



Microbiological Examination of CSF

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(Lumbar puncture)



Tube 1 (chemical and serologic test)

Tube can be frozen

Tube 2 (microbiological analysis)

Room temperature

Tube 3 (cell count and differential)

Tube can be refrigerated

Tube 4 (PCR, TB, additional tests)

Sample management

should be centrifuged directly for culture & gram stain (1500 xg, 15 min)
Supernatant ; for antigen detection
Avoid refrigeration (Neisseria meningitidis)
Do not expose it to heat or sunlight (xanthochromia)

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Typical Lumbar CSF Findings in Meningitis

Test	Bacterial	Viral	Fungal	Tuberculous
Opening pressure	Elevated	Usually normal	Variable	Variable
Leukocyte count	≥1000/µL	<100/µL	Variable	Variable
Cell differential	Mainly neutrophils*	Mainly lymphocytes [†]	Mainly lymphocytes	Mainly lymphocytes
Protein	Mild to marked increase	Normal to mild increase	Increased	Increased
Glucose	Usually ≤40 mg/dL	Normal	Decreased	Decreased: may be <45 mg/dL
CSF/serum glucose ratio	Normal to marked decrease	Usually normal	Low	Low
Lactic acid	Mild to marked increase	Normal to mild increase	Mild to moderate increase	Mild to moderate increase

The most common agents of bacterial meningitis

- group B streptococcus (neonates),
- Neisseria meningitidis (3 months and older),
- Streptococcus pneumoniae (3 months and older),
- Escherichia coli and other gram-negative bacilli (newborn to 1 month),
- Haemophilus influenzae (3 months to 18 years),
- Listeria monocytogenes (neonates, older adults, alcoholics, and immunosuppressed)



Gram stain & culture

- > An accurate, rapid method
- sensitivity ranges 60% to 90%

greatest sensitivity: concentrations of bacteria $\geq 10^5$ CFU/mI)

Listeria monocytogenes and gramnegative bacilli ≤ 50%

 Cultures have a sensitivity of 80% to 90%, but are about 30% less sensitive in partially treated cases

Typical Bacterial Meningitis

- CSF WBC > 1000, PMN predominance
- CSF protein > 500mg/dl
- CSF glucose < 45 mg/dl



Viral Meningitis

- Enteroviruses (echoviruses, coxsackieviruses, polioviruses) and arboviruses
- viral cultures

The sensitivity can be low 72% for enteroviruses to 5% for HSV

• RT-PCR

"gold standard" for the diagnosis of enterovirus, HSV, cytomegalovirus, varicella zoster.

• PCR



Fungal Meningitis

Cryptococcus neoformans



India ink or nigrosin stains for cryptococcus capsular

Sensitivity is about 25%, increasing with **multiple lumbar punctures** and **large volumes of CSF** (15-20 mL)



Latex agglutination (sensitivity, 60% -95%)

False positives: rheumatoid factor or Trichosporon asahii infection



False negatives: prozone effect or low concentration of polysaccharide antigen, Early disease, infection with nonencapsulated variants.



Tuberculous Meningitis

- Abnormal CSF with elevated protein and lymphocytic predominance
- The sensitivity of CSF acid-fast stains (10% -12% to 50%)
- Large volumes of CSF are recommended to improve the sensitivity of both acid-fast stain and culture
- Multiplex PCR analysis
- Adenosine deaminase (ADA)

increase in T lymphocytes in tuberculosis patients

Primary Amebic Meningoencephalitis (PAM)

free-living ameba Naegleria fowleri





	Bacterial	Viral	Fungal	ТВ
Opening pressure (N: 4–18 cm H ₂ 0)	Elevated	Slightly elevated	Normal or high	Usually high
WBC (N: ≤5/mm³)	> 200/mm ³	< 200/mm ³	< 50/mm ³	20–30/mm ³
Differential (N: ≤ 3 PMNs /mm ³)	PMNs	Monocytes	Monocytes	Monocytes
Glucose (N: 45–80 mg/dL)	Low (< 60% serum)	Normal	Low	Low
Protein (N: 23–38 mg/dL)	Very high	Normal	High	High
RBC (N:≤ 5 /mm³)	Few	None	None	None