

SOP

Collection, Storage and Transportation of Cerebrospinal Fluid (CSF) Samples in Paediatric Patients

**Department of Paediatrics
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Standard operating procedure (SOP) for the Collection, Storage and Transportation of Cerebrospinal Fluid (CSF) Samples in Paediatric Patients

(1) Title

Standard Operating Procedure for Collection, Storage and Transportation of Cerebrospinal Fluid (CSF) Samples in Paediatric Patients

(2) Purpose

To provide a standardised procedure for the safe and effective collection, handling, storage, and transportation of cerebrospinal fluid (CSF) samples obtained from paediatric patients to ensure specimen integrity, accurate laboratory diagnosis, and patient safety.

(3) Scope

This SOP applies to healthcare professionals involved in the collection, handling, documentation, storage, and transportation of CSF specimens obtained from paediatric patients undergoing lumbar puncture for diagnostic purposes.

(4) Responsibilities

(4.1) Medical Officer / House officer

- Assess the clinical indication and safety of the lumbar puncture.
- Request and document informed consent from parents/legal guardians.
- Perform or supervise the CSF collection using a strict aseptic technique.
- Ensure correct labelling and comprehensive documentation.

(4.2) Nursing Staff

- Prepare the paediatric patient and all required clinical equipment.
- Assist the proceduralist during the clinical procedure.
- Coordinate and ensure proper transport arrangements to the laboratory.

(4.3) Laboratory Personnel

- Receive, verify, process, and store specimens according to standard laboratory protocols.
- Document and report specimen inadequacies, contamination, or transportation delays.

(5) Ethical Considerations

- Request informed consent from parents or legal guardians before the procedure.
- Maintain patient confidentiality during documentation, transport, and storage.
- Perform the procedure only when clinically indicated.
- Minimise discomfort and distress in paediatric patients through appropriate preparation and analgesia/supportive measures.

(6) Prerequisites

(6.1) Patient-related preparation

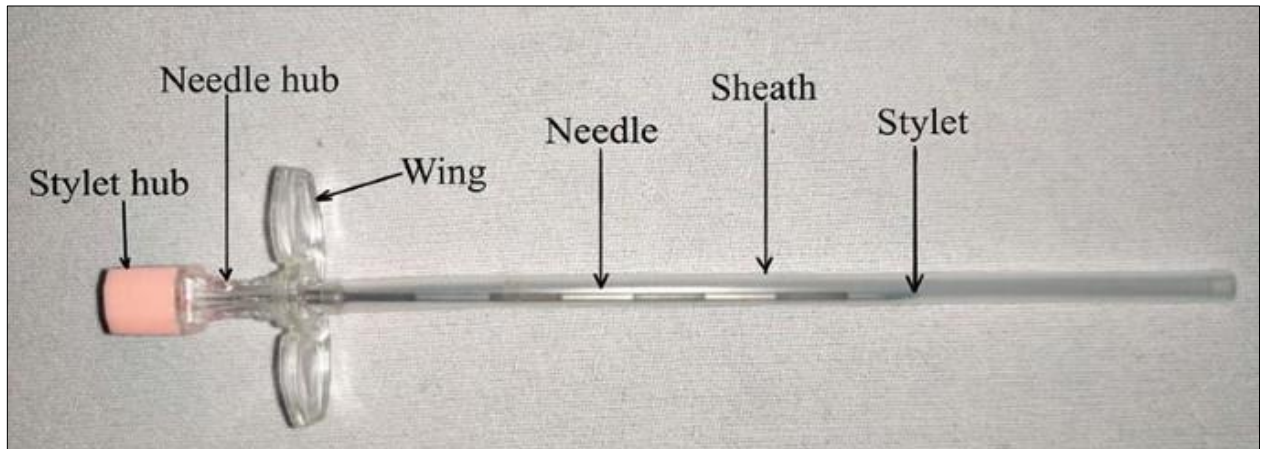
- Confirm patient identity using appropriate identifiers.
- Review clinical indication and contraindications for lumbar puncture.
- Explain the procedure to parents/guardians.
- Ensure appropriate positioning and monitoring.

(6.2) Required equipment

- **Sterile Lumbar Puncture Set:** Sterile syringes, kidney dish, towel clips, forceps, sterile bowls, sterile drapes, and gauze.



- **Spinal needle**
 - Use a 22 or 25G bevelled spinal needle with stylet
 - Consider using 25G pencil point needles for older children/adolescents to reduce the risk of headache, evidence is not convincing in younger children



	14 GAUGE COLOR: OLIVE OUTER DIAMETER: .072IN (1.83MM)
	15 GAUGE COLOR: AMBER OUTER DIAMETER: .065IN (1.65MM)
	16 GAUGE COLOR: GRAY OUTER DIAMETER: .064IN (1.63MM)
	18 GAUGE COLOR: GREEN OUTER DIAMETER: .050IN (1.27MM)
	20 GAUGE COLOR: PINK OUTER DIAMETER: .036IN (.91MM)
	21 GAUGE COLOR: PURPLE OUTER DIAMETER: .033IN (.83MM)
	22 GAUGE COLOR: BLUE OUTER DIAMETER: .027IN (.70MM)
	23 GAUGE COLOR: ORANGE OUTER DIAMETER: .025IN (.63MM)
	25 GAUGE COLOR: RED OUTER DIAMETER: .020IN (.53MM)
	27 GAUGE COLOR: WHITE OUTER DIAMETER: .016IN (.42MM)

