

Etio-pathogenesis and lab diagnosis of meningitis

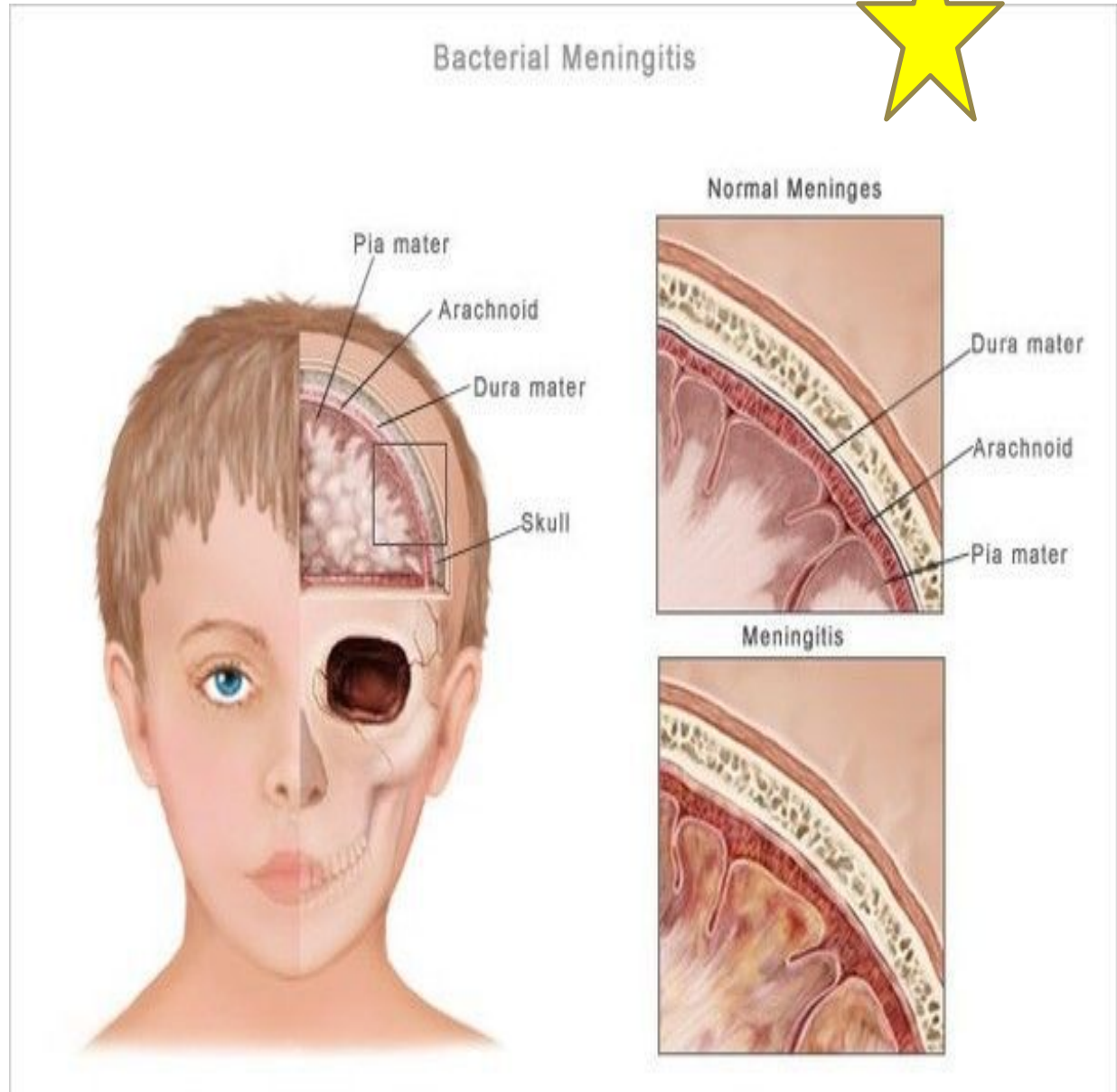
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MENINGITIS



