Chapter 15

Gram positive cocci

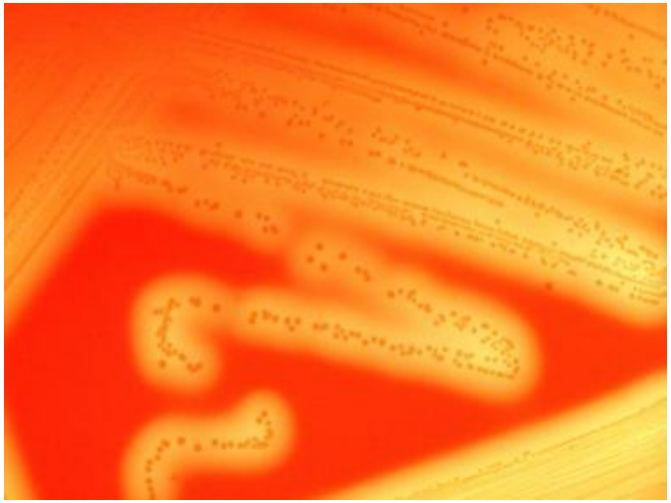
Streptococcus pyogenes (contd)

Laboratory diagnosis

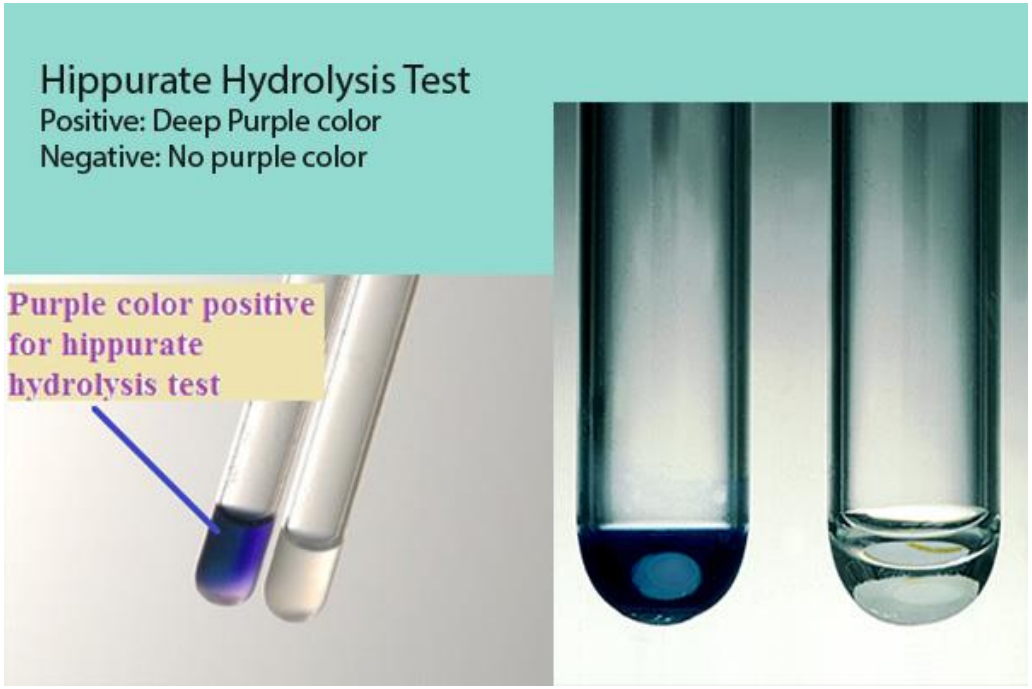
Mirobiology

-- In **pharyngitis**, Gram stained smears are useless because ***Strept. pyogenes*** cannot be differentiated, but those from **skin lesions** or **wounds** are usually diagnostic.

-- **beta-hemolytic**, small, translucent colonies on blood agar produced in 18-48 hours and **inhibited by bacitracin** are indicative.



-- ***Strept.agalactiae*** is differentiated by its **ability to hydrolyze hippurate** and by **enhanced hemolysis** on sheep blood agar when combined with β-hemolysin of ***Staph. aureus*** (**CAMP test**).



-- **Group D streptococci** (type species *Strept. faecalis*) hydrolyze **esculin** in the presence of bile (black pigment on bile-esculin agar).

-- Further differentiation is done by **growth in 6.5% NaCl**. **Enterococci** can grow, others cannot.

-- Because culture takes time, **rapid tests** may have to be used.

