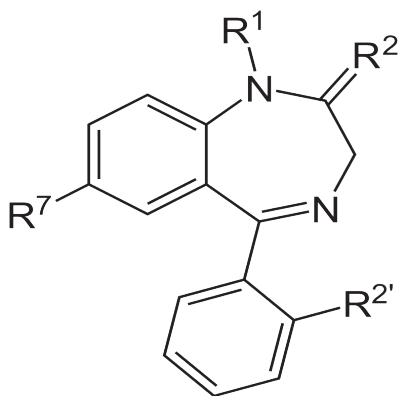


Benzodiazepine Screen: Blood, Serum, Urine and Tissue For GC-GC/MS Using: 200 mg Clean Screen® BNZ Extraction Column



UCT Part Numbers

CSBNZ203
Clean Screen® BNZ
200 mg, 3 mL

Procedure:

1. Prepare Sample

- To 1 mL of 100 mM phosphate buffer (pH= 6) add internal standards*
- Add 1 mL blood/ urine or 1 g of (1:4) tissue homogenate
- Vortex mix. Add 3 mL of 100 mM phosphate buffer (pH= 6).
- Sample pH should be 6.0 ± 0.5
- Adjust pH accordingly with 100 mM monobasic or dibasic sodium phosphate
- Vortex and centrifuge as appropriate

2. Procedure for Urine

- To 1 mL of acetate buffer (pH 5.0) containing 5000 F units/ mL β -glucuronidase. Add internal standards* To this solution add 1 mL of urine
- Mix / Vortex
- Hydrolyze for 3 hrs at 65°C
- Allow to cool
- Centrifuge for 10 minutes at 2000 rpm and discard pellet
- Add 3 mL of 100 mM phosphate buffer (pH 6.0) and mix

3. Condition Clean Screen® BNZ Extraction Column

- 1 x 3 mL MeOH
 - 1 x 3 mL 100 mM phosphate buffer (pH = 6)
- Note:** Aspirate at < 3 inches Hg to prevent sorbent drying out

4. Apply Sample

- Load sample at 1 to 2 mL/minute

5. Wash Column

- 1 x 3 mL of 5 % (v/v) acetonitrile in 100 mM phosphate buffer (pH = 6)
- Dry column (5 minutes at > 10 inches Hg)
- 1 x 3 mL of hexane.
- Dry column (5 minutes at > 10 inches Hg)

6. Elute Benzodiazepines

- 1 x 3 mL ethyl acetate: ammonia (98:2 v/v)
- Collect eluate at 1-2 mL /minute

7. Evaporation

- Evaporate eluate under a gentle stream of nitrogen < 40°C

8. Derivatize

- Add 50 μ L acetonitrile and 50 μ L BSTFA with 1% TCMS
- Heat for 30 minutes at 70° C
- Remove from heat source to cool
- Inject 1 μ L into GC/MS



Compound	Primary ion	Secondary Ion	Tertiary Ion	Cerilliant #
Alprazolam	308	279	204	A-903
*Alprazolam-D5	513	284		A-910
Alpha-hydroxyalprazolam	318	396	383	A-907
Alpha-hydroxyalprazolam-D5	386	401		A-902
Diazepam	256	283	284	D-907
*Diazepam-D5	287	289		D-902
Lorazepam	429	430	347	L-901
*Lorazepam-D4	433	435		L-902
Nordiazepam	341	342	343	N-905
*Nordiazepam-D5	345	347		N-903
Oxazepam	429	313	430	O-902
*Oxazepam-D5	435	433		O-901
Temazepam	343	257	283	T-907
*Temazepam-D5	348	262		T-902

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