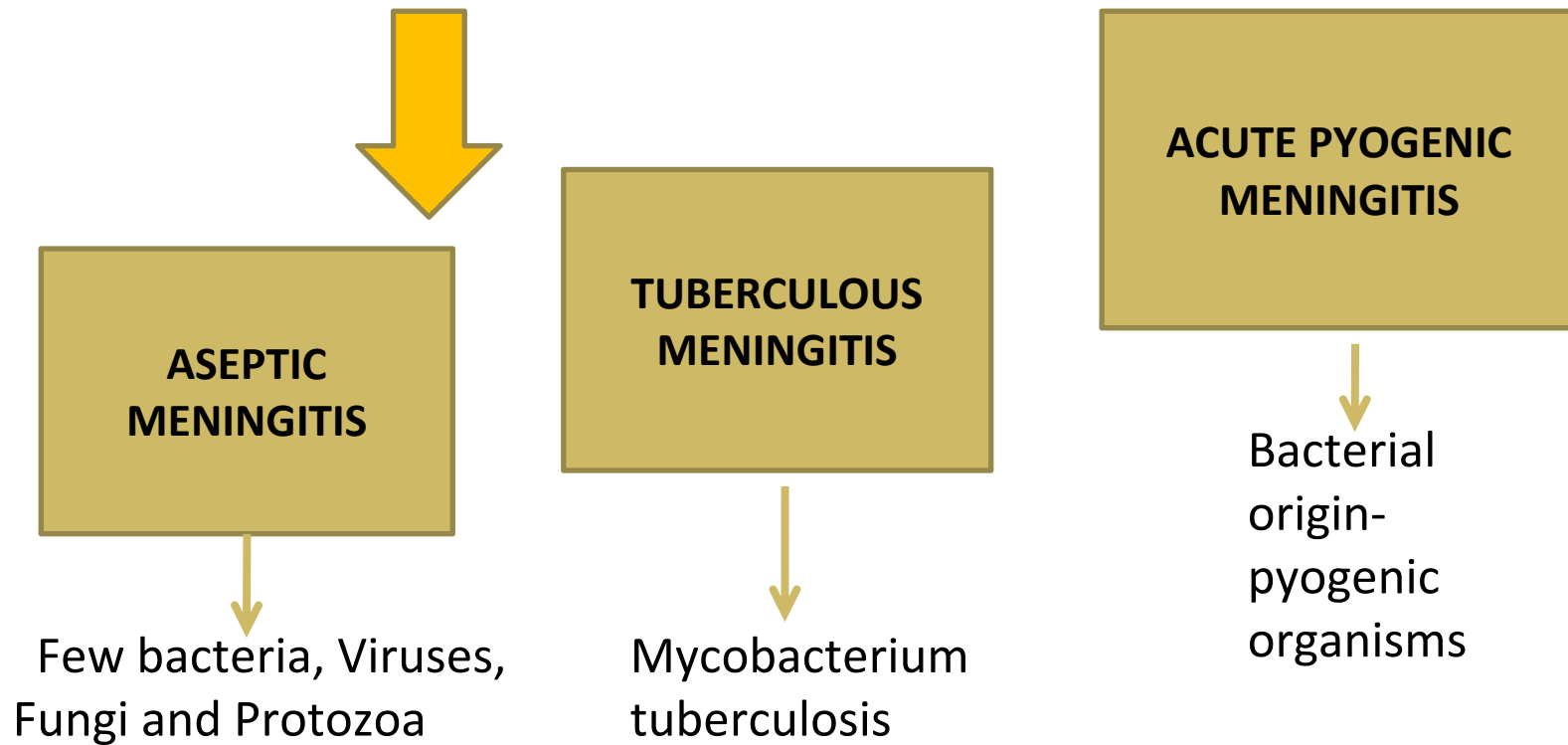
- Meningitis is an inflammation of the meninges surrounding the brain and spinal cord
- (leptomeninges).



