



















**Worklist: 4739**

<u>LAB_CASE</u>	<u>ITEM</u>	<u>ITEM_TYPE</u>	<u>DESCRIPTION</u>	
C2020-2566	2	UCK	AM 30 Urine THC Screen by LC-QTOF	
C2020-2572	3	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2609	1	UCK	AM 30 Urine THC Screen by LC-QTOF	
C2020-2611	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2615	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2618	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2618	4	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2021-0009	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2021-0023	2	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2021-0037	1	UCK	AM 30 Urine THC Screen by LC-QTOF	
C2021-0047	3	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2021-0048	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2021-0049	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2021-0050	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2021-0056	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2021-0066	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2021-0068	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2021-0070	1	BCK	AM 30 Blood THC Screen by LC-QTOF	

AM# 30: Screening of THC and Metabolites in Blood and Urine by LC-QTOF

Extraction Date: 1/11/21

Analyst: Anne Nord

Plate lot#: 200723

Plate Expiration: 1-23-2021

Mobile phase A: 0.1% formic acid in water

Mobile phase B: 0.1% Formic acid in Methanol

Blank Blood Lot: 20J20793

Column: Phenomenex Phenyl Hexyl (4.6x50mm, 2.6um)

LCMS-QQ ID: 70044

Blank Urine Lot: 10120

Pre-Analytic:

- 1. Check levels of mobile phases and needle wash refill as needed. Ensure waste is not full.
- 2. Ensure correct column is installed and begin mobile phase flow allow to equilibrate ~ 30 minutes.

Analytic:

- 1. Remove standards, plate, controls, and samples from cold storage. Allow to reach room temperature.
- 2. Urine hydrolysis: add 1.5mL urine to blank plate, add 250µl 1N KOH. Shake and incubate at 40 degrees for 15 minutes.
Using a calibrated pipette, add **1000µl blood and urine (if applicable) (calibrated pipette)** into the appropriate wells of analytical (standards) plate. **Pipette ID: K52558G**
- 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 4. Pipette **500µL 0.1% formic acid in water blood sample, 500 µL saturated phosphate buffer in urine** in wells of analytical plate.
- 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 6. Transfer **800µL of blood+acid or urine+acid** mixture to corresponding wells of SLE+ plate.
- 7. Apply positive pressure for approx. 10-15 seconds (or until no liquid remains on top of sorbent).
(Load at 85-100 PSI- Selector to the right) Manifold ID: 66792
- 8. Wait 5 minutes.
- 9. Add **2.25mL MTBE. (Add in 3 increments of 750uL)**
- 10. Wait 5 minutes.
- 11. Apply positive pressure for approx. 15 seconds. **(10-15 PSI- Selector to the left).**
- 12. Add **2.25mL Hexane. (Add in 3 increments of 750uL)**
- 13. Wait 5 minutes.
- 14. Apply positive pressure for approx. 15 seconds. **(10-15 PSI- Selector to the left).**
- 15. Remove plate containing eluate. Place on SPE Dry and evaporate to dryness at approx. 35°C.
SPE Dry ID: 66819
- 16. Reconstitute in **100µL 100% MeOH** and heat seal plate with foil. Place in autosampler and run worklist.

Post-Analytic

- 1. Create batch and process data.
- 2. Make any necessary integration changes, r^2 values ≥ 0.98 for each analyte
- 3. Evaluate samples: RT within +/- 2% or 0.1 min of administrative control, Mass Accuracy of 0 (+/- 10), and/or Mass Abundance Score of 40 or greater.
- 4. Did all QCs pass for each analyte? (if not, describe in comments section)
- 6. Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS: Evaluated THC-COOH only. THC and THC-OH had poor response compared to baseline at lower end of the curve.