-- In case of ***Strept. pyogenes***, antigens are extracted from **throat swabs** and reacted with **latex bound antibodies**. **Agglutination** indicates positive reaction.

-- Specificity is acceptable but sensitivity is low so in case of negative result, culture should be done. Similar test is available for ***Strept.agalactiae.***

Serology

-- **ASO titres are high** soon after ***Strept. pyogenes*** infection.

-- In suspected cases of **rheumatic fever**, elevated titres are indicative of previous infection because throat cultures are often negative.

-- **Titres of anti-DNase B** are high in group A streptococcal infections and serve as an indicator of previous streptococcal infection in patients suspected of having **AGN**.

Treatment

-- ***Strept. pyogenes*** infections can be treated with **penicillin G** or **amoxicillin** but **rheumatic fever and AGN are not affected** after onset.

-- In **mild infections**, **penicillin V**, or in case of **allergic patients**, **erythromycin** or **azithromycin** may be used. **Clindamycin** may also be used.

-- In general, streptococci are **not resistant to penicillins**.

-- **Endocarditis caused by viridians streptococci** is **curable** with **prolonged penicillin therapy**.

-- **Enterococci** resistant to **multiple drugs** (aminoglycosides, penicillins and vancomycin) have emerged.

-- **Vancomycin resistant** strains are problematic and mechanism of resistance is same as ***Staph. aureus***, i.e., production of enzymes which substitute D-lactate for D-alanine in peptidoglycan.

-- These strains can be treated with **linezolid** and **daptomycin**.

-- ***Strept.agalactiae*** can generally be treated with **penicillin G** or **ampicillin**, sometimes in combination with aminoglycosides.

-- ***Strept.bovis*** and **peptostreptococci** can be treated with **penicillin G**.

Prevention

-- **Rheumatic fever** can be prevented by **prompt diagnosis** and **treatment of streptococcal pharyngitis**. Special care is needed for patients known to have rheumatic fever in past.

-- In patients with **damaged heart valves** undergoing invasive dental procedures, **preventive use of amoxicillin preoperatively** is recommended to ward off viridians streptococci.

-- In infections by ***Strept. agalactiae***, neonatal sepsis can be reduced.

-- If vaginal and rectal cultures of women with 35-37 weeks' gestation are positive, penicillin G (or ampicillin) should be administered i.v.at the time of delivery.

Streptococcus pneumoniae

Diseases

-- ***Steptococcus pneumoniae*** or **pneumococcus** causes **pneumonia, bacteremia, meningitis**, and infection of upper respiratory tract such as **otitis media**, and **sinusitis**.

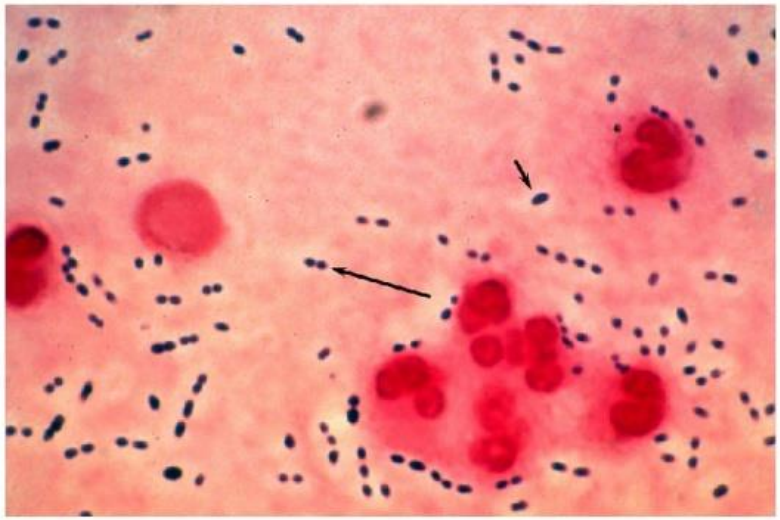
-- It is also a common cause of **conjunctivitis**.