Age of child	Weight of child (kg)	Median spinal cord depth (cm)	Length of needle (cm)
Neonates and infants	1-5	0.9-1.7	2
12-18 months	10	2.4	3
4 years	15	2.6	3 or 4
6 years	20	2.8	4
9 years	30	3.2	4
12 years	40	3.6	5
14 years	50	4	5-6

- Sterile CSF collection tubes (preferably polypropylene, leak-resistant screw-cap tubes).
- Sterile gloves and personal protective equipment.
- Skin antiseptic solution (e.g., chlorhexidine/alcohol preparation according to hospital policy)
- Sterile drapes and gauze
- Adhesive dressing/plaster for puncture site
- Sharps disposal container
- Labels and request forms.
- Transport container.
- Cryovials (if storage is required).

Polypropylene tubes with low protein-binding properties and secure caps are recommended for CSF collection to reduce contamination and leakage risks.

(7) Procedure

(7.1) CSF Collection Procedure

(7.1.1) Timing: Specimens should preferably be collected before antimicrobial therapy is initiated wherever possible. However, critical antimicrobial treatment must not be delayed if the lumbar puncture is temporarily contraindicated, unsafe, or delayed.

(7.1.2) Pre-Procedure Preparation: Confirm patient identity, explain the procedure to caregivers, assemble all necessary equipment, and verify indications against absolute and relative contraindications.

(7.1.3) Contraindications Assessment:

- **Absolute Contraindications:** Glasgow Coma Scale (GCS) <8 or a deteriorating/fluctuating level of consciousness; clear signs of raised intracranial pressure (e.g., diplopia, abnormal pupillary responses, decerebrate or decorticate posturing, or Cushing's triad: low HR + elevated BP + irregular respirations); and papilloedema. *Note: a bulging fontanelle in the absence of other signs of raised intracranial pressure is not an absolute contraindication to a lumbar puncture.*
- **Relative Contraindications:** Septic shock or haemodynamic compromise; significant respiratory compromise; new focal neurological signs or recent seizures with persisting reduced consciousness; a seizure within the previous 30 minutes and/or an ongoing decreased conscious state following a seizure; and coagulopathy (INR >1.5 or platelets <50 x 10⁹/L for a child on anticoagulant medication).

(7.1.4) Positioning

- LP may be performed with the child sitting up or lateral decubitus position (with proceduralist's dominant hand at caudal end)

Sitting position



Lateral decubitus position



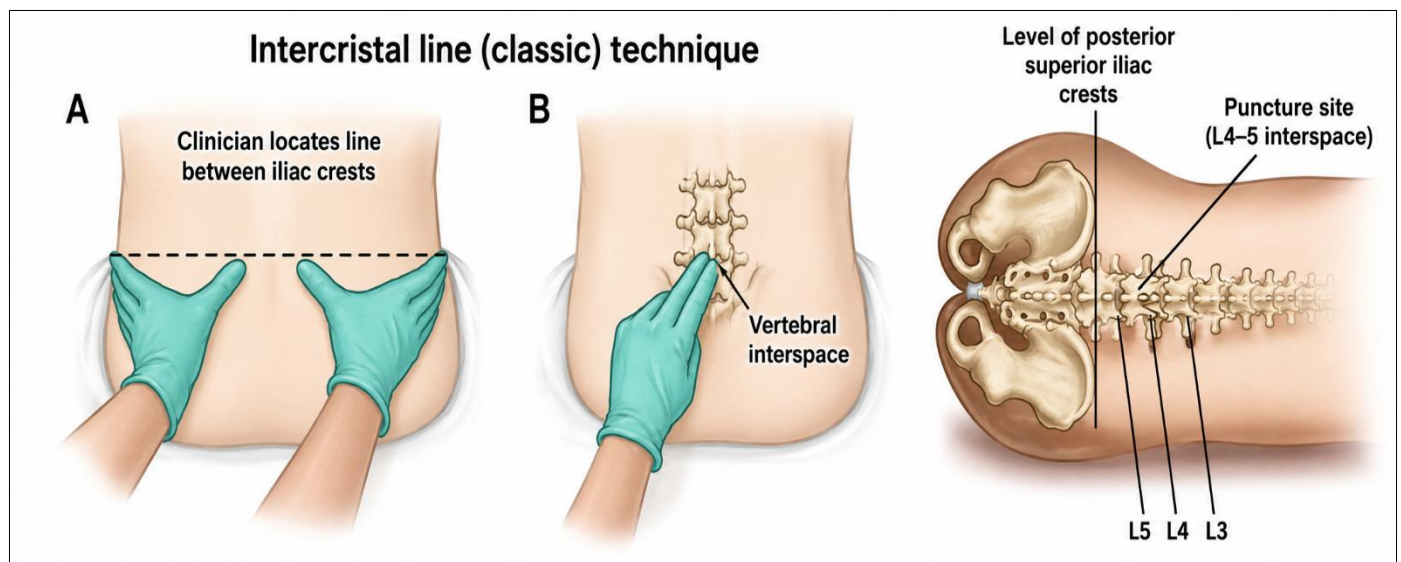
- Position back and bottom close to edge of bed
- Aim for maximum flexion of spine (curl into foetal position), but avoid over-flexing neck, especially in infants, as this may cause airway obstruction and respiratory compromise
- Position plane of the back at 90° to the bed and make sure hips and shoulders are in line with each other

