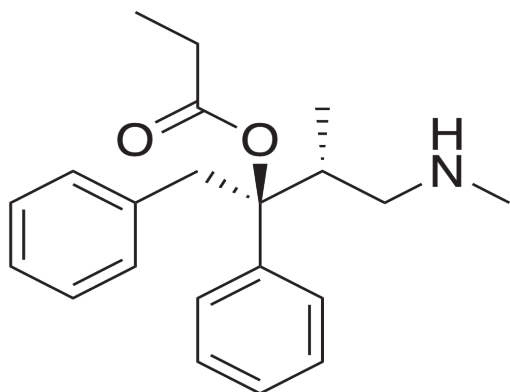


Propoxyphene and Norpropoxyphene In Blood, Plasma/Serum, Urine, Tissue By LC-MS/MS or GC-MS Using: 200 mg Clean Screen® DAU Extraction Column



UCT Part Numbers

ZSDAU020
Clean Screen® DAU
10 mL, 200 mg Column

SLDA50ID21-5UM
SELECTRA® DA HPLC
Column 50 x 2.1 mm, 5 µm

Procedure:

1. Prepare Sample

- To 1 mL of 100 mM phosphate buffer (pH=6) add internal standards.
- Add 1 -2 mL of blood, plasma/ serum, urine, or 1 g (1:4) tissue homogenate.
- Mix/vortex and let stand 5 minutes.
- Add 2 mL of 100 mM phosphate buffer (pH 6.0). Mix/vortex.
- Sample pH should be 6.0 ± 0.5 .
- Adjust pH accordingly with 100 mM monobasic or dibasic sodium phosphate.
- Centrifuge for 10 minutes at 2000 rpm and discard pellet.

2. Condition Clean Screen® Extraction Column

- 1 x 3 mL CH₃OH.
- 1 x 3 mL D.I. H₂O.
- 1 x 3 mL 100 mM phosphate buffer (pH 6.0).

Note: Aspirate at full vacuum or pressure.

3. Apply Sample

- Load at 1 to 2 mL/minute

4. Wash Column

- 1 x 3 mL D.I. H₂O.
- 1 x 3 mL 100 mM acetic acid.
- 1 x 3 mL CH₃OH.
- Dry column (5 minutes at full vacuum or pressure).

5. Elute Basic Analytes

- 1 x 3 mL CH₂Cl₂/IPA/NH₄OH (78:20:2).
- Collect eluate at 1 to 2 mL/minute.

Note: Prepare elution solvent daily
Add IPA/NH₄OH, mix, then add CH₂Cl₂ (pH 11-12)

6. Dry Eluate

- Add 100 µL of 1% HCl in Methanol to each test tube
- Evaporate to dryness at < 40 °C

7. Reconstitute / Derivatize

LC-MS/MS: Reconstitute sample in 100 µL of mobile phase
Inject 10 µL.

GC-MS: Dissolve residue in 100 µL of ethyl acetate

Note: To improve the analysis for Norpropoxyphene, the primary metabolite of Dextro-propoxyphene, add 1 drop of 35% sodium hydroxide solution to the urine sample and then after mixing bring the pH to 6 for SPE extraction. This step converts the Norpropoxyphene to Norpropoxyphene amide, a more stable compound.



LC-MS MRM Transitions

Analyte	Q1	Q3
Propoxyphene	340.0	58.0
Propoxyphene D11	351.2	64.0
Norpropoxyphene	326.0	252.0
Norpropoxyphene D5	331.0	257.0

GC-MS IONS

Analyte	Quantify Ion	Qualifier Ion 1	Qualifier Ion 2	Other Ions
Propoxyphene D5	63	120	213	255,270
Propoxyphene	58	115	208	250,265

References:

- [1] Amalfitano G, Bessard J, Vincent F, Esseric H and Bessard G Gas Chromatographic Quantitation of Dextropropoxyphene and Norpropoxyphene in Urine after Solid Phase Extraction Journal Analytical Toxicology 20:547-554 (1996)

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