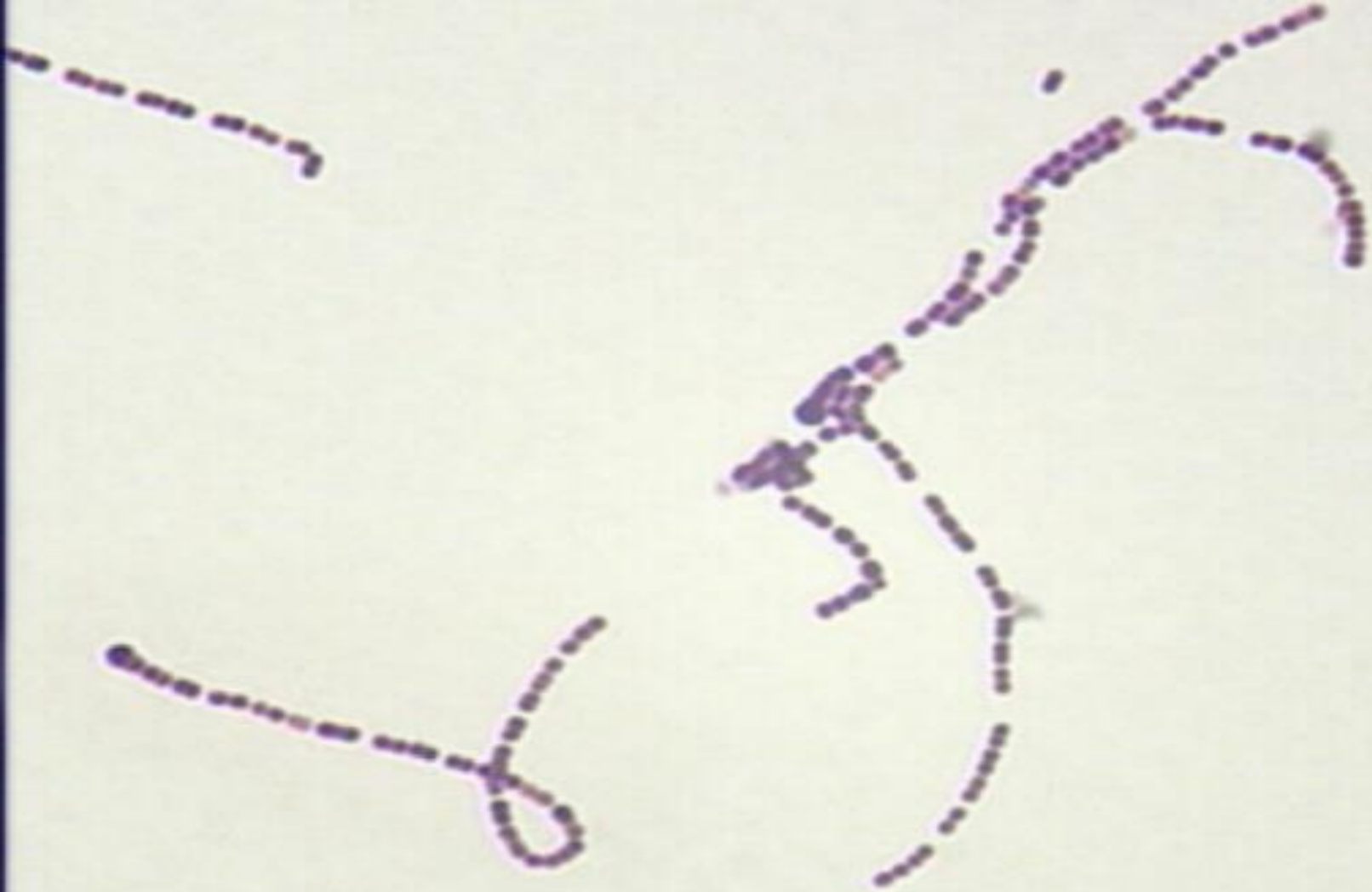


STREPTOCOCCUS

STREPTOCOCCUS

- **General Features:**
 - Gram positive cocci in pairs and chains
 - Catalase negative



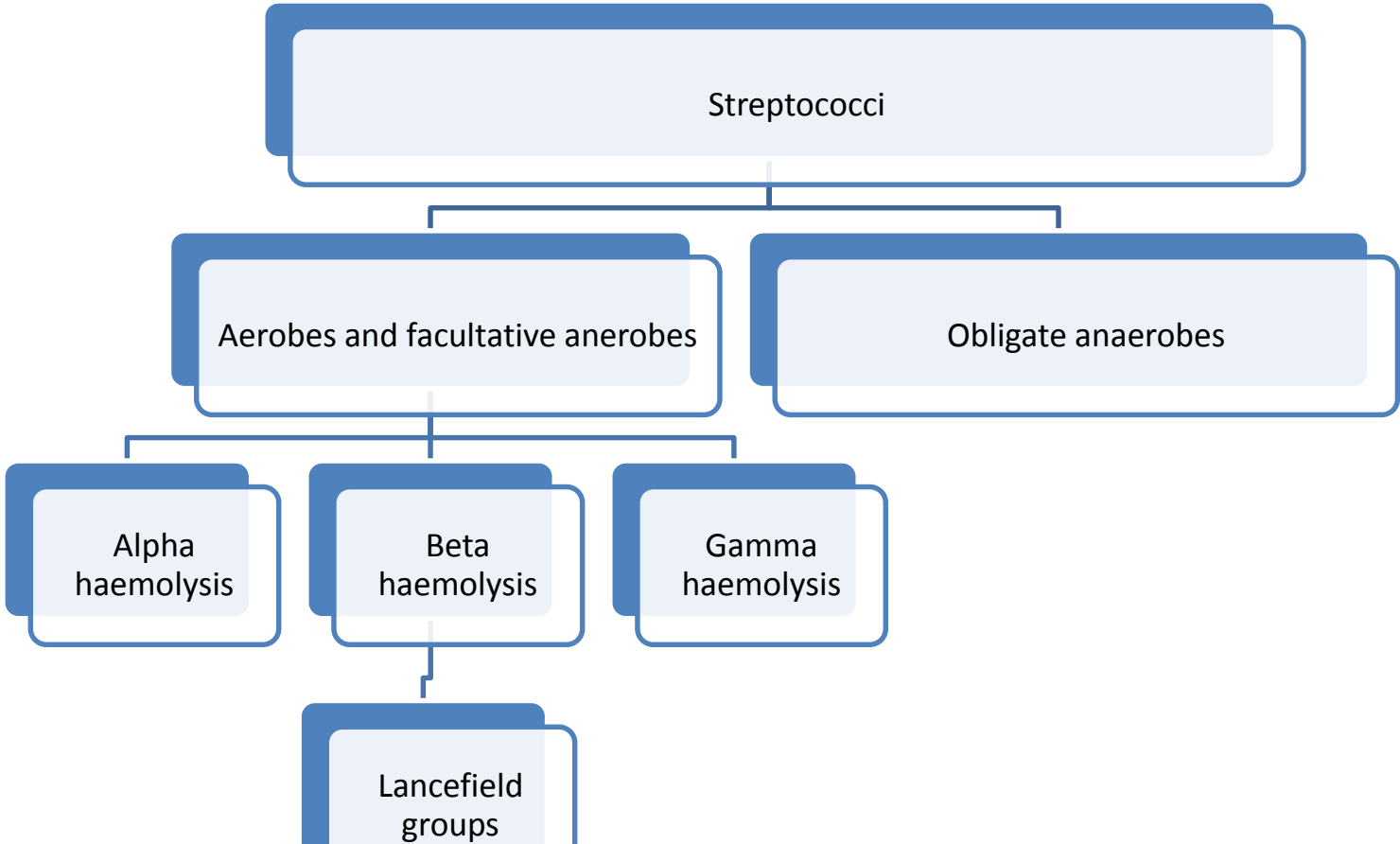
A microscopic image showing numerous clusters of small, dark, spherical bacteria. The clusters vary in size and shape, some appearing as tight spheres while others are more irregular. The background is a light, pinkish-tan color.