(7.1.5) Hand Hygiene and Personal Protective Equipment

- Perform proper hand hygiene.
- Wear sterile gloves and appropriate protective equipment.

(7.1.6) Identify and Mark the Puncture Site

- Locate the **L3–L4 or L4–L5 intervertebral space** (line joining iliac crests—Tuffier's line).
- Draw an imaginary line between the top of the iliac crests. This intersects the spine at approximately the L3-4 interspace (mark this if necessary)



(7.1.7) Skin Preparation

- Clean the area with antiseptic solution (e.g., povidone-iodine or chlorhexidine) in a circular motion from centre outward.
- Allow the area to dry.
- Drape the site with sterile drapes.

(7.1.8) Local Anaesthesia (if applicable)

- Infiltrate local anaesthetic (e.g., lidocaine) at the puncture site.

(7.1.9) Needle Insertion

- Insert the sterile spinal needle with stylet in the midline, slightly toward the umbilicus.
- Advance slowly until a “give” is felt indicating entry into the subarachnoid space.

(7.1.10) CSF Collection

- Remove the stylet and allow CSF to flow.
- Collect CSF into sterile tubes in appropriate order.
 - The first sample should be labelled # 1 and should be used for bacterial culture if the cell count is to be done in Haematology
 - The second sample to be collected should be labelled # 2 and should be used for Chemistry testing – glucose and protein levels
 - The third sample to be collected should be labelled # 3 and should be used for white blood cell (WBC) and RBC cell count.
- Observe the appearance of CSF:
 - Normal CSF: clear and colourless.



- Cloudy/turbid CSF may indicate increased cellular content.



- Blood-stained CSF may occur due to traumatic lumbar puncture.



(7.1.11) Completion of Procedure

- Reinsert the stylet before withdrawing the needle.
- Apply gentle pressure with sterile gauze.
- Cover the site with a sterile dressing.

(7.1.12) Immediate post-procedure Care

- Monitor vital signs and neurological status.
- Keep the child comfortable; encourage rest.
- Observe for complications (e.g., headache, bleeding, apnea in neonates).

(7.2) Labelling and Documentation

Immediately after collection label specimen with:

- Patient name/identifier.
- Date and time of collection.
- Specimen type (CSF).
- Collector details.

The request form should include:

- Clinical diagnosis/suspicion.
- Age of patient.
- Relevant clinical details.
- Date and time of collection.
- Any abnormal appearance of CSF.

(7.3) Transportation of CSF Samples

- **Speed of Transport:** Send CSF specimens to the laboratory immediately after collection.
- **Chain of Custody:** Maintain proper specimen identification and verification throughout transport.
- **Containment:** Transport samples inside approved, leak-proof biological specimen containers.
- **Orientation:** Keep the collection tubes upright during transportation to significantly reduce the risk of leakage or capsular contamination.

- **Temperature and Time Considerations:** CSF should ideally be processed as soon as possible. Delays in processing can cause rapid lysis of leukocytes, which severely alters cellular analysis, cell counts, and cytology studies. Never refrigerate samples intended for bacterial culture, as fastidious organisms like *Neisseria meningitidis* or *Streptococcus pneumoniae* may fail to survive cold storage

(8) Storage of CSF Samples

(8.1) Short-term storage

- Store according to laboratory requirements.

(8.2) Long-term storage

- Aliquot CSF into sterile cryovials.
- Avoid repeated freeze–thaw cycles as they may alter specimen components.
- Store samples at ultra-low temperature (approximately –80°C) when long-term preservation is required.

(9) Post-Procedure Instructions

- Monitor the child meticulously following the lumbar puncture for any signs of:
- Active bleeding or cerebrospinal fluid leakage from the puncture site.
- Neurological changes, fluctuating consciousness, or signs of herniation.
- Persistent or worsening discomfort, spinal stiffness, or severe headache.
- Systemic or local signs of infection (e.g., fever, redness at the site).
- Respiratory compromise or sudden apnoea, especially in neonates and young infants.

(10) Infection Prevention and Safety

- Follow standard precautions during the procedure.
- Use appropriate personal protective equipment.
- Dispose of sharps according to institutional policy.
- Handle CSF as a potentially infectious biological specimen.
- Ensure safe transport according to biological specimen transport guidelines.

(11) Follow-Up

- Review CSF laboratory results with clinical findings.
- Initiate appropriate treatment based on results.
- Document complications related to the procedure or specimen handling.

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