Streptococci

Streptococci are Gram-positive, nonmotile, non-spore forming, catalase-negative cocci that occur in pairs or chains. Most streptococci are facultative anaerobes, and some are obligate (strict) anaerobes. Most require enriched media (blood agar).

Classification of Streptococci

Streptococci can be classified according to:

1- Hemolysis on blood agar

Alpha hemolysis

When α -hemolysis is present, the agar under the colony is dark and greenish. *Streptococcus pneumoniae* and a group of oral streptococci (*Streptococcus viridans* or viridans streptococci) display alpha hemolysis. This is sometimes called green hemolysis because of the color change in the agar. Other synonymous terms are incomplete hemolysis and partial hemolysis. Alpha hemolysis is caused by hydrogen peroxide produced by the bacterium, oxidizing hemoglobin to green methemoglobin.

Beta hemolysis

Streptococcus pyogenes, or Group A Streptococci (GAS), and Streptococcus agalactiae, or Group B Streptococci (GBS) on blood agar cultures display beta hemolysis. Beta hemolysis (β -hemolysis), sometimes called complete hemolysis, is a complete lysis of red cells in the media around and under the colonies: the area appears lightened (yellow) and transparent.

Gamma hemolysis

If an organism does not induce hemolysis, the agar under and around the colony is unchanged, and the organism is called non-hemolytic or said to display gamma hemolysis (γ hemolysis). Enterococcus faecalis (formerly called Group D Streptococci) displays gamma hemolysis

2- Oxygen requirements

Obligate anaerobes – Pepto streptococci

Aerobic or facultative anaerobes - Streptococci

3- Serological (Lancefield Classification) (A – V)

Serologic grouping is based on antigenic differences in cell wall carbohydrates (groups A to V). Rebecca Lancefield developed the serologic classification scheme in 1933. β -hemolytic strains possess group-specific cell wall antigens, most of which are carbohydrates.



Streptococcus pyogenes (GAS)

It is 0.5 to 1 μ m in diameter and arranged in chain. Chain formation is due to cocci dividing in one plane only and failure of daughter cell to separate completely. It is usually encapsulated, non-sporing and non-motile. When capsule is present it is composed of **hyaluronic acid** or **polysaccharide** capsule.



Blood agar: After 24 hours' incubation colony is **small**, 0.5 to 1 mm (**pin point** colonies), **circular**, **transparent**, **low convex** with area of hemolysis.

Strains with capsules produce mucoid colonies. Virulent strains produce matted colonies (granular). A virulent strain produces glossy colonies.



Selective media: Blood agar medium having 1:500,000 crystal violet may be used as selective medium.



Bacitracin susceptibility

Streptococcus pyogenes can be differentiated from other non-group A β -hemolytic streptococci by their increased sensitivity to bacitracin. The bacitracin test, along with the Lancefield antigen A test, is used for greater specificity in the identification of *S. pyogenes*, since other β -hemolytic strains of streptococci that may contain the group A antigen are resistant to bacitracin.

Streptococcus agalactiae

Streptococcus agalactiae belongs to Lancefield group B and is the only species that carries the group B antigen. Human pathogenic group B strains possess a polysaccharide capsule which appears to confer virulence.

CAMP test (Christie, Atkins and Munch-Peterson)

The **CAMP** test is a test to identify group B β hemolytic streptococci (*Streptococcus agalactiae*) based on their formation of a substance (CAMP factor) that enlarges the area of hemolysis formed by the β hemolysin elaborated from *Staphylococcus aureus*.



	Hemolysis	Bacitracin sensitivity	CAMP test
S. pyogenes	β	Susceptible	Negative
S. agalactiae	β	Resistant	Positive

Streptococcus Pneumonia

Pneumococci are gram-positive cocci in pairs (diplococci). The cocci are about 1 μ m, slightly elongated cocci, with one end broad or rounded and the other pointed, presenting a **flame shaped or lanceolate appearance**. They may occur singly, in pairs, or in short chains but most often are seen as pairs (diplococci).



Culturing

Specimen is inoculated on plates of blood agar and heated blood agar incubated in air with 5-10 percent CO₂ for 18-24 hours. Typical colonies develop with α -hemolysis. The colonies are small (0.5-1 mm), dome shaped and glistening, with an area of green discoloration (α -hemolysis) around them.



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S. pneumoniae can be differentiated from the viridans streptococci, some of which are also alpha-hemolytic, using an optochin test, as *S. pneumoniae* is optochin-sensitive. *S. pneumoniae* can also be distinguished based on its sensitivity to lysis by bile, the so-called "bile solubility test". The encapsulated, Gram-

positive, coccoid bacteria have a distinctive morphology on Gram stain, lancetshaped diplococci. They have a polysaccharide capsule that acts as a virulence factor for the organism

	hemolysis	Optochin	Bile solubility	Inulin
		sensitivity		Fermentation
S. pneumoniae	α	Sensitive (≥ 14	Soluble	Not ferment
		mm)		
Viridans strep	α	Resistant (<13	Insoluble	Ferment
		mm)		
		mm)		