

Overview

Useful For

Detecting drug use involving benzodiazepines such as alprazolam, chlordiazepoxide, clonazepam, diazepam, midazolam, oxazepam, temazepam, clobazam, flunitrazepam, flurazepam, lorazepam, prazepam, triazolam, and zolpidem, in urine specimens handled through the chain-of-custody process

Additional Tests

Test ID	Reporting Name	Available Separately	Always Performed
COCH	Chain of Custody Processing	No	Yes
ADLTX	Adulterants Survey, CoC, U	Yes	Yes

Testing Algorithm

Testing for adulterants will be performed on all chain of custody urine samples per regulatory requirements.

Method Name

Liquid Chromatography-Tandem Mass Spectrometry (LC-MS/MS)

NY State Available

Yes

Specimen

Specimen Type

Urine

Specimen Required

Supplies: Chain-of-Custody Kit (T282)

Container/Tube: Chain-of-Custody Kit containing the specimen containers, seals, and documentation required.

Specimen Volume: 20 mL

Collection Instructions: Collect specimen in the container provided, seal, and submit with the associated documentation to satisfy the legal requirements for chain-of-custody testing.

Additional Information: Submitting <20 mL will compromise the ability to perform all necessary testing.

Forms

[Chain-of-Custody Request](#) is included in the Chain-of-Custody Kit (T282).

If not ordering electronically, complete, print, and send a [Therapeutics Test Request](#) (T831) with the specimen.

Specimen Minimum Volume

5 mL

Reject Due To

Gross hemolysis	Reject
Gross icterus	Reject

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Urine	Refrigerated (preferred)	7 days	
	Frozen	14 days	

Clinical and Interpretive
Clinical Information

Benzodiazepines are any of a group of compounds having a common molecular structure and acting similarly as depressants of the central nervous system. As a class of drugs, benzodiazepines are among the most commonly prescribed drugs in the western hemisphere because of their efficacy, safety, low addiction potential, minimal side effects, and high public demand for sedative and anxiolytic agents.

Chain of custody is required whenever the results of testing could be used in a court of law. Its purpose is to protect the rights of the individual contributing the specimen by demonstrating that it was under the control of personnel involved with testing the specimen at all times; this control implies that the opportunity for specimen tampering would be limited. This includes a record of the disposition of a specimen to document the personnel who collected it, who handled it, and who performed the analysis. When a specimen is submitted in this manner, analysis will be performed in such a way that it will withstand regular court scrutiny.

Reference Values

Negative

Cutoff concentrations:

Alprazolam by LC-MS/MS: 10 ng/mL

Alpha-Hydroxyalprazolam by LC-MS/MS: 10 ng/mL

Chlordiazepoxide by LC-MS/MS: 10 ng/mL

Clonazepam by LC-MS/MS: 10 ng/mL

7-Aminoclonazepam by LC-MS/MS: 10 ng/mL

Diazepam by LC-MS/MS: 10 ng/mL

Nordiazepam by LC-MS/MS: 10 ng/mL

Midazolam by LC-MS/MS: 10 ng/mL

Alpha-Hydroxy Midazolam by LC-MS/MS: 10 ng/mL

Oxazepam by LC-MS/MS: 10 ng/mL

Temazepam by LC-MS/MS: 10 ng/mL

Clobazam by LC-MS/MS: 10 ng/mL

N-Desmethyloclobazam by LC-MS/MS: 10 ng/mL

Flunitrazepam by LC-MS/MS: 10 ng/mL

7-Aminoflunitrazepam by LC-MS/MS: 10 ng/mL

Flurazepam by LC-MS/MS: 10 ng/mL

2-Hydroxy Ethyl Flurazepam by LC-MS/MS: 10 ng/mL

Lorazepam by LC-MS/MS: 10 ng/mL

Prazepam by LC-MS/MS: 10 ng/mL

Triazolam by LC-MS/MS: 10 ng/mL

Alpha-Hydroxy Triazolam by LC-MS/MS: 10 ng/mL

Zolpidem by LC-MS/MS: 10 ng/mL

Zolpidem Phenyl-4-Carboxylic acid by LC-MS/MS: 10 ng/mL

Interpretation

Benzodiazepines are extensively metabolized, and the parent compounds are not detected in urine. This test screens for (and confirms) the presence of:

Alprazolam

Alpha-hydroxyalprazolam (metabolite of alprazolam)

Chlordiazepoxide

Clonazepam

7-Aminoclonazepam (metabolite of clonazepam)

Diazepam (separate prescribable drug and metabolite of medazepam)

Nordiazepam (metabolite of clorazepate, halazepam, prazepam, diazepam and medazepam)

Midazolam

Alpha-hydroxy midazolam (metabolite of midazolam)

Oxazepam (separate prescribable drug and metabolite of clorazepate, halazepam, prazepam, medazepam, temazepam, and diazepam)

Temazepam (separate prescribable drug and metabolite of medazepam and diazepam)

Clobazam

N-Desmethyloclobazam (metabolite of clobazam)

Flunitrazepam

7-minoflunitrazepam (metabolite of flunitrazepam)