DEFINITIONS



□ TYPES OF MENINGITIS



Bacteria causing Purulent Meningitis related to age



AGE	MOST COMMON CAUSES
Neonates or infants of 0-2 months	Escherichia coli, Streptococcus agalactiae(Group B) Klebsiella pneumoniae Listeria monocytogenes
Children	Neisseria meningitidis(Most Common) Haemophilus influenzae Streptococcus pneumoniae
Adolescents	N. Meningitidis
>20 years(Adults)	Streptococcus pneumoniae Haemophilus influenzae Neisseria meningitidis
Old age	Listeria monocytogenes

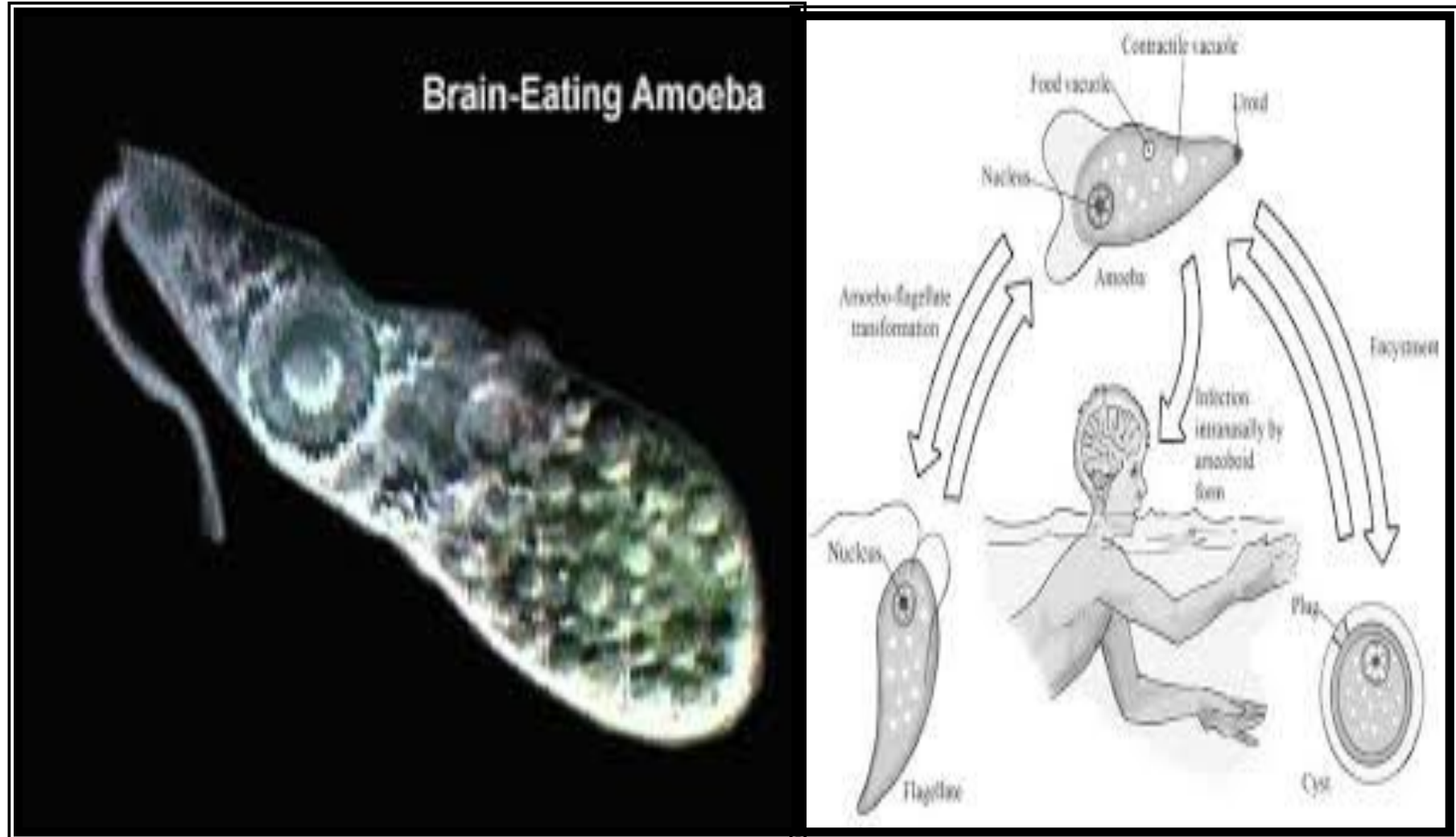
ASEPTIC MENINGITIS

Causative agents



VIRUSES	BACTERIA	FUNGI	PROTOZOA
Enteroviruses (echo, polio, coxsackie)	Mycobacterium tuberculosis	Cryptococcus neoformans	Naegleria fowleri
Myxoviruses (Mumps and Influenza)	Leptospira interrogans		Acanthamoeba species
Adenovirus and Arbovirus	Treponema pallidum		Toxoplasma gondii
HSV, VZV, EBV, CMV			
Measles, Rubella and HIV, Rabies			

