



BLOOD GHB EXTRACTION PROCEDURE

Part #: ZSGHB020

By: Mr. Jim Oeldrich,
Wisconsin State Crime Lab, Milwaukee, WI

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 less than 3 inches of 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.
(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.

<u>Compound</u>	<u>Primary Ion *</u>	<u>Secondary</u>	<u>Tertiary</u>	<u>Cerilliant #</u>
GHB-D6-di-TMS	239,	240,	241	G-006
GHB-di-TMS	233,	234,	235	G-001

** Part # SBSTFA-1-1,10,25,100