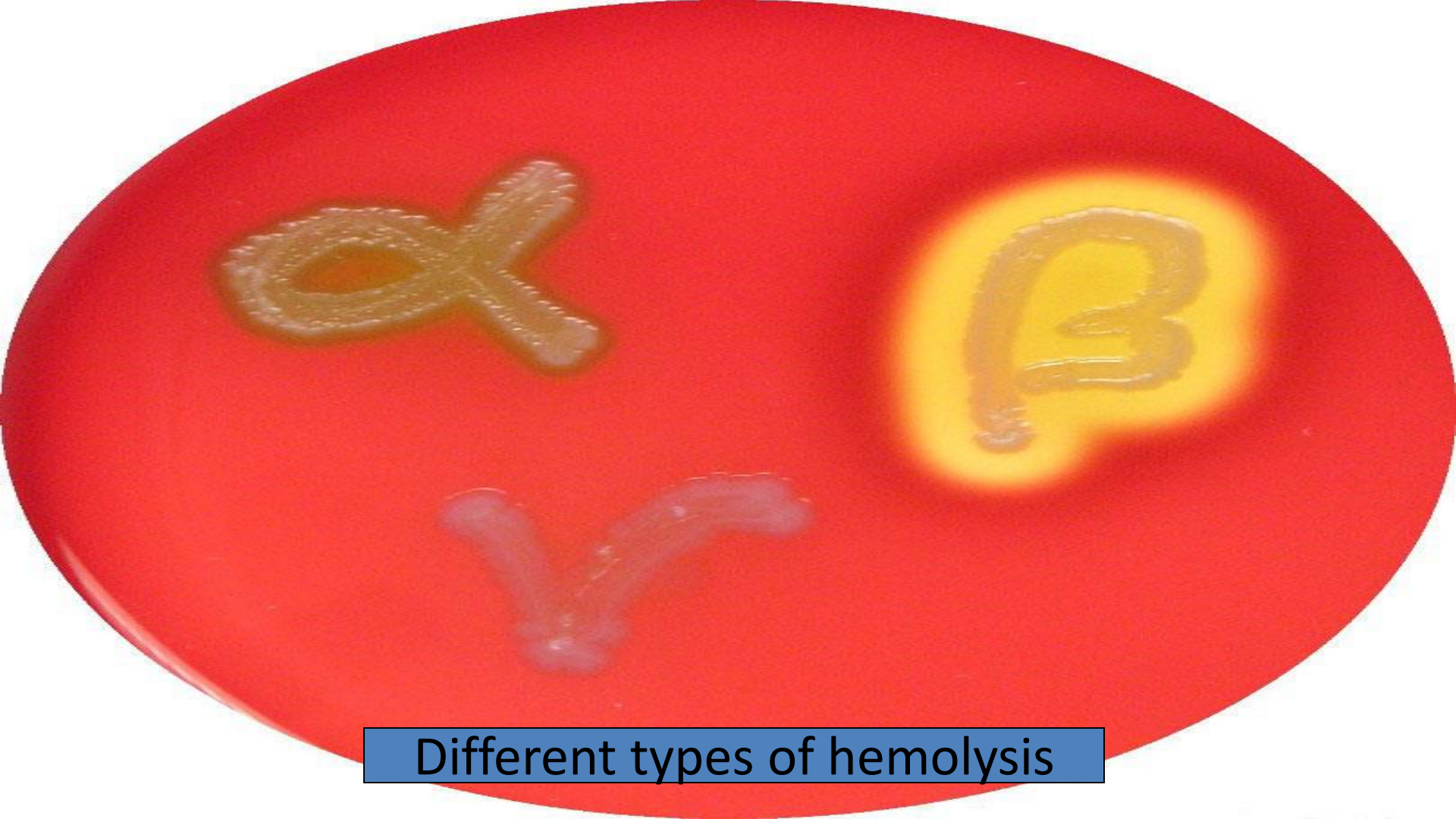
Staphylococci

A microscopic image showing chains of small, dark, spherical bacteria. The chains are of varying lengths and some are curved. The background is a light, pinkish-tan color.

Streptococci

Classification of Streptococcus





Different types of hemolysis

Streptococcus pyogenes

Morphology

- Size : 0.5-1.0 micrometer in diameter
- Shape : spherical or oval
- Arrangement : in chains, the length of which varies within wide limits, being longer in the liquid medium. Chain formation is due to the cocci dividing in one plane only.
- Non motile and non sporing
- Group A and C have capsules composed of hyaluronic acid while group B and D have polysaccharide capsules.

Cultural characteristics

- Facultative anaerobe
- Optimum temp. for growth is 37°C (range 22-42°C).
- Exacting in nutritive requirements, grows only in presence of glucose or serum.
- On blood agar, after incubation for 24hrs, the colonies are small (0.5 to 1.0mm), circular, semitransparent, low convex discs with an area of clear hemolysis around them.



Cultural characteristics cont.