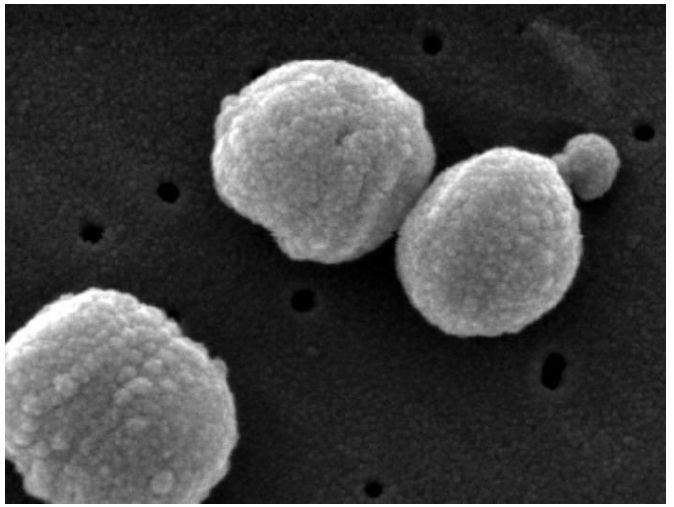
Important properties

Streptococcus pneumoniae cells are **Gram-positive**, **lancet-shaped cocci**.

These are usually **diplococci**, but they may also occur singly and in short chains.

They **do not form spores**, and are **non motile**.



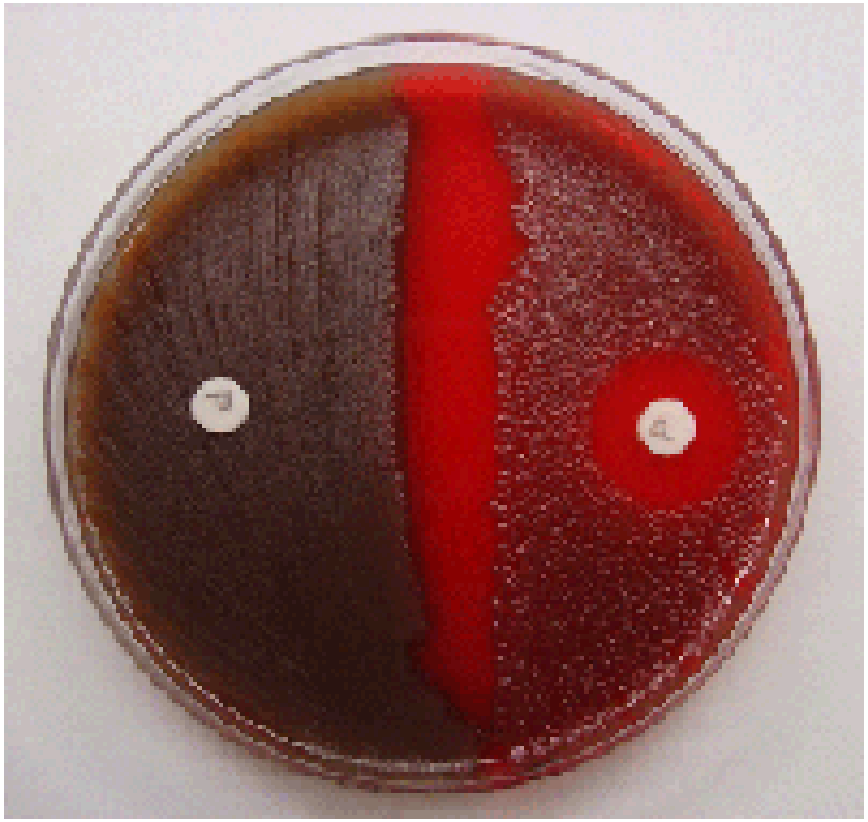


-- ***Streptococcus pneumoniae*** grows best in **5% carbon dioxide**.

