



SPEware uSPE (ultraSPE) Faster / Cleaner Sample Preparation

Opiates from Whole Blood

For GC or GC-MS confirmations using:
Trace-B 50mg.

1. PREPARE SAMPLE

To 1.0ml whole blood add 2.0 ml 100 mM phosphate buffer pH = 6.0. Vortex for 30 seconds then sonicate for 15 minutes. Centrifuge sample for 5 min at 3000 rpm,

2. APPLY SAMPLE

Decant the supernatant into the Trace-B column and draw through the column at a flow of 2-5 ml/minute.

3. WASH COLUMN (All wash steps flow at 5-10 ml/min)

1 x 1ml pH 9.0 buffer***
1 x 1 ml DI H₂O;
1 x 1 ml 100 mM HCl
1 x 1 ml CH₃OH
Dry for 2 minutes at 25 psi

4. ELUTE OPIATES

1 x 2.0 ml Ethyl Acetate containing 2% NH₄OH; collect eluate at 1 to 2 ml/minute.
NOTE: Prepare elution solvent daily.

5. DRY ELUATE

Evaporate to dryness at 40 °C.

6. DERIVATIZE

Add 50 ul ethyl acetate vortex and transfer to a/s vial (**This transfer step is a very important clean up step. Do not omit.**)

Add 50 ul MSTFA. (Important—do not add MSTFA to dry down vial)

React 20 minutes at 70 °C. Remove from heat source to cool.

NOTE: Do not evaporate MSTFA solution.

7. QUANTITATE

Inject 1 to 3 ul sample onto chromatograph.

Monitor the following ions (GC/MS):

TMS-Codeine: 371**, 372, 343 TMS- D3 -Morphine : 432**, 417

TMS-D3 -Codeine: 374**, 346 TMS-Morphine: 429**, 414, 401

* Suggested internal standards for GC/MS: D3 -Codeine, D3 -Morphine

** Quantitation ion