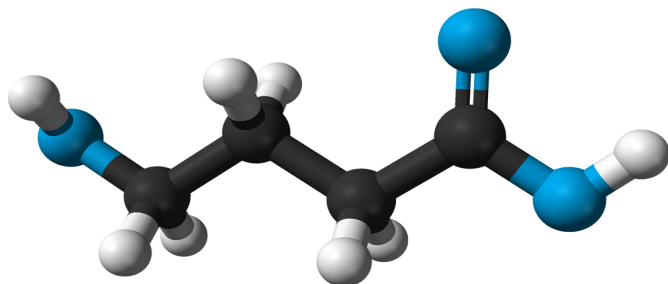


Blood GHB Extraction Procedure



UCT Part Numbers

ZSGHB020
Clean Screen® GHB
200 mg, 10 mL

SBSTFA-1-1
Selectra-Sil BSTFA
with 1% TMCS 1g vial

Procedure:

1. Prepare Sample

- To 1 mL blood sample add internal standard and 0.5 mL of 100 mM phosphate buffer (pH 6.0).
- Mix/vortex.
- Rock for 10 minutes.
- Centrifuge for 10 minutes at 2700 rpm.

2. Condition Clean Screen® GHB Extraction Column

- 1 x 3 mL CH₃OH
 - 1 x 3 mL D.I. H₂O
 - 1 x 1 mL 100 mM phosphate buffer (pH= 6.0)
- Note:** Aspirate at < 3 inches Hg to prevent sorbent drying.

3. Apply Sample

- Place centrifuge tubes into vacuum manifold for collection.
- The sample loading is collected.
- Decant sample onto column. Aspirate at about 1 inch Hg.
- After the sample is off the columns apply full vacuum for about 15 seconds to remove any residual blood.

4. Elute GHB

- Remove centrifuge tubes, set aside.
- Place clean centrifuge tubes into vacuum manifold for collection.
- 1 x 2 mL of CH₃OH/NH₄OH (99:1).
- Aspirate at about 1 inch of Hg.

5. Concentrate

- Remove test tubes from vacuum manifold.
- Vortex the sample prior to concentrating.
- Evaporate to dryness at 60 °C using a stream of nitrogen.

6. Sample Clean Up

- Add 200 µL of dimethylformamide.
- Add 1 mL of hexane saturated with dimethylformamide.
- Rock for 5 minutes.
- Centrifuge at 5 minutes at 2700 rpm.
- Transfer lower dimethylformamide layer to a clean test tube.

Note: If necessary transfer all liquid to a clean tube and allow to separate, then proceed to extract the lower layer

7. Concentrate

- Evaporate to dryness at 50 °C using a stream of air or nitrogen.

8. Derivatize

- Add 25 µL ethyl acetate and 25 µL BSTFA w 1% TMCS.
- Mix/vortex.
- Heat at 70 °C for 30 minutes.

9. Quantitate

- Inject a 1 to 2 µL of the sample onto GC/MS.



Compound	Primary Ion	Secondary	Tertiary	Cerilliant #
GHB-D6-di-TMS	239	240	241	G-006
GHB-di-TMS	233	234	235	G-001

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