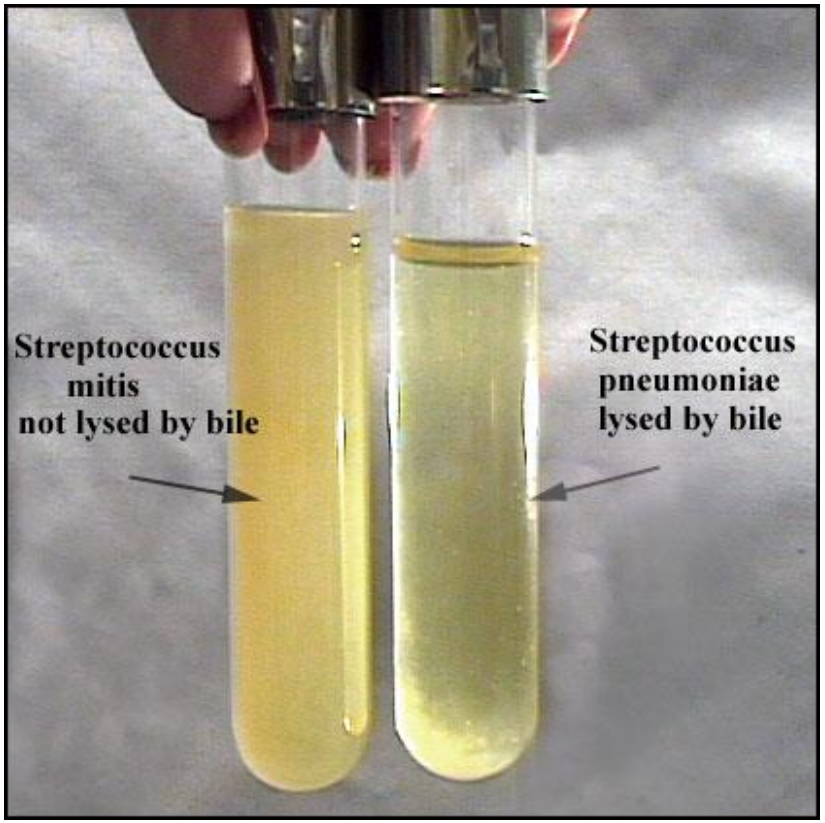
-- On **blood agar**, colonies characteristically produce **α (green) hemolysis.**

-- This differentiates them from ***S. pyogenes*** but not from **viridans streptococci.**

-- Special tests such as **inulin fermentation, bile solubility**, and **optochin** (an antibiotic) sensitivity must be routinely employed to differentiate.



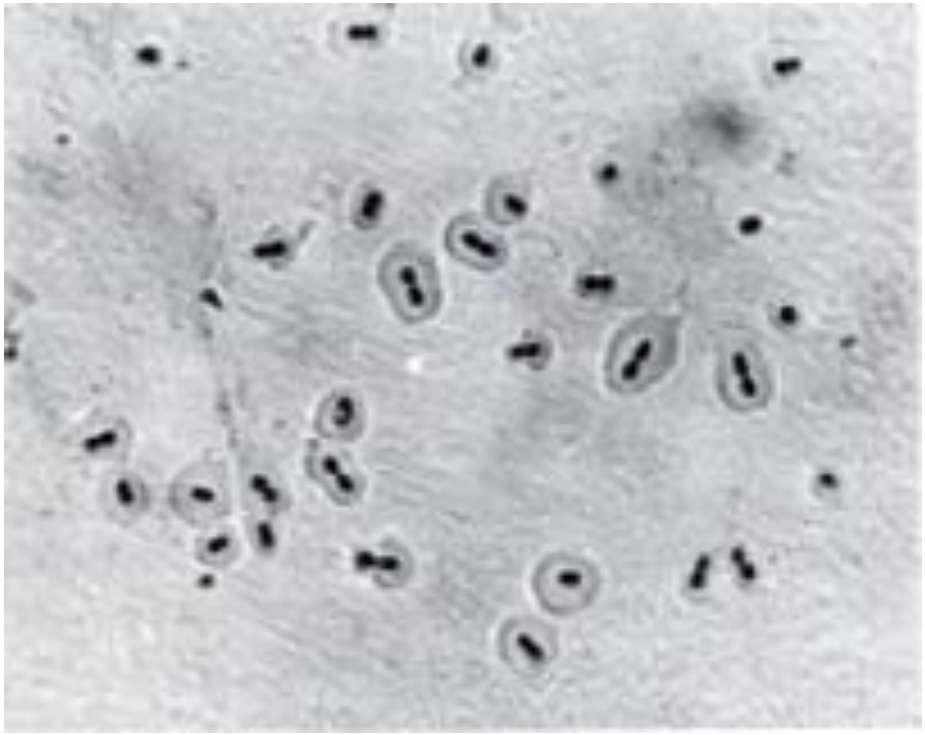
Optochin sensitivity test



Bile sensitivity test

Pneumococci possess **polysaccharide capsule** of more than 85 antigenically distinct types.

The **Quellung reaction** (swelling reaction) forms the basis of serotyping and relies on the swelling of the capsule upon binding of homologous antibody.



-- **Capsule** is a **virulence factor** and specific antibody osponizes the organism, facilitates phagocytosis, and promotes resistance.

-- The antibody develops due to infection or vaccination.

-- Another important surface component, **C-substance** or **C-polysaccharide** is a teichoic acid in cell wall.