Naegleria -PAM



Other causes

- Stapylococccal meningitis
- Brucella
- H.capsulatum
- Coccidoides immitis..etc

PATHOGENESIS



PATHOGENESIS



- 3 major pathways

Meningococcal
Cryptococcal,
Syphilitic,
Pneumococcal

Naegleria
fowleri
Rabies, HSV

Pathogenesis



Bacterial invasion



Parameningeal focus/colonisation by adhesion, binding receptors, pili.



Microbial virulence factors-inflammatory response-
endotoxin/lipopolysaccharide, peptidoglycan, lipoteichoic
acid, IgA proteases



Release of inflammatory mediators by astrocytes,
microglial cells and mast cells

PATHOGENESIS



BACTERIAL MENINGITIS

- Increased concentration of
- $TNF-\alpha$,
- IL-1, IL-8
- IL-6,

- NO, PAF, PGE2

ASEPTIC MENINGITIS

- Increased levels of
- IL-6,
- $TNF-\alpha$,
- IFN- γ

LABORATORY DIAGNOSIS



SPECIMEN COLLECTION AND TRANSPORT



SPECIMEN:

- CSF- lumbar puncture under aseptic conditions
- Blood, serum.

- CSF collection: In THREE sterile containers



- Volume of CSF collected is critical for detection of certain microorganisms (mycobacteria, fungi)
- Minimum of 5- 10 ml is recommended for collection.
- Processing of too little specimen lowers sensitivity.



- CSF to be delivered immediately to laboratory.
- Incubator at 37°C or at RT
- **CSF should never be refrigerated.**
- Exception-CSF for viral studies may be refrigerated for as long as 23 hrs or frozen at -70 °C

Microbiological Investigations



- Microscopy
- Culture
- Antigen detection
- Antibody detection
- Endotoxin detection
- Molecular methods

MICROSCOPIC EXAMINATION



- **Stained smear of sediment:**

Gram staining-done on all CSF sediments

Heaped smear

Acridine orange flurochrome stain for faster examination.