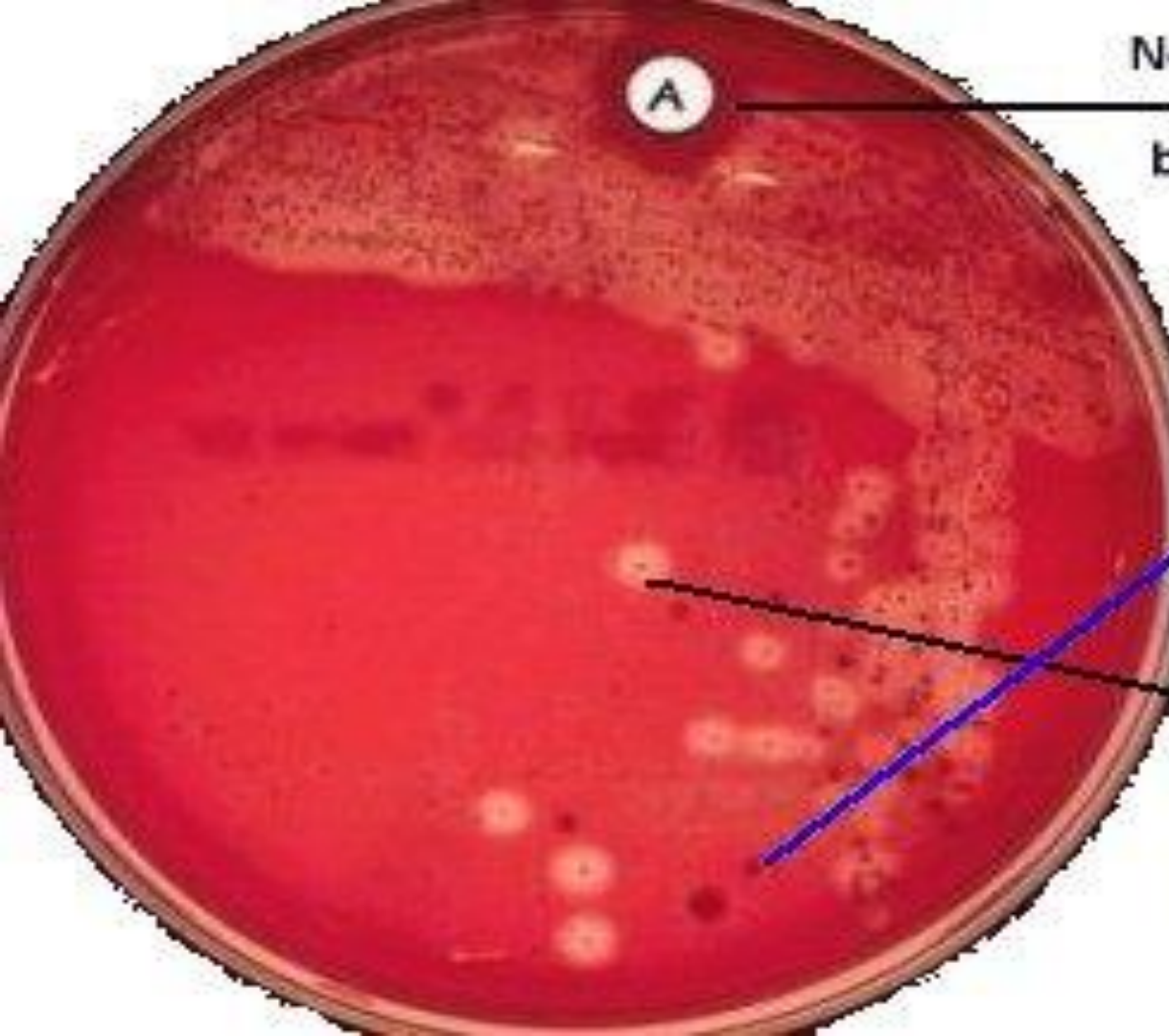
- Growth and hemolysis are promoted by 10% CO₂.
- In liquid media, growth occurs as a granular turbidity with powdery deposit.
- Colony types :
 - 1) Matt type** : fresh isolate of virulent strain (finely granular)
 - 2) Glossy type**: a virulent strains
 - 3) Muroid type**: capsular strain

Biochemical reactions

- Catalase negative and insoluble in bile like other streptococci
- Ferments variety of sugars in sugar serum peptone waters.
- *S.pyogenes* is positive in PYRase test, which distinguishes it from non-group A hemolytic streptococci.

Resistance

- Delicate organism, easily destroyed by heat (54° C for 30 minutes).
- Crystal violet (1mg/L), nalidixic acid (15mg/L) and colistin sulphate (10mg/L) added to blood agar provide a good selective medium.
- Sensitive to most antibiotics
- Sensitive to Bacitracin unlike other streptococci.



Notice the zone of growth inhibition around the bacitracin (Taxo A disc).

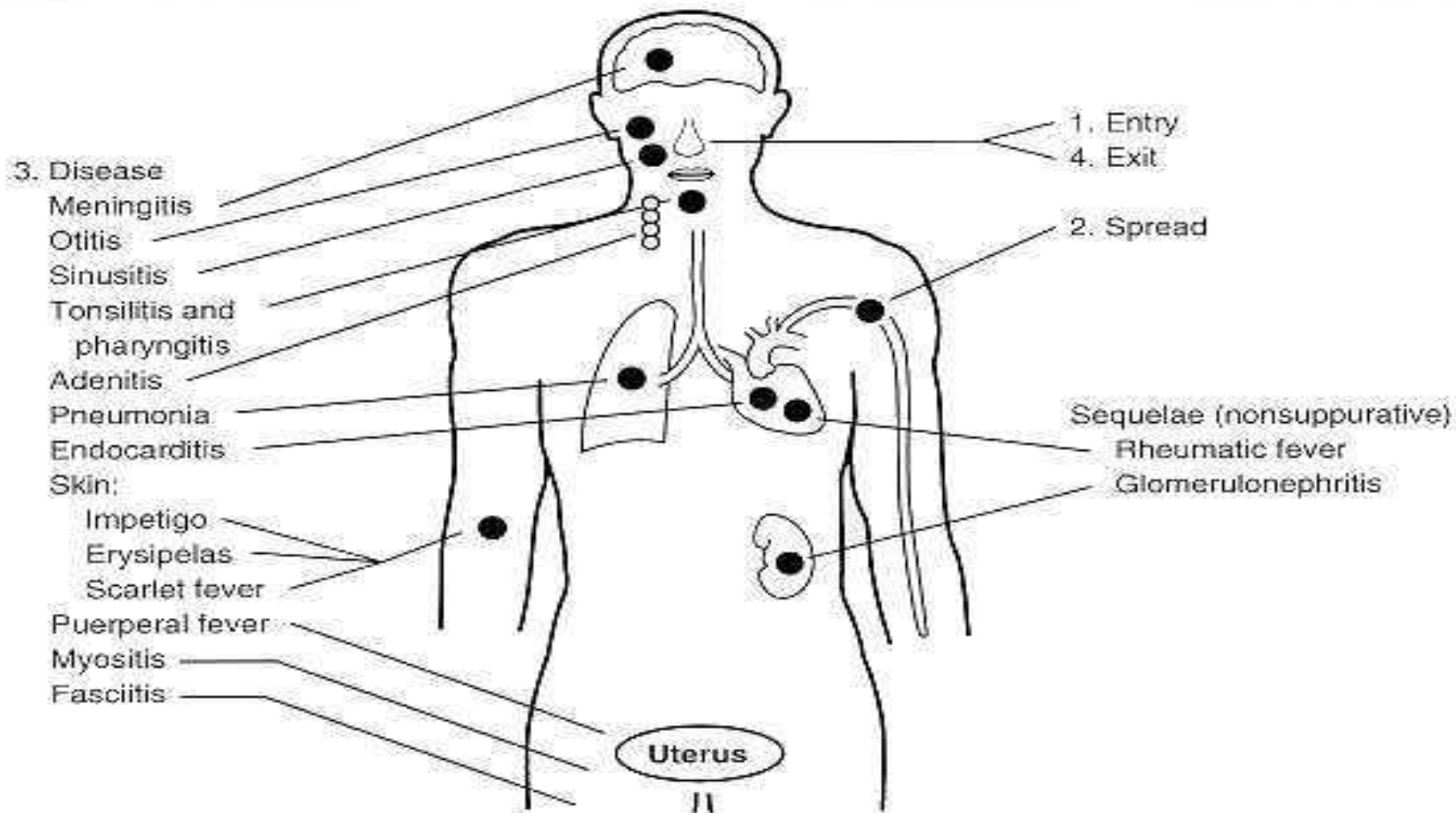
A throat culture taken from a 5 year-old with Streptococcal pharyngitis.

