



## GHB (GAMMA-HYDROXYBUTYRIC ACID)

### FORENSIC USE ONLY

#### INTENDED USE

The GHB test strips can detect the presence of GHB in human urine at concentrations from 10 µg/ml to 50 µg/ml. The range of detection compares favorably with the manufacturer's specified working range shown on device label.

Test	Cut-off µg/ml
GHB	10

The GHB test is used to obtain a visual, qualitative result and is intended for forensic use only

**This assay provides only a preliminary result. Clinical consideration and professional judgment must be applied to any drug of abuse test result, particularly in evaluating a preliminary positive result. In order to obtain a confirmed analytical result, a more specific alternate chemical method is needed. Gas chromatography/mass spectroscopy (GC/MS) is the preferred confirmation method.**

#### SUMMARY AND EXPLANATION

Gamma-hydroxybutyric acid (GHB) is a colorless, odorless chemical and has become one of the most dangerous illicit drugs of abuse today. It has become known as one of the "date rape" drugs and there is an increasing trend that GHB is being used as a recreational drug.

Manufactured lots of GHB test strips were tested for sensitivity, assay response range, cross-reactivity, precision and stability. Specifically, the lowest and highest ranges of detectable response were determined. The precision of the assay response between device lots was also examined. The influence of common urine interferants as well as compounds with similar chemical structure was also determined. Stability of the strip response after exposure to high temperatures and moisture in and outside of original packaging was also examined.

The devices appear to have good precision. Within lot precision was within a color scale unit based on the IDS color chart from day to day and between storage containers. The color intensities were also within a color unit between device lots.

The specificity of test strip appears high and will not detect closely related drug structures at concentrations as high as 50 µg/ml.

The test strip appears to be relatively resistant to common interferants but a strong false positive signal can be produced with extreme concentrations of L-Ascorbic acid. The impact of this interference is expected to be low in human urine or serum as concentrations are expected to be much less. Further titration is required to ascertain at what concentration this effect is observed. L-ascorbic acid may be more problematic for beverage testing applications.

Highly colored samples such as those urines containing high amounts of blood or riboflavin can interfere with interpretation of the color signal. Grossly hemolyzed samples will require clean-up or possibly re-sampling.

The test strip reaction zone appears relatively sensitive to moisture. Storage of the devices in the original packaging materials will likely be critical for maintaining stability.

Shelf Life of product is determined to be 9 months from the date of manufacture and will be indicated on the product label.

#### TEST PRINCIPLE

GHB-DH catalyses the reaction of GHB and NAD to produce NADH, and a diaphorase couple tetrazolium dye reaction results in the production of a purple dye complex. The reagents were stabilized and used to produce the dip test to screen for low levels of GHB in urine samples. The dip test has a 3.4 x 5 mm reagent pad on a 80mm strip. The reagent pad is dipped into the test sample for 5 secs and results can be read by comparing pad to color chart after 2 minutes.

#### REAGENTS & MATERIALS SUPPLIED

\* GHB Test Strip / Vitamin C test strip

\* One instruction sheet

#### MATERIAL REQUIRED BUT NOT PROVIDED

\* Timer

\* Specimen collection cup (for dip test)

\* External positive and negative controls (OPTIONAL)

#### WARNINGS AND PRECAUTIONS

\* Urine specimens may be potentially infectious. Proper handling and disposal methods should be established.

\* Avoid cross-contamination of urine samples by using a new specimen collection container for each urine sample.

\* Test device should remain sealed until ready for use.

\* Do not use the test kit after the expiration date.

\* A positive test result does not always mean an individual has taken the drug illegally as the drug can be administered legally.

\* Do not store and or expose reagent kits at temperature greater than 30°C. Do not freeze.

#### STORAGE

The GHB test should be stored at 2-30°C (36-86°F) in the original sealed pouch. Do not freeze. Do not store and or expose reagent kits at temperature greater than 30°C (86°F).

#### SPECIMEN COLLECTION AND HANDLING

Fresh urine does not require any special handling or pretreatment. A fresh urine sample should be collected in the container provided. Alternately, a clean, dry plastic or glass container may be used for specimen collection. If the specimen will not be tested after the specimen collection, the specimen may be refrigerated at 2-8°C up to 2 days or frozen at -20°C for longer period of time. Specimens that have been refrigerated must be equilibrated to room temperature prior to testing. Specimens previously frozen must be thawed and mixed thoroughly prior to testing.

Note: Urine specimens and all materials coming in contact with them should be handled and disposed as if capable of transmitting infection. Avoid contact with skin by wearing gloves and proper laboratory attire.