Flurazepam

2-Hydroxy ethyl flurazepam (metabolite of flurazepam)

Lorazepam

Prazepam

Triazolam

Alpha-hydroxy triazolam (metabolite of triazolam)

Zolpidem

Zolpidem phenyl-4-carboxylic acid (metabolite of zolpidem)

The clearance half-life of long-acting benzodiazepines is more than 24 hours. It takes 5 to 7 half-lives to clear 98% of a drug dose. Therefore, the presence of a long-acting benzodiazepine greater than the limit of quantification indicates exposure within a 5 to 20-day interval preceding specimen collection. Following a dose of diazepam, the drug and its metabolites appear in the urine within 30 minutes. Peak urine output is reached between 1 and 8 hours. See Mayo Clinic Laboratories Drugs of Abuse Testing Guide at <https://www.mayocliniclabs.com/test-info/drug-book/index.html> for additional information including metabolism, clearance (half-life), and approximate detection times.

Cautions

No significant cautionary statements.

Clinical Reference

1. Gudin JA, Mogali S, Jones JD, Comer SD: Risks, management, and monitoring of combination of opioid, benzodiazepines, and/or alcohol use. *Postgrad Med*. 2013;125(4):115-30. doi: [10.3810/pgm.2013.07.2684](https://doi.org/10.3810/pgm.2013.07.2684)
2. Manchikanti L, Abdi S, Atluri S, et al: American Society of Interventional Pain Physicians (ASIPP) guidelines for responsible opioid prescribing in chronic non-cancer pain: Part 2-guidance. *Pain Physician*. 2012;15(3 Supp):S67-116
3. Barkin RL: In: Baselt RC, ed. *Disposition of Toxic Drugs and Chemicals in Man*. 8th ed. Biomedical Publications; 2008
4. Langman LJ, Bechtel LK, Meier BM, Holstege C: Clinical toxicology. In: Rifai N, Horvath AR, Wittwer CT, eds.

Tietz Textbook of Clinical Chemistry and Molecular Diagnostics. 6th ed. Elsevier; 2018: 1328-1333

Performance

Method Description

Benzodiazepines are extensively metabolized by the liver and subsequently exist in the urine primarily as conjugated esters (-glucuronides). The conjugated metabolites are cleaved during a mild hydrolysis utilizing the enzyme glucuronidase. Stable isotope forms of the compounds are added as internal standards to account for extraction losses. An aliquot of the hydrolyzed sample is diluted and the analytes are separated by liquid chromatography tandem mass spectroscopy (LC-MS/MS) and analyzed by multiple reaction monitoring. (Unpublished Mayo method)

PDF Report

No

Day(s) Performed

Tuesday through Saturday

Report Available

2 to 6 days

Specimen Retention Time

14 days

Performing Laboratory Location

Rochester

Fees and Codes

Fees

- Authorized users can sign in to [Test Prices](#) for detailed fee information.
- Clients without access to Test Prices can contact [Customer Service](#) 24 hours a day, seven days a week.
- Prospective clients should contact their Regional Manager. For assistance, contact [Customer Service](#).

Test Classification

This test was developed and its performance characteristics determined by Mayo Clinic in a manner consistent with CLIA requirements. This test has not been cleared or approved by the U.S. Food and Drug Administration.

CPT Code Information

80347

G0480 (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC Value
BNZX	Benzodiazepines Conf, CoC, U	90890-5



Result ID	Test Result Name	Result LOINC Value
608452	Benzodiazepines Immunoassay Screen	14316-4
608280	Alprazolam by LC-MS/MS	59615-5
608281	Alpha-Hydroxyalprazolam by LC-MS/MS	16348-5
608282	Chlordiazepoxide by LC-MS/MS	20522-9
608283	Clonazepam by LC-MS/MS	16229-7
608284	7-aminoclonazepam by LC-MS/MS	51776-3
608285	Diazepam by LC-MS/MS	16227-1
608286	Nordiazepam by LC-MS/MS	16228-9
608287	Midazolam by LC-MS/MS	16233-9
608288	Alpha-Hydroxy Midazolam by LC-MS/MS	59590-0
608289	Oxazepam by LC-MS/MS	16201-6
608290	Temazepam by LC-MS/MS	20559-1
608291	Clobazam by LC-MS/MS	59635-3
608292	N-Desmethyclobazam by LC-MS/MS	In Process
608293	Flunitrazepam by LC-MS/MS	20528-6
608294	7-aminoflunitrazepam by LC-MS/MS	51777-1
608295	Flurazepam by LC-MS/MS	16231-3
608296	2-Hydroxy Ethyl Flurazepam by LC-MS/MS	In Process
608297	Lorazepam by LC-MS/MS	17088-6
608298	Prazepam by LC-MS/MS	17479-7
608299	Triazolam by LC-MS/MS	16232-1
608300	Alpha-Hydroxy Triazolam by LC-MS/MS	49876-6
608301	Zolpidem by LC-MS/MS	72770-1
608302	Zolpidem Phenyl-4-Carboxylic acid by LC-MS/MS	72768-5
608449	Benzodiazepines Interpretation	16195-0
608450	Chain of Custody	77202-0