Notice the non-hemolytic bacterial colonies

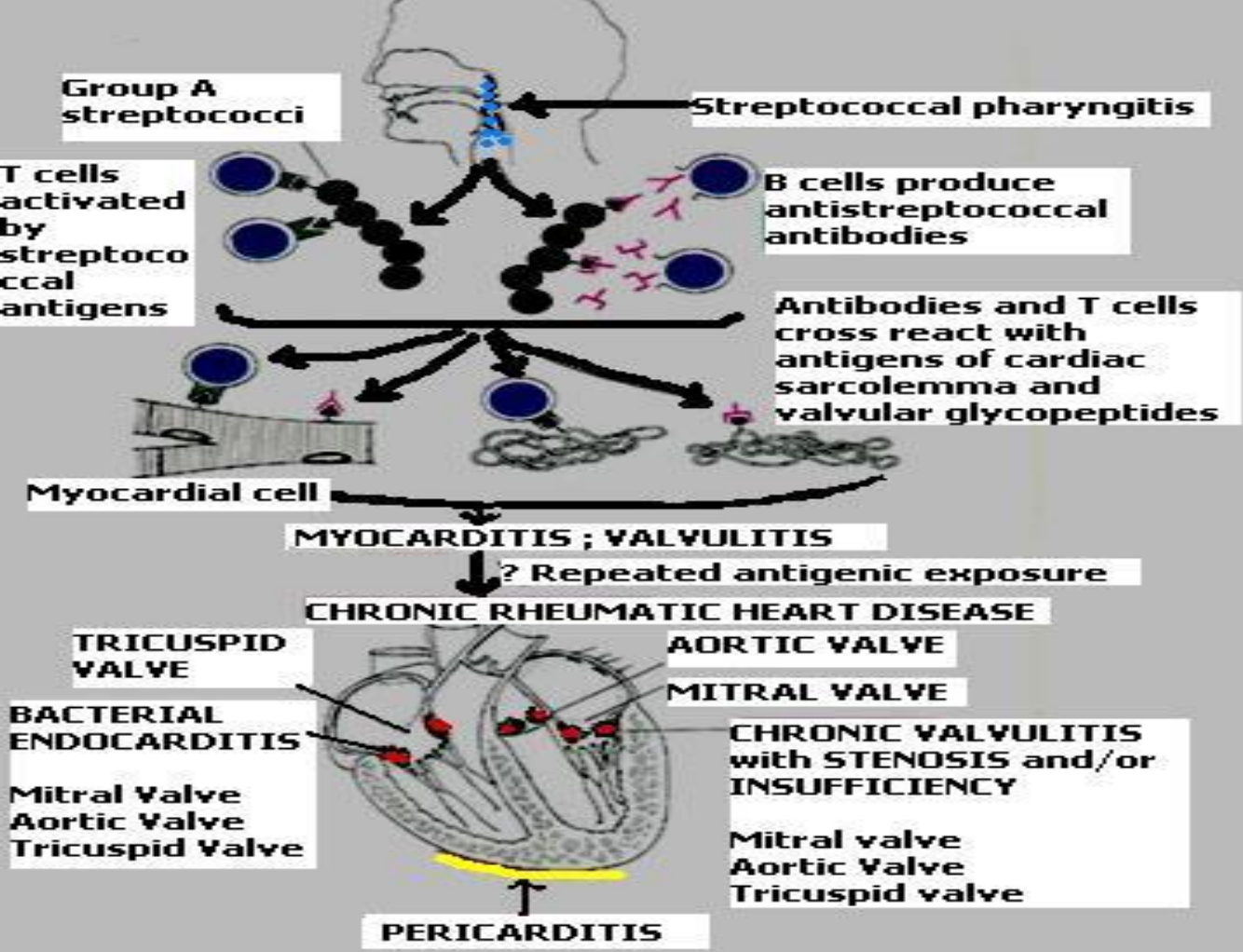
Notice the clear zone of hemolysis around the *Streptococcus pyogenes* grown on a blood agar plate.

Table 22.2: Suppurative and non-suppurative manifestations of *Streptococcus pyogenes*

Suppurative	Non-suppurative
Respiratory infections: <ul style="list-style-type: none">• Pharyngitis/sore throat• Pneumonia• Empyema	Acute rheumatic fever
Scarlet fever	Acute glomerulonephritis
Skin and soft tissue infections: <ul style="list-style-type: none">• Impetigo (pyoderma)• Cellulitis and erysipelas	Guttate psoriasis Reactive arthritis
Deep soft tissue infections: <ul style="list-style-type: none">• Necrotizing fasciitis• Streptococcal myositis• Toxic shock syndrome	PANDAS (Pediatric Autoimmune Neuropsychiatric Disorders Associated with Streptococcal infections)
Bacteremia leading to toxic shock syndrome, osteomyelitis, meningitis, etc.	

