1/4/2024

Worklist: 6633

<u>LAB CASE</u> <u>ITEM</u> <u>ITEM TYPE</u> <u>DESCRIPTION</u>

C2023-2239 2 UCK AM 6 Urine GHB

REVIEWED

By Britany Wylie at 2:49 pm, Jan 07, 2024





AM 6: Urine GHB Screening Extraction

Extraction Date: 01/04/24 Analyst: Anne Nord

Mobile phase A: 0.1% Formic Acid in Water Mobile phase B: 0.1% Formic Acid in MeOH

0.1% formic acid in methanol

Blank Urine Lot: 1324

Column: Agilent poroshell 120 (4.6x50mm, 2.7um)

LCMS-OOO ID: 69679 GHB Control Lot: 1424

Pre-Analytic:

- Working Solution: Preparation of 200,000 ng/mL Positive Control Working Solution: Add 200μL of GHB 1 mg/mL stock solution to 800μL negative urine.
- Preparation of 10,000 ng/mL Positive Control: Add 10μL of GHB 20,000 ng/mL working solution to 190 μL negative urine.
- ☑ 2. Check levels of mobile phases and needle wash refill as needed. Ensure waste is not full.
- ☑ 3. Ensure correct column is installed and begin mobile phase flow allow to equilibrate ~ 30 minutes.

Analytic:

- ☑ 3. Label ALS or LCMS vials for positive control, negative control, and case samples. Place insert in all vials.
- ☑ 4. Place on tube rocker at ambient temp for approx. 10 minutes.
- Σ 5. Pipette positive and negative controls (for negative control, 200 μL urine will be added to the appropriate tube). Add 200μL urine to each centrifuge tube for case samples.
- ⊠ 6. Add 100μL of the GHB-D6 Internal Standard Working Solution to each tube.
- X 7. Add 900μL of 0.1% formic acid in methanol to each tube. Vortex. Made fresh 100 ul 195725 formic acid fisher, 100 ml Honeywell lot ED456-US 01-4-24 AMN
- \boxtimes 8. Centrifuge at ~3400 rpm for 15 minutes.
- × 9. Add 100μL 0.1% formic acid in water to each vial insert.
- ⊠ 10. Transfer 10μL of sample from each centrifuge tube to the corresponding vial insert (avoid disturbing the pellet at the bottom). Vortex.

Post-Analytic

- ☑ 1. Open quantitation software and create a new quantitation batch.
- ☑ 2. Using the positive control, a 1-point calibration curve will be established. The curve will be set to linear, non-weighted and origin set to force.
- ☑ 3. If a sample gives a response that is greater than 10,000 ng/mL, a statement on the report will be included saying that preliminary testing indicated a possible presence of an elevated level of GHB and that it is recommended that the sample be sent to a private lab for quantitation. If a sample gives a response between 7,000 and 10,000 ng/mL, an inconclusive statement can be added to the report.
- ☑ 4. The S/N for samples and controls at and over 10,000 ng/mL must be 5 or greater
- ⊠ 5. Case samples and negative controls will generally be considered negative if the calculated concentration is less than 7,000 ng/mL.
- ☑ 6. Central File Packet to include: LIMS Worklist, Method Checklist, Working solution prep sheet(s), Calibration and Control Reports

COMMENTS:

GHB controls

200000 ng/ml working solution 200 ul 1 mg/ml GHB into 800 ul neg urine (1324)

ppd 1/4/24 Exp 7/4/24 lot 1424 by AMN

Drug lot expiration GHB FE04111903 5/1/2024

20000 ng/nl working internal standard solution 1ml 100ul/ml GHB D6 stock in 4000 ul methanol

Ppd 1/4/24 exp 1/4/25 lot GHB-D6 01424 by amn

Drug lot

GHB-D6 FE07031801

^{*} AM 6 Control: add 10uL of working solution to 190uL negative urine and extract. Approx conc 10,000ng/mL



GHB Screen results

Batch results D:\MassHunter\Data\2021 Data\ghb 010424\QuantResults\ghb.batch.bin

Calibration Last Update 1/4/2024 12:45:35 PM

Instrument 69679 **Type** Cal

Acq. Method GHB urine screen.m Sample Position Vial 2

Sample Position Vial **Injection Volume** 3

Acq. Date-Time 1/4/2024 11:09:10 AM

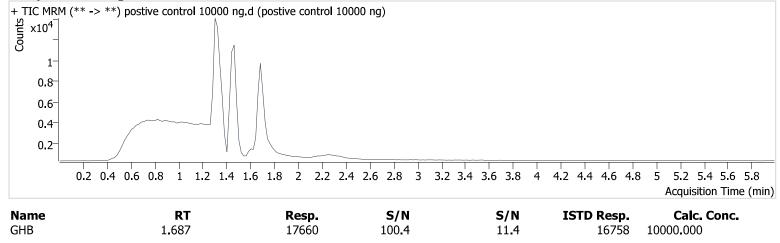
Sample Info.

Data File Sample Operator Comment

postive control 10000 ng.d postive control 10000 ng

Anne Nord

Sample Chromatogram





GHB Screen results

Batch results D:\MassHunter\Data\2021 Data\ghb 010424\QuantResults\ghb.batch.bin

Calibration Last Update 1/4/2024 12:45:35 PM

Instrument 69679 **Type** Sample Acq. Method

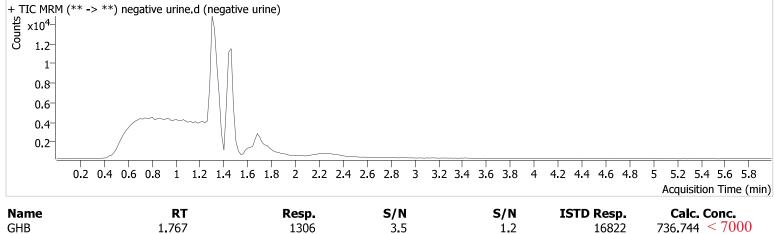
GHB urine screen.m **Sample Position** Vial 3

Injection Volume 3 Acq. Date-Time 1/4/2024 11:15:37 AM

Sample Info.

Data File Sample Operator Comment negative urine d negative urine Anne Nord





3.5

1.2

16822

736.744 < 7000