



THC AND THC-COOH FROM ORAL FLUID BY LC-MS/MS OR GC-MS STYRE-SCREEN[®] POLYMERIC QAX EXTRACTION COLUMN

Part #

SSQAX056 – Styre Screen[®] QAX 50 mg, 6 mL Tube

SBSTFA-1-1 – SELECTRA-SIL[®] BSTFA w/ 1% TMCS

SMSTFA1-1 – SELECTRA-SIL[®] MSTFA w/ 1% TMCS

SLDA100ID21-5UM – Selectra[®] DA HPLC Column, 100 x 2.1 mm, 5 μ m

1. PREPARE SAMPLE:

To 1 mL of oral fluid specimen (diluted in Quantisal buffer), add appropriate internal standards

Mix/vortex for 30 seconds

3. APPLY SAMPLE:

Load at 1 to 2 mL/minute.

Dry column (2 minutes at full vacuum or pressure)

4. WASH COLUMN:

1 x 750 μ L of 85:15:1 D.I. H₂O: ACN: NH₄OH

Dry Column at full vacuum or pressure for 10 minutes

5. ELUTE ANALYTES:

1 x 750 μ L Hexane/: Ethyl Acetate/ Glacial Acetic Acid (90:10:2)

Collect eluate at 1 to 2 mL/minute

NOTE: Before proceeding, insure there are no water droplets at the bottom of the collection tube. This may increase drying time and decrease BSTFA derivatizing efficiency

6. DRY ELUATE:

Evaporate to dryness at < 40 °C.

7. RECONSTITUTE / DERIVATIZE:

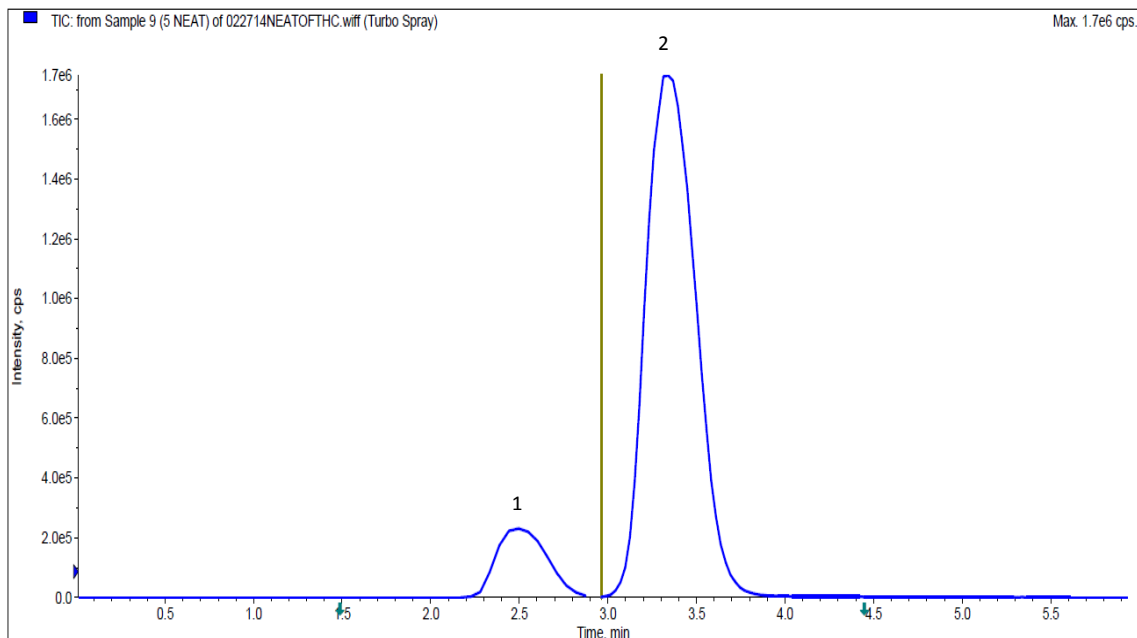
- **LC-MS/MS:** Reconstitute sample in 100 μ L of mobile phase
Inject 20 μ L.
- **GC-MS:** Dissolve residue in 50 μ L of Ethyl Acetate and
50 μ L BSTFA w/ 1% TMCS
Overlay with N₂ and cap. Mix/vortex
React 30 minutes at 70 °C; Cool and inject 1 μ L

Alternate Derivatization

- Form TMS Derivatives by adding 50 μ L MSTFA w/ 1% TMCS and 50 μ L of Ethyl Acetate;
React 45 minutes at 70 °C

INSTRUMENT CONDITIONS (LC-MS/MS):