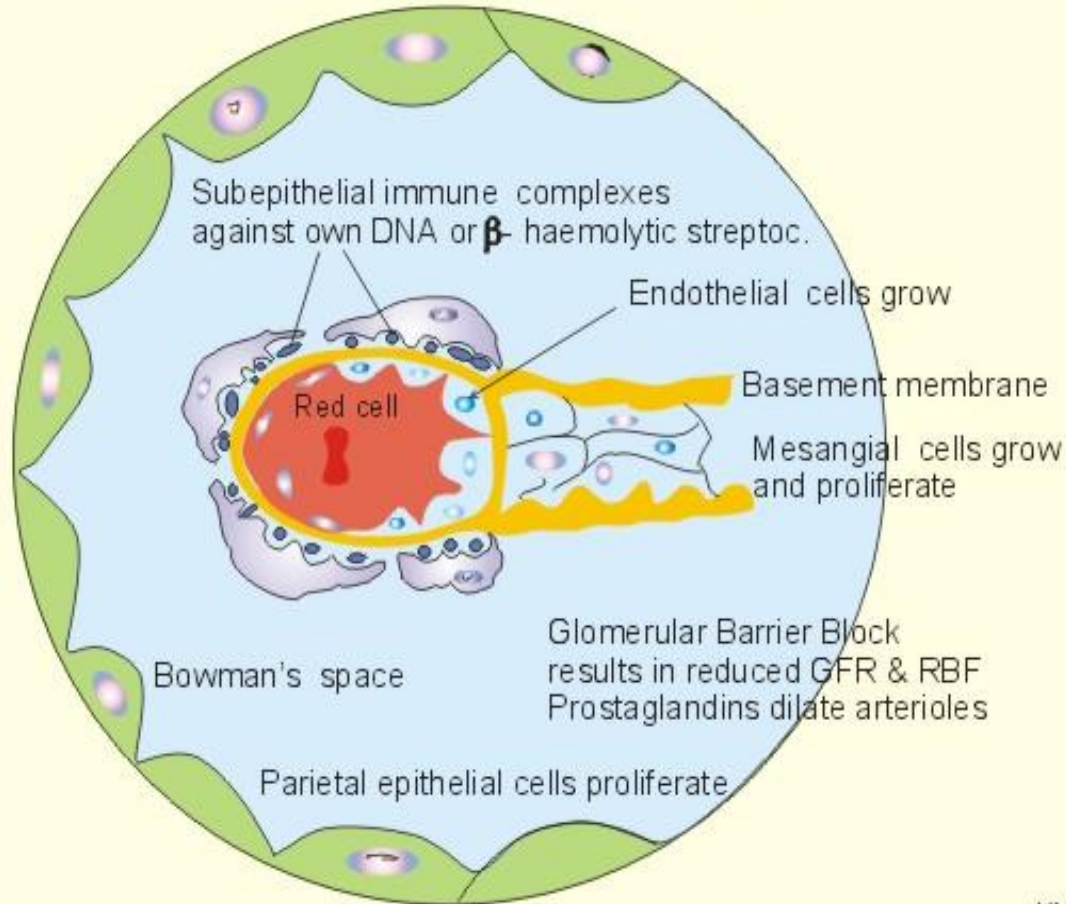


ETIOLOGICAL FACTORS IN RHEUMATIC HEART DISEASE



Antigenic cross reaction between streptococci and cardiac tissues

Post- Streptococcal Glomerulo-nephritis



Immune
Complex
Mediated
damage

Fig. 25-19

Laboratory diagnosis

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

- Direct microscopy \longrightarrow Culture on blood agar

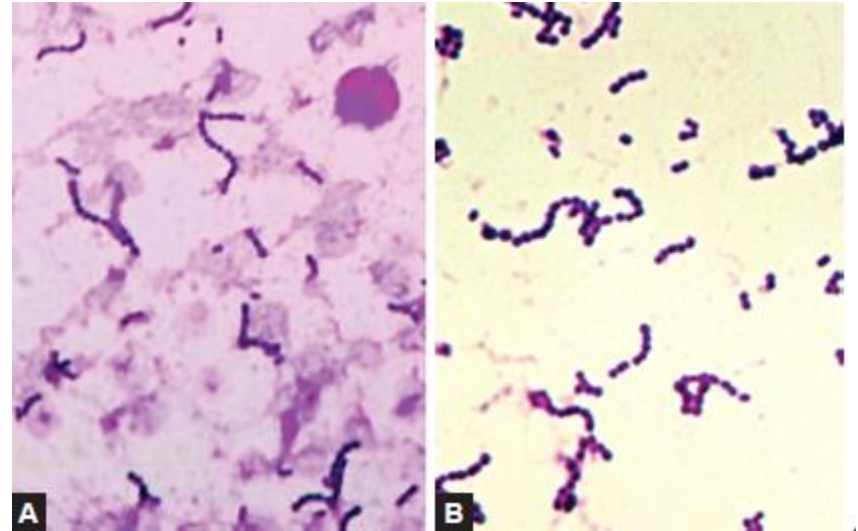
Antibiotic sensitivity

Biochemical
test

Use selective
media

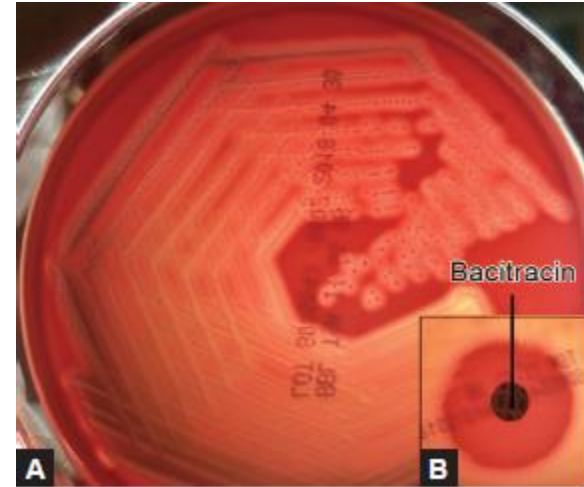
- Serology (ASO titer, Anti DNAase B)

- **Specimens:**
 - Throat swab, pus swab, exudates and blood
 - Pike's transport media
- **Microscopy**
 - Gram positive cocci in chains



- Aerobe & Fastidious anaerobes
- Blood Agar – Beta hemolytic pinpoint colonies
- Liquid medium – Granular turbidity
- Selective Medium
 - Crystal violet blood agar
 - PNF medium

- Catalase – Negative
- Bacitracin - sensitive



- **ASO (Anti-streptolysin O) antibodies**
- Titer >200 Todd unit/ml in most of the streptococcal infections except pyoderma and PSGN.
- detected by latex agglutination test.

- **Anti-DNase-B antibodies –**
- Titre >300-350 units/ml is diagnostic of PSGN and pyoderma
- **Anti-hyaluronidase antibodies**
- **Anti-streptokinase antibodies**