- **Wet mount:**

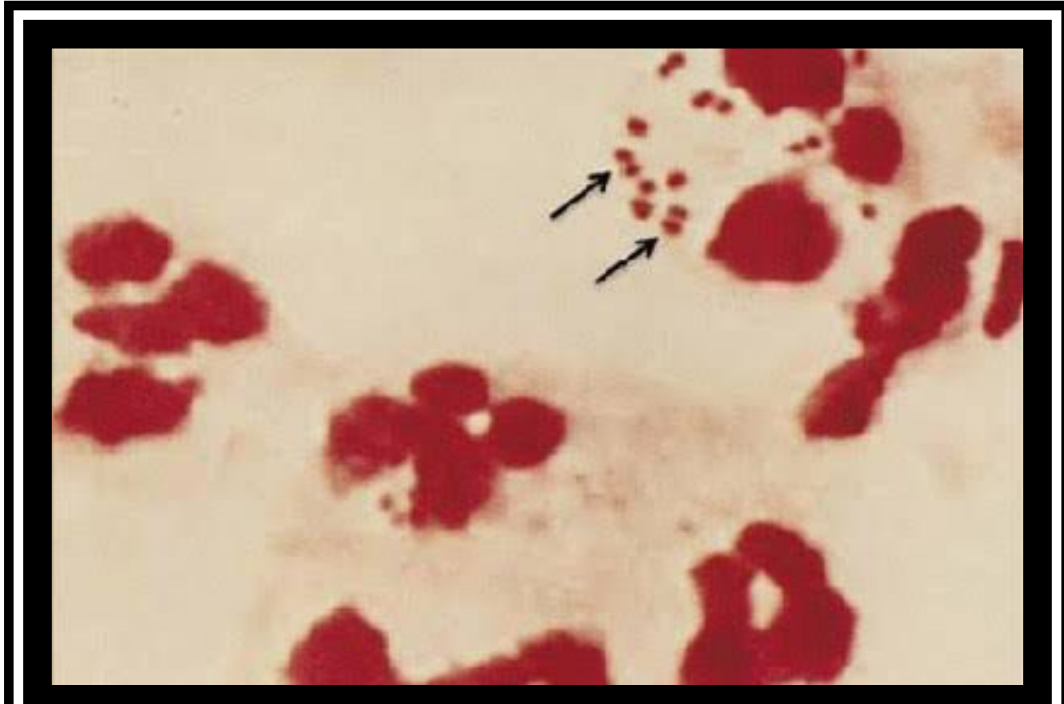
To detect motile amoeba

Phase contrast microscopy

- **Zeihl Neelsen Stain /Acid fast stain:**

When tuberculous meningitis is suspected

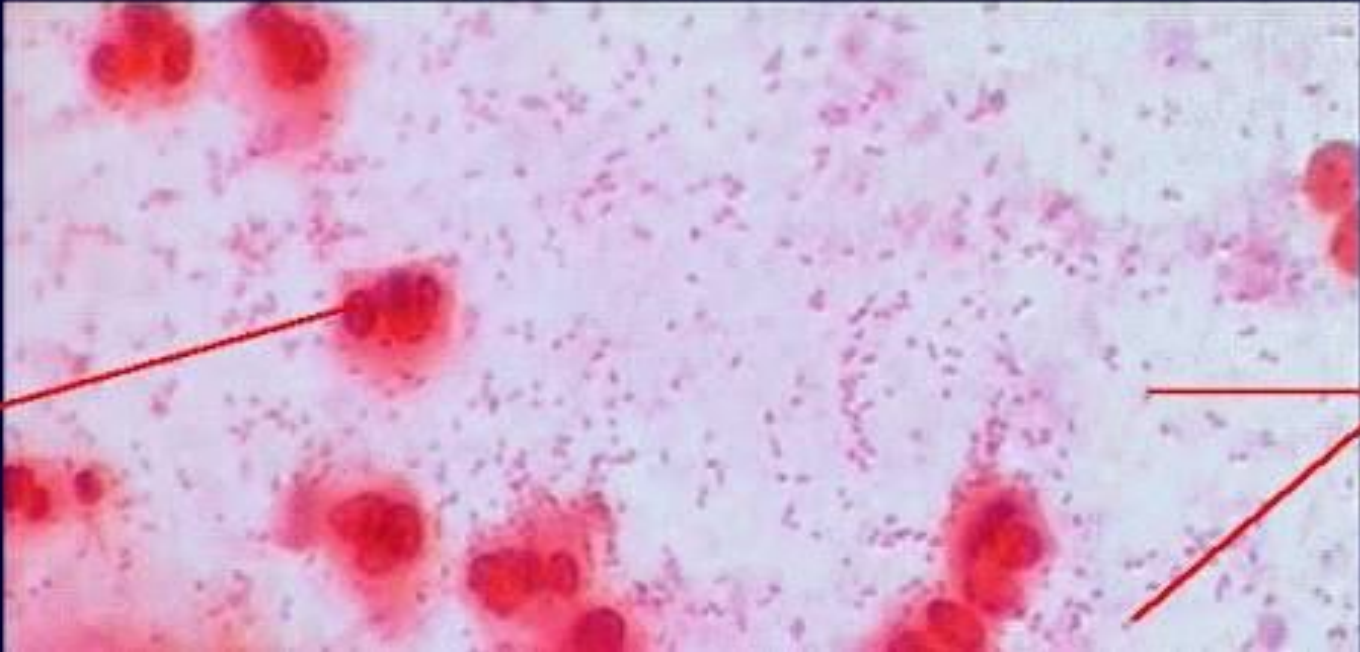
AFB is difficult to detect in CSF



Haemophilus influenzae - Respiratory

Typical Morphology:
small, pale,
pleomorphic
Gram-negative
rods.

