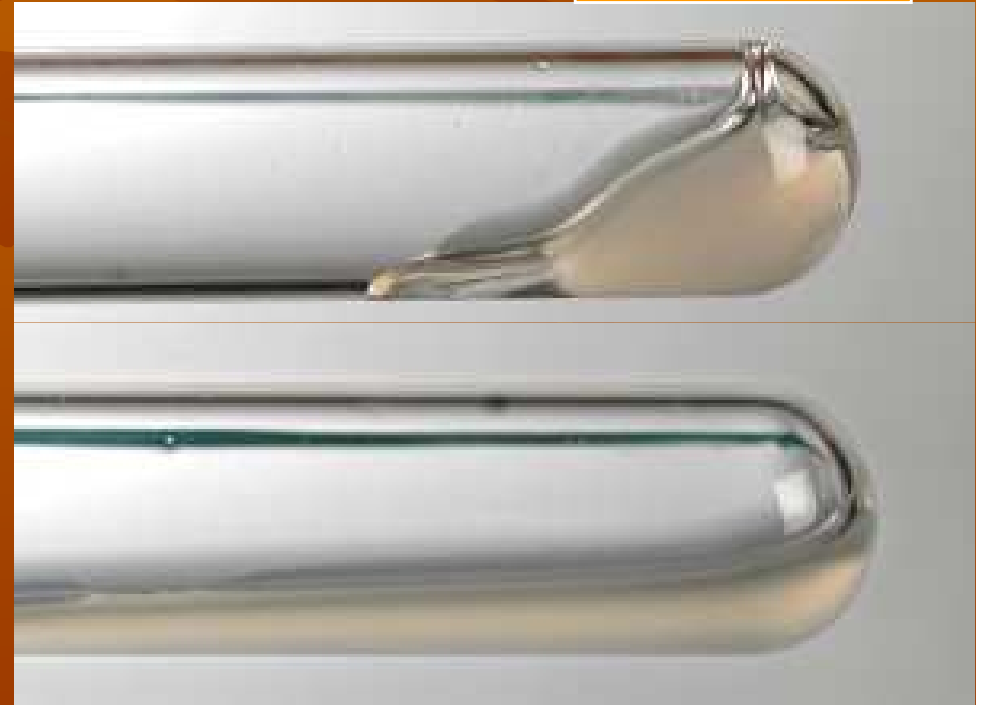
Tube Coagulase test

- the coagulase test is another method for differentiating between pathogenic and non-pathogenic strains of *Staphylococcus*. Bacteria that produce coagulase use it as a defense mechanism by clotting the areas of plasma around them, thereby enabling themselves to resist phagocytosis by the host's immune system.
- The sample in question is usually inoculated onto 0.5 ml of rabbit plasma and incubated at 37 °C for one to four hours. A positive test is denoted by a clot formation in the test tube after the allotted time.

Tube coagulase



positive



negative

Resistance

- *S.aureus* is among the hardiest of the non-sporing bacteria and survive well in the environment under both moist and dry conditions. It withstands moist heat at 60°C for 30 min.
- Readily killed by phenolic and hypochlorite disinfectants at standard in-use conc. And antiseptic preparations such as chlorhexidine and povidone-iodine.

Penicillin resistance

- Three types :
 - 1) Production of beta-lactamase (penicillinase) which inactivates penicillin by splitting the beta lactum ring.
 - 2) Changes in bacterial surface receptors, mutation in PBP-2. This change is chromosome in nature and is expressed at 30°C. These strains have been called Epidemic methicillin resistant S.aureus (EMRSA)
 - 3) Development of tolerance to penicillin

Pathogenicity and virulence

- Cell associated polymers :
 - 1) Cell wall polysaccharide
 - 2) Teichoic acid, facilitates adhesion of the cocci to the host cell surface
 - 3) Capsular polysaccharide inhibits opsonisation.

Cell Surface Proteins

- 1) Protein A : present on *S.aureus* strains has many properties as chemotactic, antiphagocytic and anti complementary.

Coagglutination: It binds to the Fc terminal of IgG molecule leaving the Fab region free to combine with its specific antigen producing agglutination.
e.g. Streptococcal grouping and gonococcal typing.

- 2) Clumping factor, bound coagulase which is responsible for slide coagulase.

Extra cellular Enzymes

- 1) Free Coagulase
- 2) Lipases
- 3) Hyaluronidases
- 4) Nuclease
- 5) Protein receptors

Toxins

- Cytolytic toxins :

- 1) Alpha hemolysin: protein inactivated at 70°C, but reactivated at 100°C lyses rabbit RBCs and leucocidal, cytotoxic, dermonectrotic and neurotoxic.