OTHER BETA HEMOLYTIC STREPTOCOCCI

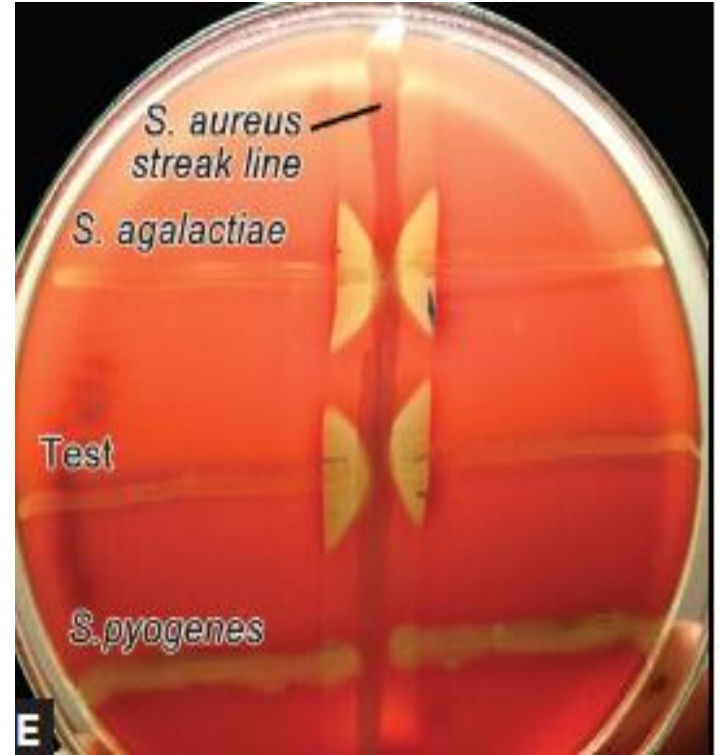
S.agalactiae

- Group B Streptococcus
- Carriage in vagina and rectum in 30% women
- Neonatal sepsis & meningitis
- Pregnancy → peripartum fever
- Cellulitis and soft tissue infections urinary tract infection, pneumonia, and endocarditis – in diabetics, cancer patients

Early v/s Late GBS Disease

Characteristics	Early-onset disease	Late-onset disease
Age of onset	0–6 days of birth	7–90 days of birth
Increased risk following obstetric complications	Prematurity and prolonged labor	Not associated
Mode of transmission to the baby	During or before birth from the colonized maternal genital tract	Contact with a colonized mother and nursing personnel
Common clinical manifestations	Pneumonia and/or respiratory distress syndrome followed by meningitis	Bacteremia&meningitis (most common)
Common serotypes	Ia, III, V, II, Ib	III predominates
Case fatality rate	4.7%	2.8%

- Lab Diagnosis
- CAMP positive: CAMP factor (named after the discoverers: Christie, Atkins-Munch-Petersen)



- **Other Tests:**

- Hippurate hydrolysis test positive
- Bacitracin resistant
- PYR test negative
- Orange pigment - Islam's medium
- Mucoid β haemolytic colonies

- **Treatment**

- Penicillin or higher penicillins

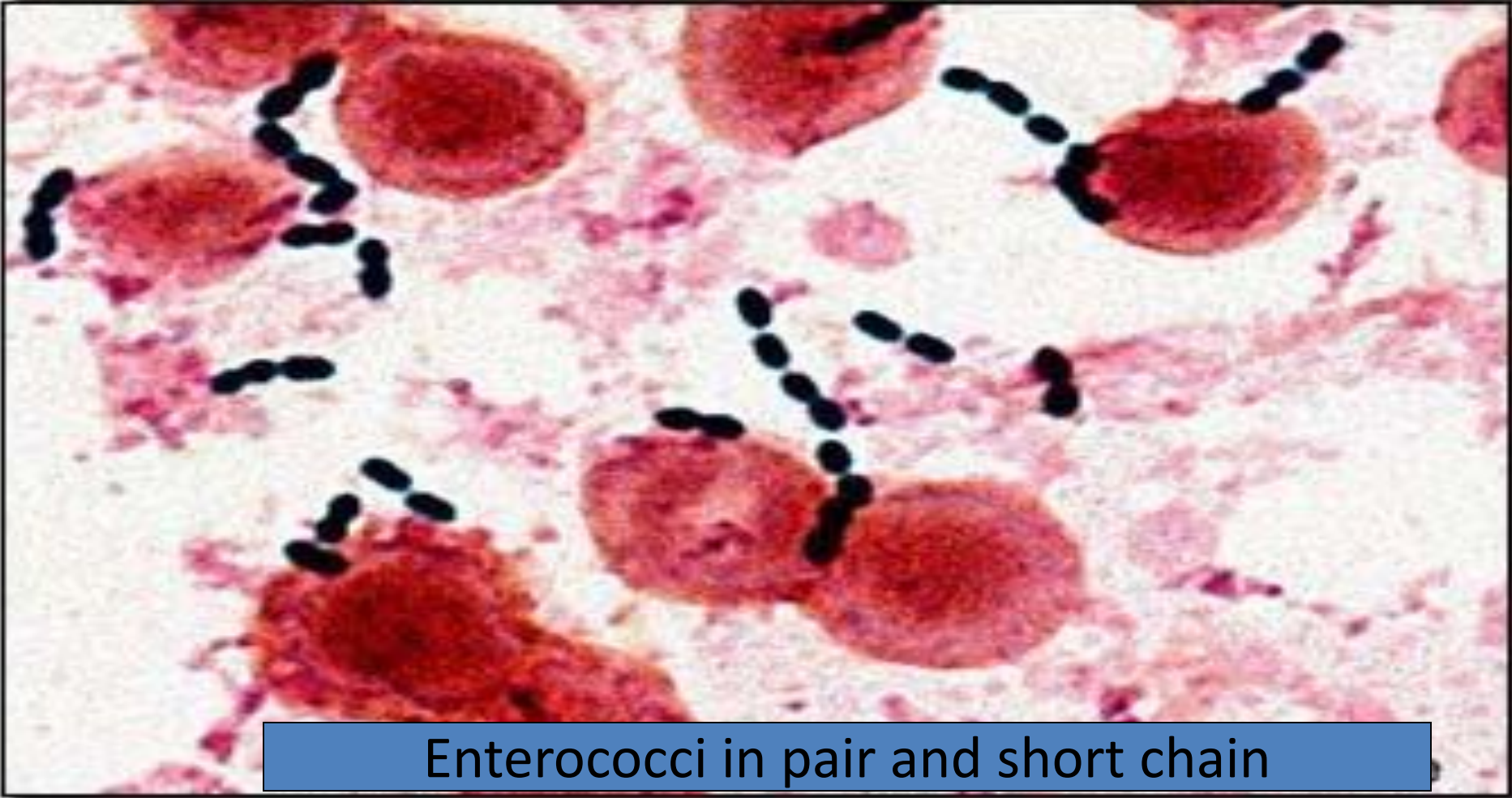
- **Prevention**

- Screening at 35-37 wks of pregnancy
- Ampicillin or penicillin to carrier mothers

Group D Streptococci

- Two groups :
 - 1) Enterococcus group E. faecalis, E. faecium
 - 2) Nonenterococcal group S. bovis, S. equinus

Gram stain of *Enterococcus faecalis*



Enterococci in pair and short chain

Characteristics of Enterococci

- Ability to grow in the presence of 40% bile
- 6.5% NaCl
- At pH 9.6 and at 45° C
- On mac Conkey`s medium, they produce tiny deep pink colonies.
- They appear as pairs of oval cocci, the pair arranged at an angle to each other
- Non hemolytic

