

Protocol for collecting CSF samples for metabolic screening

Concerns the following analyses:

- Biogenic amine neurotransmitter metabolites
- Pterines
- 5-methyl-tetrahydrofolate (5-MTHF)
- Amino acids
GABA
- Pilocarpic acid.

General considerations

- Preferably collect CSF samples between 8 and 10 h in the morning, before any medication is taken.
- Always mention the actual medication on the request form.
- Collect the CSF samples in small clean tubes without additives.
- Due to the ventriculo-lumbar concentration gradient for homovanillic acid (HVA) and 5-hydroxyindoleacetic acid (5-HIAA), lumbar CSF samples should be taken in fractions in order to analyze the right fraction for these neurotransmitter metabolites.

Sample collection and storage

- For newborns up to six months of age: collect a sample of 1-2 ml.
- For ages over six months collect fractions in separate **numbered** tubes according to the following scheme:

Age	Fraction 1 (# ml)	Fraction 2 (# ml)	Fraction 3 (# ml)
< 2 y	2	1	1
2 – 16 y	4	2	2
> 16 y	8	4	4

- In case of blood contamination immediately centrifuge samples prior to freezing and transfer the clear supernatants to labeled clean tubes
- Because of the light sensitivity of the pterins, wrap all tubes in aluminum foil.
- **Freeze samples immediately at -80°C** and store until shipment.

Shipment

- Send the frozen samples on sufficient dry ice, together with a completed laboratory form (available at www.labgmd.nl) to our laboratory by (international) courier.