Toxicology AM method 27/26 external prep information A

working solution 15 ug/ml in meoh C-THC, THC-OH, 7.5 ug/ml THC

Stock solution 1mg/ml 7.5 ul each THC, 100 ug/ml 150 ul C-THC, 150 ul THC-OH in 9692.5 ul meOH

Ppd 8/26/20 Exp: 7/1/21 lot 82620 by AMN

Drug	lot	expiration
C-THC	FE01061702	3/1/2022
THC-OH	FE07221601	7/1/2021
THC	FE01041701	3/1/2022

AM 27/26 blood control 100 ul working solution lot () in 9900 ul blood lot ()

		Concentration 7.5 ng/ml THC, 15 ng/ml C-THC, THC-OH	
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AM 27/26 urine control 400 ul working solution lot (82620) in 9600 ul urine

out of use

ppd 8/26/20 Exp 7/1/21 neg urine lot 73020	lot u82620	Concentration 30 ng/ml THC, and 60 ng/ml C-THC, THC-OH	by amn	10/4/2020
ppd 10/5/20 Exp 7/1/21 neg urine lot 10120	lot 10520	Concentration 30 ng/ml THC, and 60 ng/ml C-THC, THC-OH	by amn	1/12/2021
ppd 1/13/21 Exp 7/1/21 neg urine lot 10120	lot 11321	Concentration 30 ng/ml THC, and 60 ng/ml C-THC, THC-OH	by amn	

	1	2	3	4	5	6
a	cal 1ng	neg blood	21-0047-3	negative urine		QC 1
b	cal 3 ng	20-2615-1	21-0048-1 1-11-21	urine control		cal 100 ng
c	cal 5 ng	20-2611-1	21-0049-1	20-2566-2		cal 50 ng
d	cal 10ng	20-2615-1	21-0050-1	20-2609-1		cal 25 ng
e	25	20-2618-1	21-0056-1	21-0037-1		cal 10ng
f	50	20-2618-4	21-0066-1	21-0048-1		cal 5 ng
g	100	21-0009-1	21-0068-1			cal 3 ng
h	QC 1	21-0023-2	21-0070-1			cal 1ng

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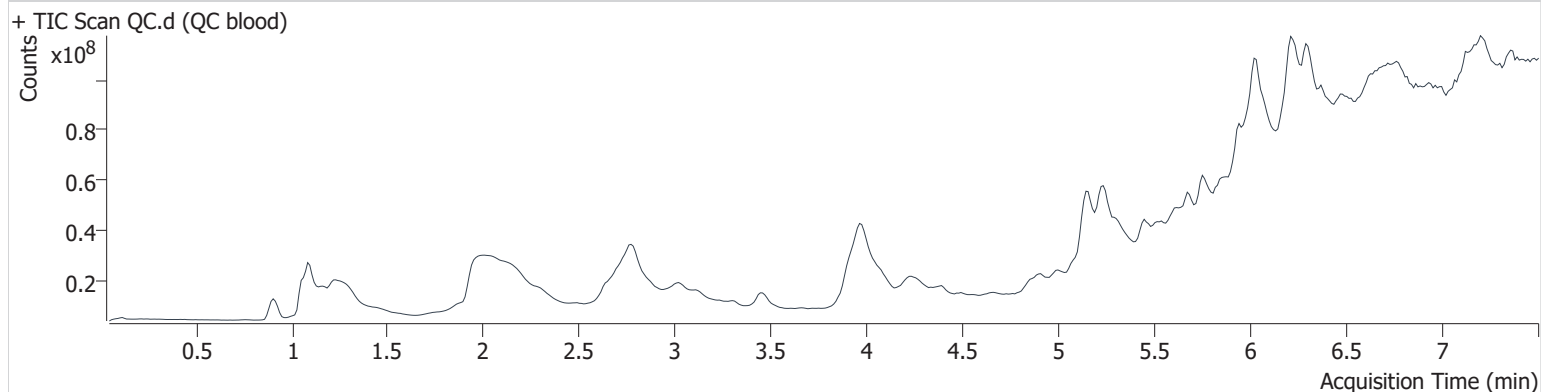
AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	QC.d
Type	QC	Sample	QC blood
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-H1	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 3:10:53 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.713	665365	1.30	74.6	2960264	15.8764 ng/ml