The viridans group

- Normal resident in the mouth and the upper respiratory tract and typically produce greenish (alpha hemolysis) discoloration on blood agar.
- *Str. mitis*, *Str. mutans*, *Str. sanguis*
- Causative agent for subacute bacterial endocarditis, most often *Str. sanguis*
- Dental caries by *Str. mutans*

PROTOZOA: Amoebae

protozoa

```
graph TD; A[protozoa] --- B[Rhizopoda]; A --- C[Mastigophora]; A --- D[Sporozoa]; A --- E[Ciliates]; B --- B1[-pseudopodia]; B --- B2[-entamoeba]; C --- C1[-flagella]; C --- C2[leishmania, Trypanosoma, Trichomonas, Giardia]; D --- D1[-amoeboid movement]; D --- D2[-plasmodia]; E --- E1[-cilia]; E --- E2[-balantidium];
```

Rhizopoda
-pseudopodia
-entamoeba

Mastigophora
-flagella
leishmania,
Trypanosoma,
Trichomonas,
Giardia

Sporozoa
-amoeboid
movement
-plasmodia

Ciliates
-cilia
-
balantidium

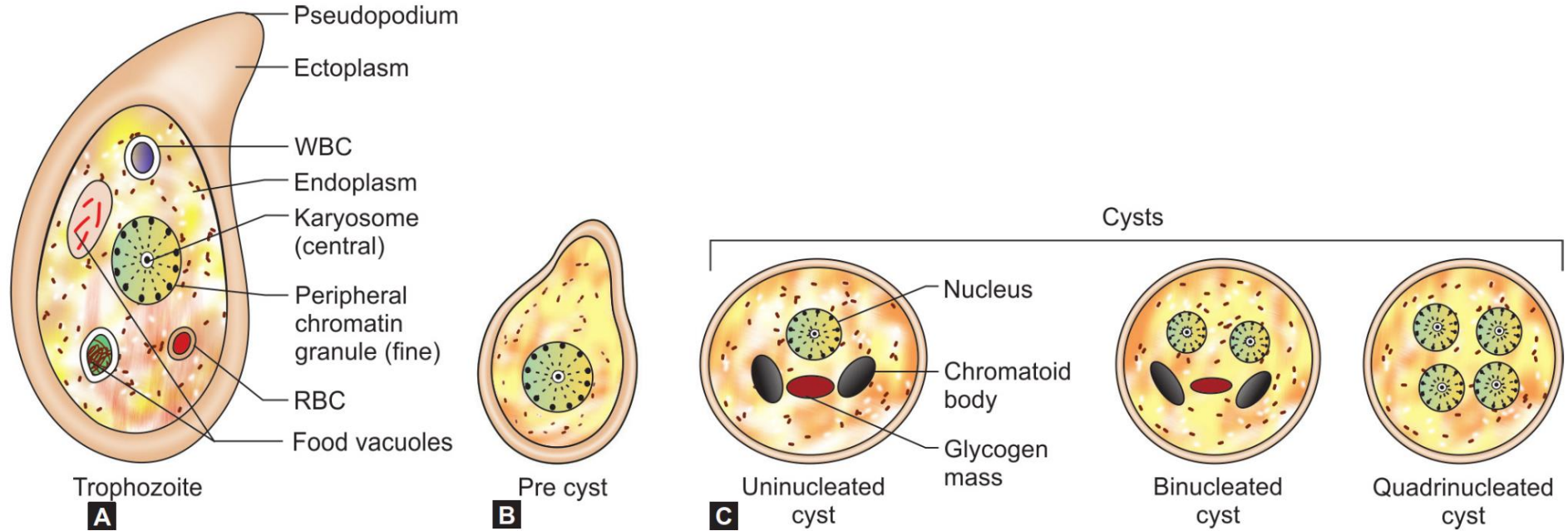
Table 3.1: Classification of amoebae based on habitat

<i>Intestinal amoebae (large intestine)</i>		<i>Free living amoebae (in soil and water)</i>
Pathogenic	Nonpathogenic	Opportunistic pathogen
<ul style="list-style-type: none">• <i>Entamoeba histolytica</i>• <i>E. moshkovskii</i> (infants)• <i>E. bangladeshi</i> (infants) (unknown virulence)	<ul style="list-style-type: none">• <i>E. dispar</i>• <i>E. coli</i>• <i>E. polecki</i>• <i>E. hartmanni</i>• <i>Endolimax nana</i>• <i>Iodamoeba butschlii</i>• <i>E. gingivalis</i>* (mouth)	<ul style="list-style-type: none">• <i>Acanthamoeba</i> species• <i>Naegleria fowleri</i>• <i>Balamuthia mandrillaris</i>• <i>Sappinia</i> species

The habitat of *E. gingivalis** is oral cavity, not large intestine.

Note: E. moshkovskii and *E. bangladeshi* are occasionally reported to be

Morphology



Figs 3.1A to C: *Entamoeba histolytica* (schematic diagram): (A) Trophozoite; (B) Precyst; (C) Cysts

Life cycle

- **Host:** *E. histolytica* completes its life cycle in single host, i.e. man.
- **Infective form:**
 - Mature quadrinucleated cyst is the infective form.
 - However, uni or binucleated cysts can also be infective.
- **Mode of transmission**
 - Feco-oral route (most common)
 - Sexual (oral, anal)
 - Vector

