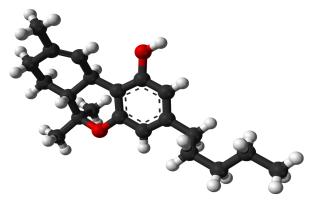
delta 9-THC (parent), delta 9-Hydroxy THC, Carboxy- delta 9-THC In Whole Blood For GC/MS Confirmations Using: 100 mg Clean Screen® SSTHC Extraction Column



UCT Part Numbers

SSTHC116

Styre Screen® THC 100 mg, 6 mL Tube

Procedure:

1. Prepare Sample

- a) To 1-2 mL whole blood add appropriate internal standards prepared in alcohol.
- b) Add drop-wise 2 mL ice cold acetonitrile.*
- c) Mix thoroughly and centrifuge.
- d) Decant acetonitrile into a clean tube. Evaporate acetonitrile under a stream or air or nitrogen to $\sim 200 \mu L$.
- e) Add 2 mL distilled water (pH~6.0-7.0)

 Note: The sample is ready to be extracted.

 Note: The acetonitrile should be cold (recommended storage in freezer at <0 C just prior to use) and it should be added very slowly to ensure proper mixing of organic phase with the whole blood. If added too quickly, the blood could precipitate to fast possibly resulting in lower recoveries.

2. Apply Sample

 Load sample directly to column without any preconditioning.

3. Wash Column

- a) Wash with 1 mL (84/15/1) Water/ Acetonitrile/ NH₄OH.
- b) Dry column thoroughly under vacuum (10 mm Hg) or positive pressure (~ 80-100 psi) for 10-15 minutes. **Note:** It is important to dry the column properly to achieve the highest recovery of all compounds. Any residual moisture will slow down the drying of the elution solvents prior to derivatization. Also, any residual moisture could reduce the reactivity of the derivatizing agent.

4. Elute THC, THC-OH, THC-COOH

- a) 1 x 3 mL Hexane/ Ethyl Acetate/ Glacial Acetic Acid (49: 49:2).
- b) Collect at 1-2 mL/ minute.

5. Dry Eluate

a) Evaporate fraction(s) to complete dryness under stream of dry air or nitrogen at <40°C.

6. Derivatize

- a) Add 50 µL ethyl acetate, vortex mix.
- b) Add 50 µL BSTFA (with 1% TMCS).
- or
- b) Add 50 µL MTBSTFA (with 1% TBMCS).
- c) Mix/vortex.
- d) React 20 minutes at 70°C.
- e) Remove from heat source to cool.

Note: Do not evaporate BSTFA.

7. Quantitate

- a) Inject 2 µL onto gas chromatograph.
- b) For MSD monitor the following ions:







Derivatization Procedure

Derivatizing Agent	THC {T-005}**	THC-OH {H-041}**	THC-COOH {T-006}**
	(D3 THC) {T-003}**	(D3 THC-OH) {H-027}**	(D9 THC-COOH) {T-007}**
BSTFA	371, 343, 386	371, 459, 474	371, 473, 488
	(374, 346, 389)	(374, 462, 477)	(380, 479, 497)
MTBSTFA	371, 428, 345	413, 369, 501	413, 515, 572
	(374, 431, 348)	(416, 372, 504)	(422, 524, 581)

^{*} Suggested internal standard for GC/MS: D9-Carboxy-delta 9-THC, D3-Hyroxy- delta 9-THC, D3-delta 9-THC

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^{**} lons common to deuterated delta-9 THC and non-deuterated compounds.