-- It reacts with **CRP** which is elevated **up to 1000 times** in acute inflammation. It is a non-specific indicator.

Transmission

-- Humans are natural host and there is **no animal reservoir**. About 5-50% of normal population harbor pneumococci.

-- Disease usually does not occur in healthy persons.

Pathogenesis

-- **Capsule** is the most important virulence factor.

-- **Lipoteichoic acid** activates complement and induces inflammatory cytokine production and **septic shock syndrome** in immunocompromised individuals.

-- **Pneumolysin**, cause of **α-hemolysis** may also contribute to disease.

-- Pneumococci produce **IgA protease** to enhance colonization of mucosa of upper respiratory tract.

-- Pneumococci **multiply in tissues** and cause inflammation.

Pneumococcal infection may be helped by:

1. **Alcohol** or **drug intoxication** that depress cough reflex and increase aspiration of secretions.
2. **Pooling of mucus**, **bronchial obstruction**, and **injury irritants**.
3. **Pulmonary congestion** and **heart failure**
4. **Splenectomy**
5. Chronic diseases such as **sickle cell anemia** and **nephrosis**.
6. **Trauma to head** that causes leakage of spinal fluid.

Clinical findings

-- **Pneumonia** often begins with a **sudden chill, fever, cough**, and **pleuritic pain**.

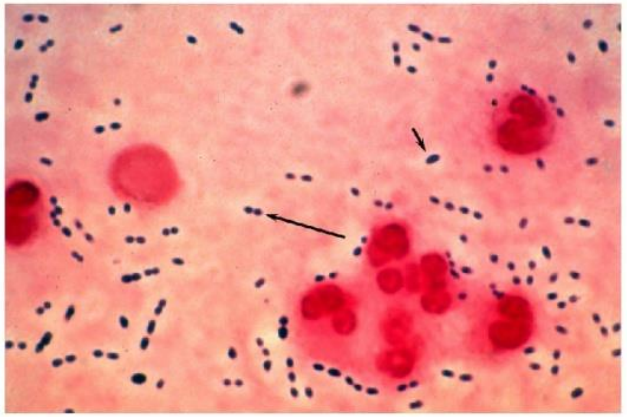
-- **Sputum** is a **red or brown** “rusty” color.

-- **Bacteremia** occurs in 15-25% cases.

-- **Spontaneous recovery**, associated with production of **anticapsular antibodies**, begins after 5-10 days.

Laboratory diagnosis

-- **Gram stained smears of sputum** show them as **lancet shaped Gram positive diplococci**.



-- **Quellung reaction** with multitype antiserum is useful.

-- Small **α-hemolytic colonies** on blood agar which are **bile soluble** and **sensitive to optochin** are almost diagnostic.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Streptococcus pneumoniae on Columbia Sheep Blood Agar

-- **Blood cultures** are positive in **15-25%** cases.

-- In **meningitis**, **culture of CSF** is usually **positive**.

-- **Rapid diagnosis** can be made by **detecting capsular polysaccharide** by **latex agglutination test**.

-- **Antibiotic sensitivity tests** must be done on organisms isolated from serious infections.

- Latex agglutination for pneumococcal antigen in serum of patients

Negative reaction Positive reaction

Pathology Basics
Notes and more.....
By Dr. Ashish Jawarkar

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Treatment

-- Generally, **pneumococci** are **sensitive to penicillins** and **erythromycin/azithromycin.**

-- In **severe infections, penicillin G** is of choice; **in mild, penicillin V** may be used.

-- A fluoroquinolone such as **levofloxacin** may also be considered.

-- Recently, **high level resistance to penicillins** (up to 35% isolates) has started to emerge **due to changes in PBP** without production of β-lactamase.

-- **Vancomycin** is the drug of choice in **very serious cases**.

-- **Less severely ill** cases may be given **ceftriaxone** or **levofloxacin**.

-- **Strains resistant to all mentioned drugs have emerged**.

Prevention

-- In spite of availability of antimicrobials, **mortality rate is high** in **elderly** (above 65 years), **immunocompromised** (esp. splenectomized) or **debilitated** persons.

-- Such persons should be **immunized** with **polyvalent (23 type)** polysaccharide vaccine.

-- **Oral penicillin** is given to **young children** with **hypogammaglobulinemia** or **splenectomy** because they are prone to pneumococcal infection and **respond poorly to the vaccine**.

-- **Conjugate vaccine** (with diphtheria toxoid) is given to children **younger than 5 years**.