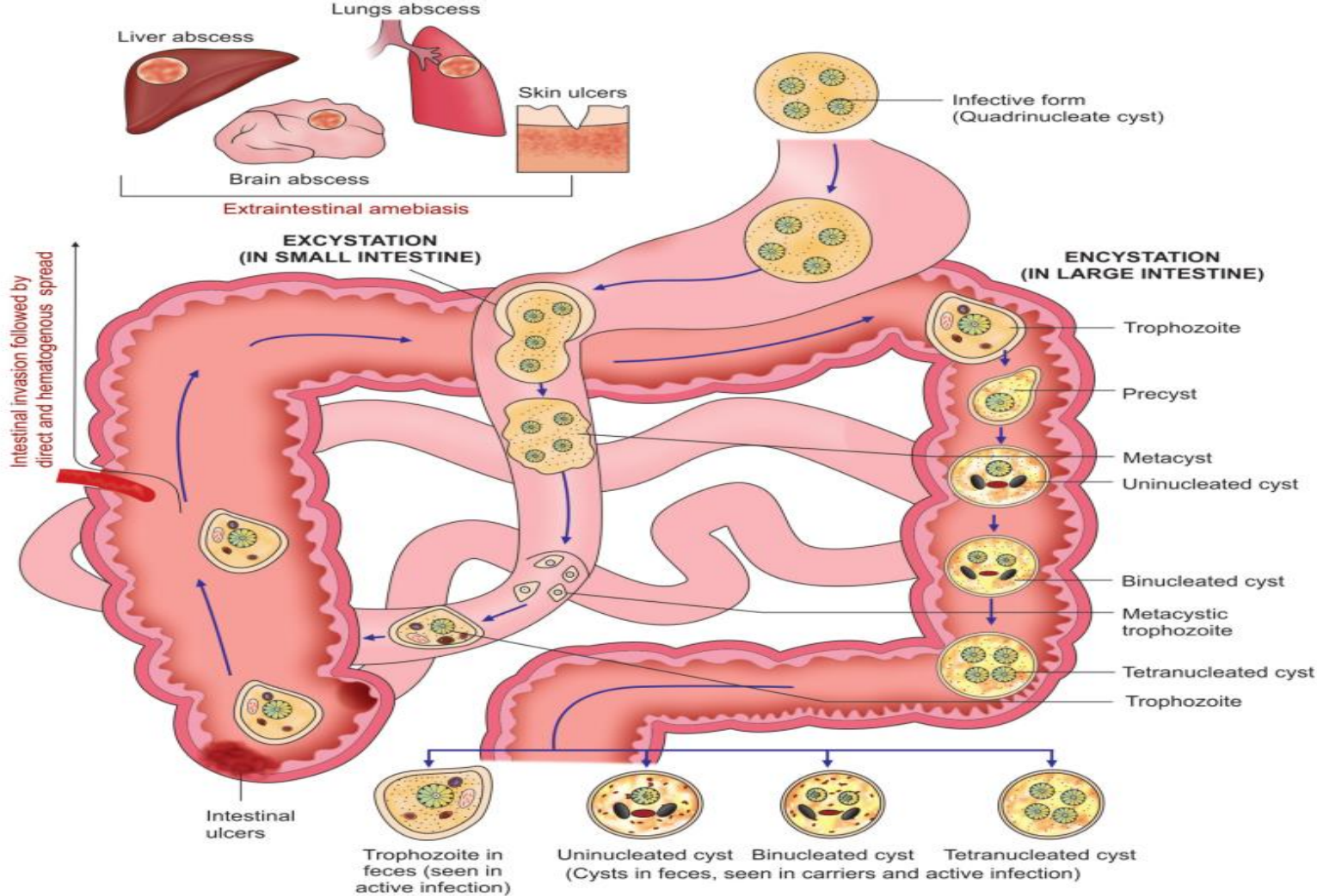


Fig. 3.2: Life cycle of *Entamoeba histolytica*

Clinical Manifestations of Amoebiasis

- **Asymptomatic amoebiasis-90%**
- **Intestinal amoebiasis**
 - Amoebic dysentery
 - Amoebic appendicitis
 - Amoeboma
 - Fulminant colitis
- **Amoebic liver abscess**
 - hepatomegaly and fever (most consistent features)
 - weight loss, sweating and weakness,
 - very rarely jaundice, and cough

- ❑ **Stool microscopy** by wet mount, permanent stains, etc.—detects cysts (round, 1–4 nuclei) and trophozoites (with finger like pseudopodia)
- ❑ **Histology**—intestinal biopsies stained with PAS or H&E stains reveal trophozoites
- ❑ **Stool culture**—Polyxenic and axenic culture
- ❑ **Stool antigen detection** (copro-antigen)—ELISA (detecting 170-kDa of lectin Ag) and ICT (detecting 29-kDa surface Ag)
- ❑ **Serology**
 - Amoebic antigen—ELISA (170-kDa of lectin Ag)
 - Amoebic antibody—IHA, ELISA and IFA
- ❑ **Isoenzyme** (zymodeme) analysis
- ❑ **Molecular diagnosis**—Nested multiplex PCR and real time PCR (18S rRNA) and Biofire FilmArray

Laboratory Diagnosis of Intestinal Amoebiasis

- **Sample collection**
- **Stool macroscopy**
- **Stool microscopy**
 - Trophozoites
 - Quadrinucleated cyst

Table 3.4: Differences in stool characters between amoebic dysentery and bacillary dysentery

<i>Character</i>	<i>Amoebic dysentery</i>	<i>Bacillary dysentery</i>
<i>Pathology</i>		
Ulcer	Deep	Shallow
Margin	Ragged and undermined	Uniform
Intervening mucosa	Normal	Inflamed
Necrosis type	Pyknosis (pyknotic bodies)	Karyolysis (ghost cells)
Cellular response	Mononuclear	Polymorphonuclear
<i>Stool macroscopic feature</i>		
Number of motion	6–10/day	> 10/day
Amount	Copious amount	Small quantity
Color	Dark red	Bright red
Odor	Offensive	Odorless
Reaction	Acidic	Alkaline
Consistency	Not adherent to the container	Adherent to the container

Table 3.4: Differences in stool characters between amoebic dysentery and bacillary dysentery

<i>Character</i>	<i>Amoebic dysentery</i>	<i>Bacillary dysentery</i>
<i>Stool microscopic feature</i>		
Red blood cells (RBCs)	In clumps	Discrete or in rouleaux
Pus cells	Few	Numerous
Macrophages	Few	Numerous*
Eosinophils	Present	Absent or rare
Charcot Leyden crystal	Present	Absent
Pyknotic body**	Present	Absent
Ghost cell	Absent	Present
Organism detected	<i>E. histolytica</i> cyst or trophozoite	Bacteria (e.g. <i>Shigella</i>)

Stool culture..

- National Institute of Health (NIH) media
- Boeck and Drbohlav egg serum medium
 - containing Lockes solution
 -

- **Stool antigen detection (coproantigen)**
 - ELISA detecting 170 kDa of lectin antigen
 - Immunochromatographic test

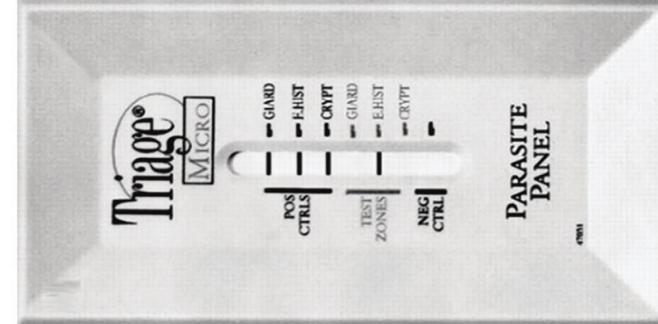


Fig. 3.7: Triage parasite panel, positive for *E. histolytica*/*E. dispar* antigen

- **Imaging method**

- Colonoscopy can be performed to detect collar button or flask shaped amoebic ulcers.

- **Other nonspecific findings**

- Charcot Leyden crystals in stool
 - Moderate leucokytosis in blood.

Free-Living Amoebae

- Small, freely living, widely distributed in soil and water and can cause opportunistic infections in humans.
- *Naegleria fowleri* is a causative agent of primary amoebic meningoencephalitis (PAM).
- *Acanthamoeba* species causes granulo matous amoebic encephalitis (GAE) and amoebic keratitis in contact lens wearers.
- *Balamuthia mandrillaris* causes GAE.
- *Sappinia* species: causes encephalitis.