- 2) Beta hemolysin

- 3) Gamma hemolysin

- 4) Delta hemolysin

- 5) Leucocidin

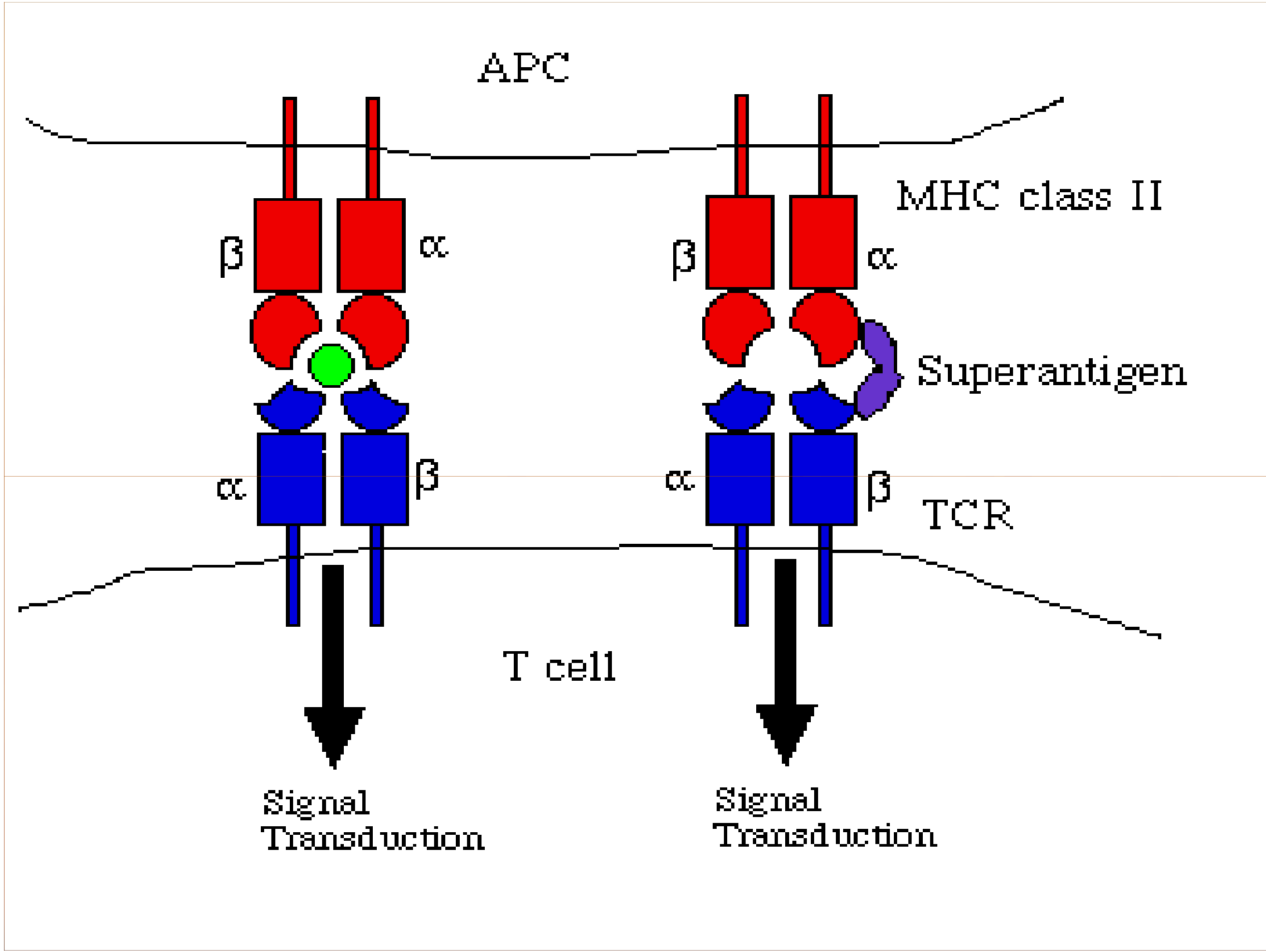
Exfoliative(epidermolytic) toxin : produce SSSS.

Enterotoxin

- Responsible for Staphylococcal food poisoning-nausea, vomiting and diarrhea 2-6 hrs after consuming contaminated meat, fish or milk containing preformed toxin. Self limited.
- Eight types A,B,C₁₋₃,D,E and H.Type A toxin is responsible for most cases.
- The toxin is believe to act upon directly on autonomous nervous system.

Toxic Shock syndrome toxin (TSST)

- Toxic shock syndrome is a fatal multisystem disease presenting with fever, hypotension myalgia, vomiting and an erythematous rash.
- First identified in the USA in menstruating women using intravaginal tampons.
- Staphylococcal enterotoxins and TSST-1 are super antigens.



Staphylococcal diseases

- Staphylococcal infections are by far the most common bacterial pathogens. They are characteristically localized Pyogenic lesions and are able to cause infections only if they enter through breaks in the skin. Infections are as follows:
 - 1) Pyogenic infections: folliculitis, impetigo, furuncles, carbuncles, breast abscess etc..
 - 2) Disseminated infections
 - 3) Toxin mediated illnesses: toxic shock syndrome, staphylococcal scalded skin syndrome, staphylococcal food poisoning

Bacteriophage Typing

- Staphylococci may be typed, based on their susceptibility to bacteriophages.



Laboratory diagnosis

- The specimens to be collected depend on the type of the lesion.

- Direct microscopy → Culture on basal media

Antibiotic sensitivity

Coagulase
test

Use selective
media



Other coagulase positive staphylococci

- *S.intermedius*
- *S.hyicus*

Coagulase negative staphylococci

- Constitute a major component of the normal flora of the human body.
- **S.epidermidis**: It is nonpathogenic ordinarily but can cause disease when the host defenses are breached. It has a predilection for growth on implanted foreign bodies with formation of biofilms such as artificial valves, shunts, I.V. catheters and prosthetic appliances leading to bacteremia.
- **S.saprophyticus**: may be present on normal human skin and the periurethral area and can cause UTI, particularly in sexually active young women. It is Novobiocin resistant.

Micrococci

- Gram positive cocci, mostly in tetrads
- Catalase and DMSO-oxidase positive
- Strict aerobe by which can be differentiated by Hugh and Leifson`s oxidation-fermentation test.