-- It **prevents both bacteremic (e.g. meningitis)** and **mucosal (e.g. otitis media) infections**.

-- The **latest conjugate vaccine** includes **13 serotypes** to cover the problem of serotype replacement.

-- Immunization of children provides **herd immunity** to adults also, as they are the main carriers.

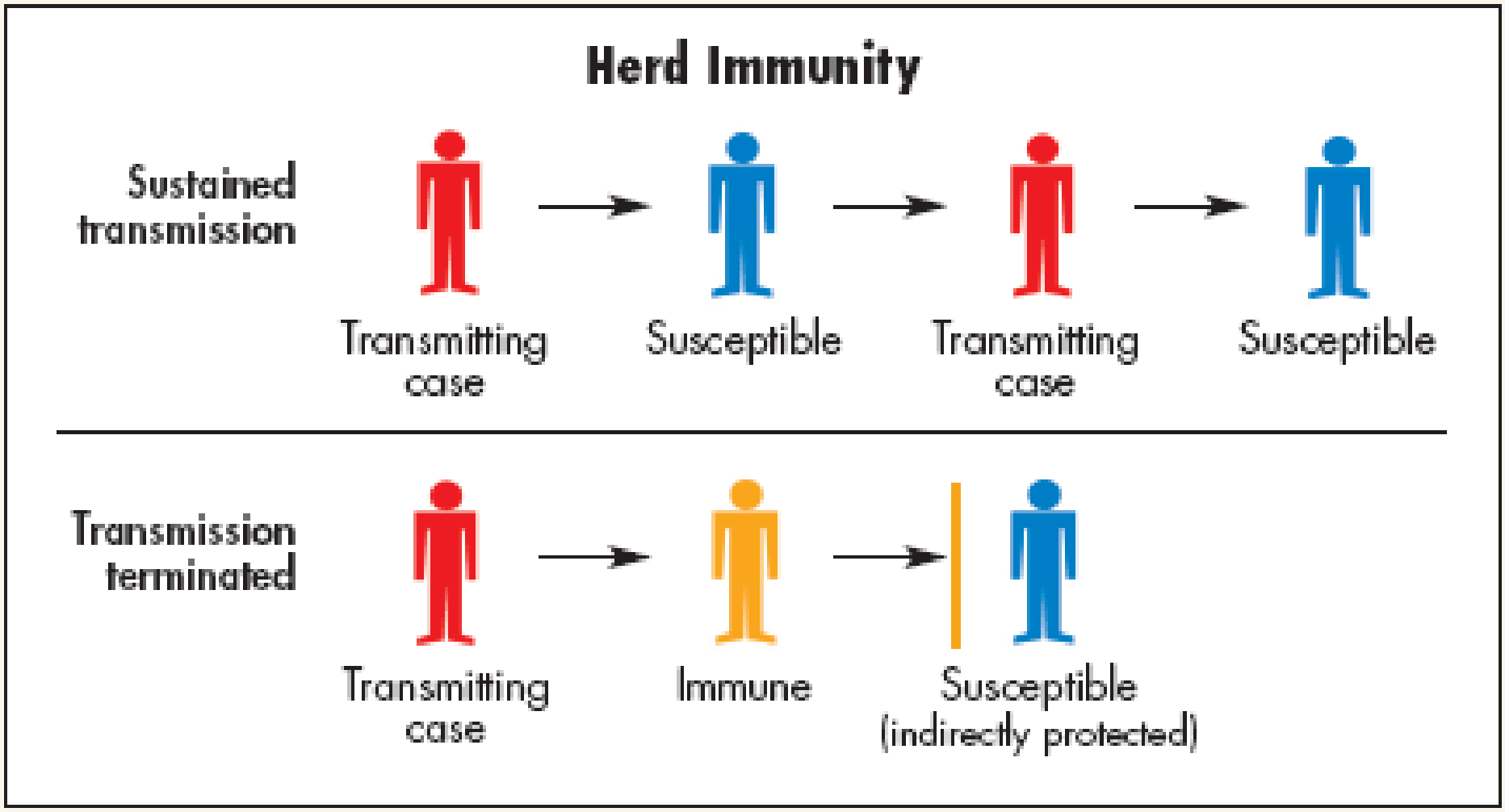

Figure – The principle underlying herd immunity is that the presence of enough immune persons in a community interrupts the transmission of an infectious agent, thereby providing indirect protection for unimmunized (or "susceptible") persons.


Thank You!