#### ASSAY PROCEDURE

##### Preparation

1. If specimen or controls have been stored at refrigerated temperatures, allow them to warm to room temperature before testing.
2. Do not open test device pouch until ready to perform the test.

##### Testing: GHB

FOR DIP STRIP GHB DRUG TEST

1. Issue a specimen cup to the individual to be tested.
2. Collect the urine into the collection cup
3. Remove test strip from the left channel of the sealed pouch.
4. Immerse sampling tip into the urine specimen for about 5 seconds and then place the test on a flat surface.
5. Read initial Results at 2 minutes. Test may take up to 10 minutes to detect lower concentrations of GHB near the 10 µg/ml cutoff.

#### INTERPRETATION OF RESULTS

Use the color chart on the product pouch to interpret GHB levels at the three indicated semi-quantitative GHB concentrations. The 0 µg/ml level indicates that no significant GHB is present, the 10 µg/ml level and the 50 µg/ml level indicates a presumptive positive.

**IMPORTANT: The GHB test has been shown to cross react with vitamin C (ascorbic acid). If the GHB test strip indicates that the sample is positive for GHB (10 µg/ml or 50 µg/ml), use the Vitamin C test strip to determine the presence of ascorbic acid.**

##### Testing: Vitamin C

1. Dip reagent end of strip in urine and remove immediately.
2. While removing, run the edge of strip against the rim of the urine container to remove excess urine.
3. Compare the test areas with the color chart after 30 seconds

#### SAMPLE INTERFERENCE

The following list of compounds were added to urine pre-spiked with 100 µg/ml of GHB. The device signal intensities were compared directly to those obtained with matched urine samples without interferants added. Increased color signal was noted with L-Ascorbic acid spiked urine. Highly colored urine test samples such as those containing high concentrations of (-) Riboflavin and Hemoglobin made signal interpretation difficult or impossible. A slight diminishment in signal intensity was observed with EDTA, potassium tetra oxalate and low pH urine.

Interferants	Interferants (mg/dl) Unless otherwise indicated	Average Observed Signal Intensity
GHB	-	5.5
Ethanol	2000	5.5
Urea	6000	5.5
Sodium Chloride	6000	5.5
Human Serum Albumin	500	5.5
Creatinine	500	5.5
Glucose	3000	5.5
L-Ascorbic acid	500	10
(-) Riboflavin	10	Too Yellow
Bilirubin, mixed isomers	30	5.5
Bile Salts	5mg/ml	5.5
Hemoglobin	800	Too Red
Urine pH	pH 5.0	4.5
Urine pH	pH 10.0	5.5
Disodium EDTA	25mM	4.5
Sodium Citrate	40mM	6
Heparin	100 USP/4ml	5.5
Potassium Oxalate	2mg/ml	4.5
Sodium Fluoride	1.25mg/ml	5.5

#### LIMITATIONS OF PROCEDURE

\* The assay is designed for use with human urine only.

\* A positive result with any of the tests indicates only the presence of a drug/metabolite and does not indicate or measure intoxication.

\* There is a possibility that technical or procedural error as well other substances as factors not listed may interfere with the test and cause false results. See SPECIFICITY for lists of substances that will produce positive results, or that do not interfere with test performance.

\* If adulteration is suspected, the test should be repeated with new sample.

#### PERFORMANCE CHARACTERISTICS

##### Precision

The device precision was tested three times over a period of four days from three different reagent storage bottles. Three GHB concentrations in human urine were examined. The lowest and highest concentrations specified by the manufacturer (0, 500 µg/ml) and a concentration in between (.50ug/ml) was examined in duplicate. Similar signals intensities were noted within the test lot at each of the tested concentrations

	Bottle 1		Bottle 2		Bottle 3	
	6/22/10		6/23/10		6/25/10	
GHB ug/ml	Run 1		Run 2		Run 3	
0	2	2.5	2	2	2.5	2.5
50	5.5	5.5	5	5	4.5	4.5
500	10	9.5	9.5	9.5	9	9

#### SPECIFICITY

##### Device Cross-Reactivity:

The following structurally similar compounds were checked for cross-reaction.

None of the tested compounds produced a color change at 500µg/ml greater than that observed for 10µg/ml of GHB. The calculated cross-reaction is predicted to be less than 2%.

Compound	Percent Cross Reactivity
GHB	100%
1,4-Butanediol	<2%
Gamma-Butyrolactone	<2%
Succinic Semialdehyde	<2%
Gabapentin	<2%
Alpha-Hydroxy-Gamma-Butyrolactone	<2%

#### REFERENCES

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