CHROMATOGRAM



Analyte	MRM Transitions		Relative Retention Time (min)
	Q1	Q3	
THC-DELTA 9-COOH D ₉	352	254	2.50
1. THC-DELTA 9-COOH	343	299	2.52
DELTA 9-THC D ₃	318	196	3.34
2. DELTA 9-THC	315	193	3.37

PARAMETERS

Mobile Phase A: 0.1% Formic Acid in D.I. H₂O

Mobile Phase B: 0.1% Formic Acid in Methanol

Flow Rate: 0.4 mL/minute

Polarity: Negative MRM (-): 2.995

Positive MRM (+): 3.018

Reconstitute: 100 µL

Injection Volume: 20 µL

LC Column: Selectra® DA HPLC Column 100 x 2.1 mm 5 µm

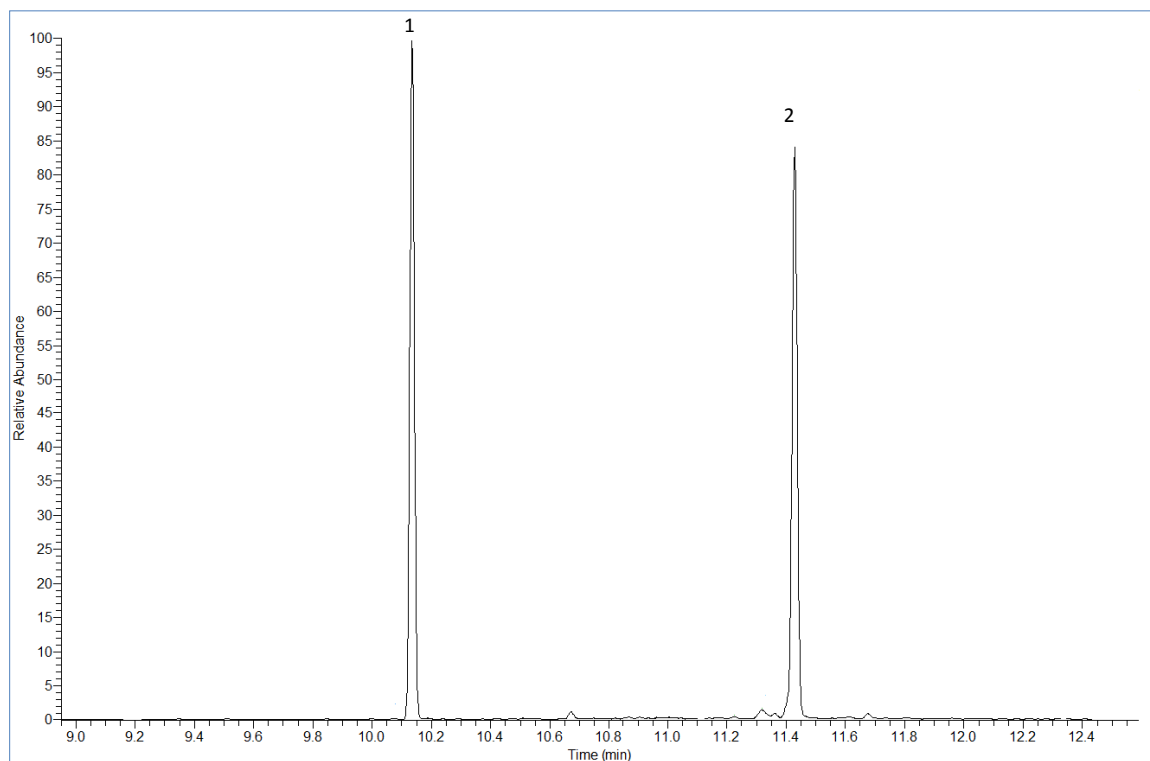
Instrument: API 4000 Qtrap MS/MS with Agilent 1200 Binary Pump SL

Gradient:

Time	%A	%B
0.00	20	80
7.00	STOP	

INSTRUMENT CONDITIONS (GC-MS):

CHROMATOGRAM



MSTFA/BSTFA TMS IONS

Analyte	Quantify Ion	Qualifier Ion 1	Qualifier Ion 2	Relative Retention Time (min)
DELTA 9 THC-D ₃	374	346	389	-
1. DELTA 9 THC	371	343	386	10.14
THC-COOH D ₃	374	476	491	-
2. THC-COOH	371	473	488	11.43

PARAMETERS

GC/MS: Thermo ISQ Trace 1300

GC capillary column: 30 m x 0.25 mm (0.25 µm) TG-1MS

Injector: 1 µL Splitless, 250 °C

Oven temperature program: 70 °C (0.5) to 320 °C (25 °C/ minute): hold (2 minutes)

Carrier gas: Carrier Gas: Helium (1.2 mL/minute)

MSD condition: Aux temperature: 280 °C, MS Source: 350 °C, MS Quad: 150 °C