white blood cell
(PMN)

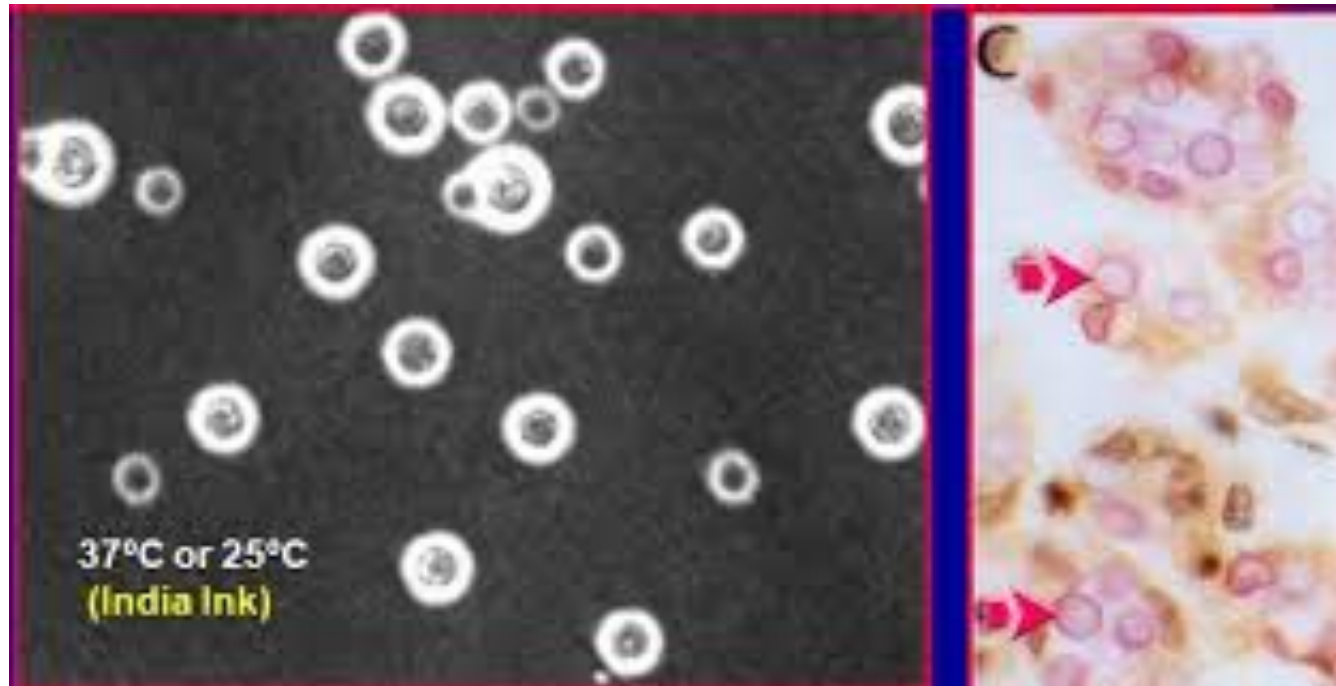




□ **India Ink preparation-**

To detect Cryptococcal meningitis

Presence of capsulated budding yeast cells



CULTURE



- Blood agar, Chocolate agar-incubate in 5% to 10% CO₂ at 35-37°C.
- Antibiotic sensitivity tests to be done for all positive cultures.
- **Primary drug susceptibility testing(DST)**
- Blood culture
- Fungal culture-SDA or BHI agar at 35°C for 4 wks



- Suspected TB meningitis-LJ medium, liquid media, automated cultures

- Viral culture-cell lines

- Naegleria or Acanthamoeba species

Bacterial overlay on non nutrient agar is used for detection.

ANTIGEN DETECTION



Bacterial Antigen Testing (BAT)



For rapid detection of antigen from CSF

Useful in previously treated patients and **gram stain negative CSF specimen**

- CSF-Supernatant is used.

