

Benzodiazepines In Whole Blood For GC OR GC/MS Confirmations Using: 300 mg Clean Screen® BNZ Extraction Column



UCT Part Numbers

ZSBNZ030
Clean Screen® BNZ
300 mg 10 mL
Without Tips

Or

ZCBNZ030
Clean Screen® BNZ
300mg 10mL
With CLEAN-THRU® Tips

Procedure:

1. Prepare Sample

- To 1 mL of pH 6 buffer add internal standards*, add 2 mL of whole blood and vortex mix
- Add 5 mL of pH 6 buffer, sonicate with a probe sonifier for ~10 seconds and centrifuge at ~2700 rpm for 15 minutes.

2. Condition Clean Screen® BNZ Extraction Column

- 1 x 3 mL Ethyl Acetate.
- 1 x 3 mL MeOH.
- 1 x 3 mL DI H₂O.
- 1 x 3 mL 0.1M phosphate buffer (pH 6.0).

3. Apply Sample

- Load sample by gravity.

4. Wash Sample

- 1 x 3 mL DI H₂O.
- 1 x 3 mL 5% acetonitrile in 0.1M phosphate buffer (pH 6.0).
- Dry columns 5 minutes at full vacuum or > 10 inches Hg.
- 1 x 3 mL Hexane.

5. Elute Benzodiazepines

- 2 x 3 mL Ethyl Acetate.

6. Dry Eluate

- Evaporate to dryness under nitrogen at ~ 55° C.
- Add external standards.*

7. Derivatize

- Add 100 µL acetonitrile and 100 µL MTBSTFA w/1% t-BDMCS.
- Heat for 30 minutes at 70° C.
- Remove from heat source to cool.
- Inject 1 µL into GC/MS-NCI.

Note: Do not evaporate MTBSTFA solution.



GC/MS Conditions
Agilent 6890/5975 inert MSD
Inlet 260 °C, Split injection 2:1
Column: ZB-5MS, 15 m, 0.25 mm ID, 0.25 µm film
Temperature program: 70 °C hold 0.25 min, ramp 20 °C per min to 300 °C, hold 4 min.
NCI parameters: Source 150 °C, Quad 106 °C, Methane as collision gas

*Suggested internal standards : Diazepam-D5 and Lorazepam-D4.

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