- **Latex agglutination**

Streptococcus pneumoniae

Streptococcus agalactiae

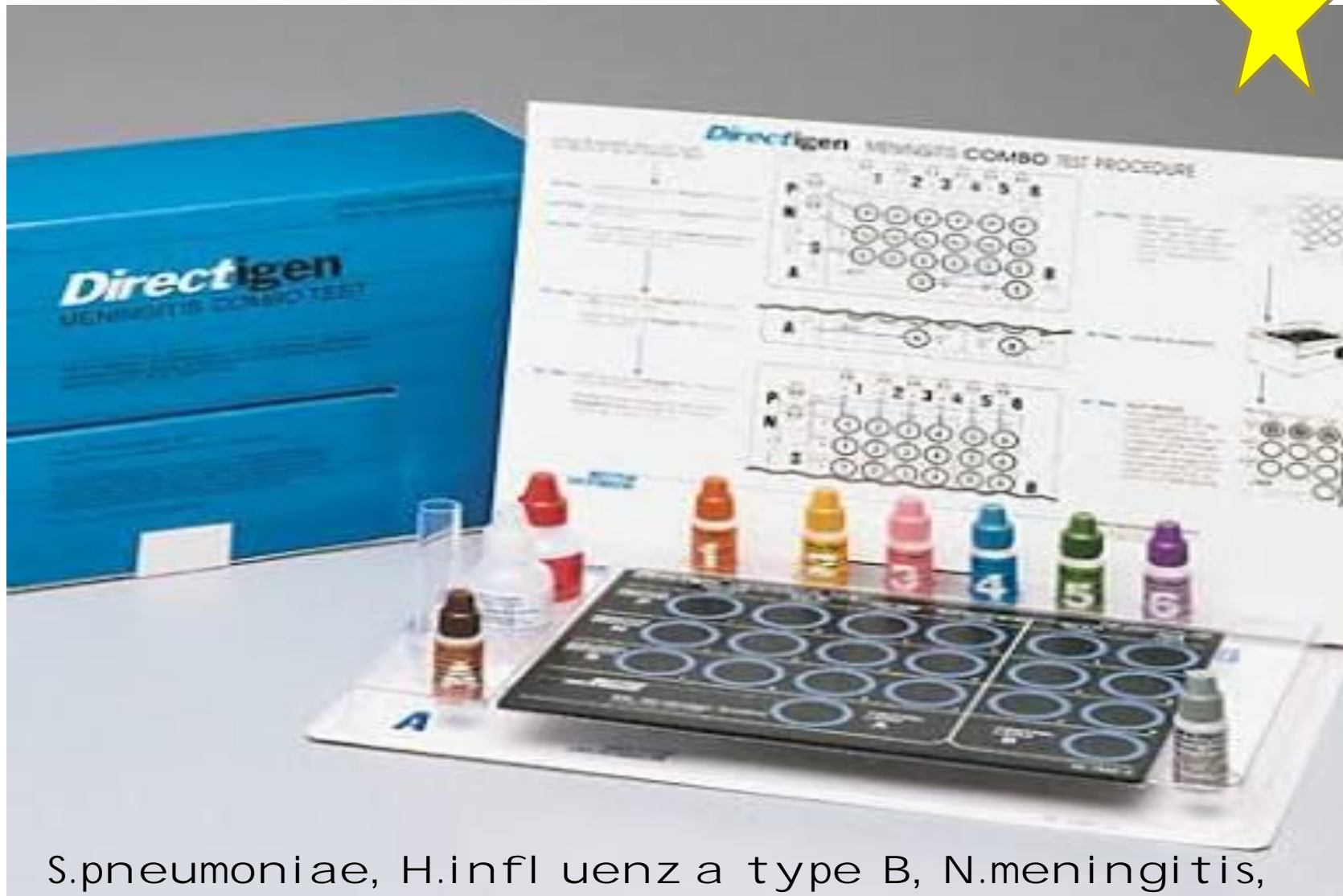
Neisseria meningitidis

Haemophilus influenzae

- **CIEP**

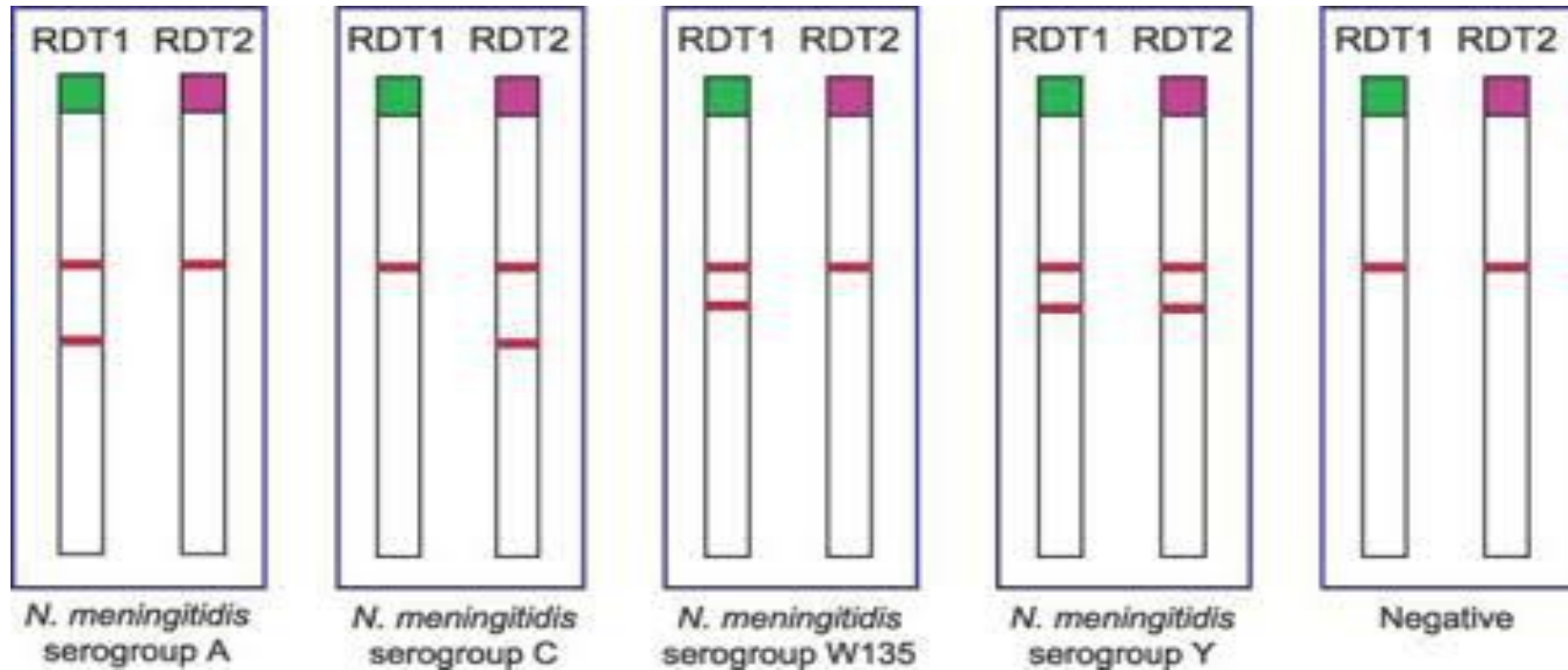
Latex agglutination test for
cryptococcal antigen

CALAS-Cryptococcal Latex
Agglutination System



S.pneumoniae, H.infl uenz a type B, N.meningitis, S.agal actiae, E.col i K1

RAPID DIAGNOSTIC TEST



ANTIBODY DETECTION



- Ab against viral infections –Herpes group
- ELISA
- CIEP

BACTERIAL ENDOTOXIN DETECTION



- Limulus Lysate Assay
- Patients blood + amoebocytes → coagulation
- To detect endotoxins from gram negative bacteria

MOLECULAR METHODS



- Polymerase chain reaction (PCR)
- Done to detect Mycobacterium tuberculosis specific genes such as IS 6110 gene- Tubercular meningitis
- **Herpes simplex virus- Most commonly used**
- Enteroviruses(RT-PCR)
- Pyogenic meningitis- Multiplex PCR can be done targeting genes of common etiological agent.



**We all
Smile
In the
Same
Language**