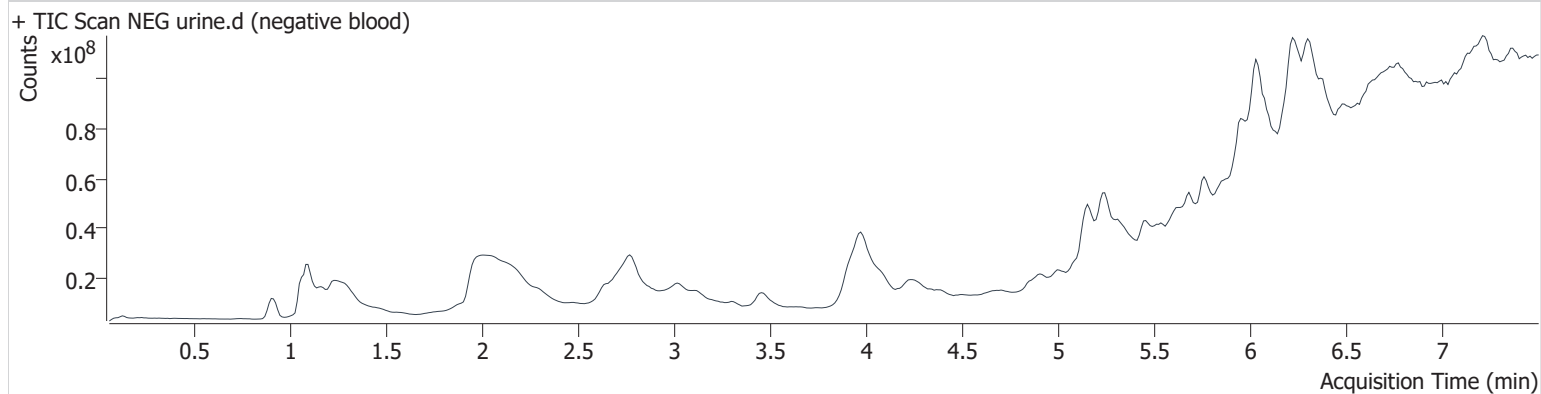
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AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	NEG urine.d
Type	Sample	Sample	negative blood
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-A2	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 3:20:31 PM		
Sample Info.			

Sample Chromatogram



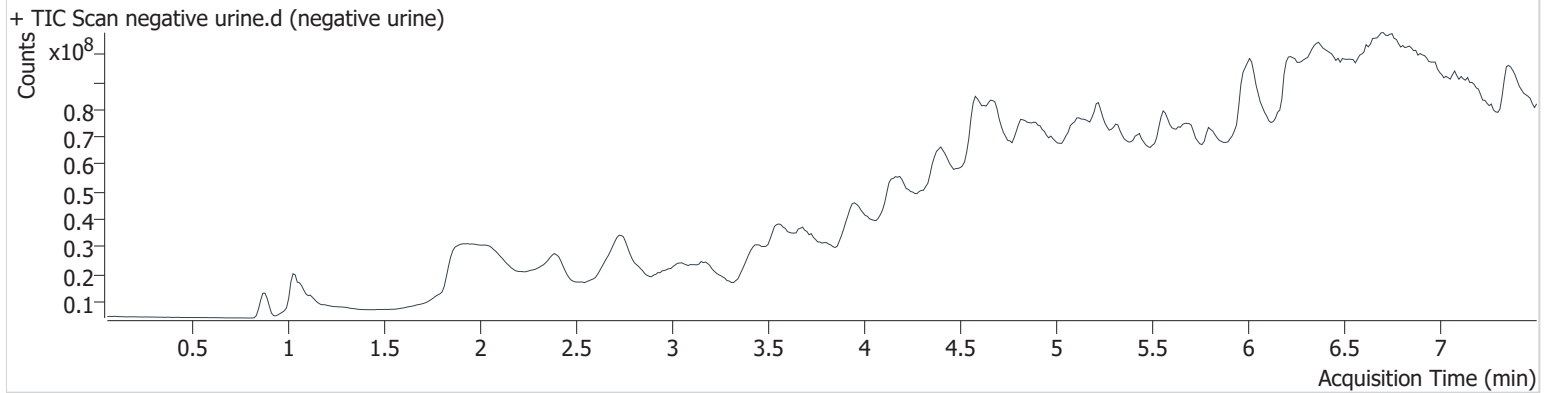
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AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	negative urine.d
Type	Sample	Sample	negative urine
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-A4	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 5:58:21 PM		
Sample Info.			

Sample Chromatogram

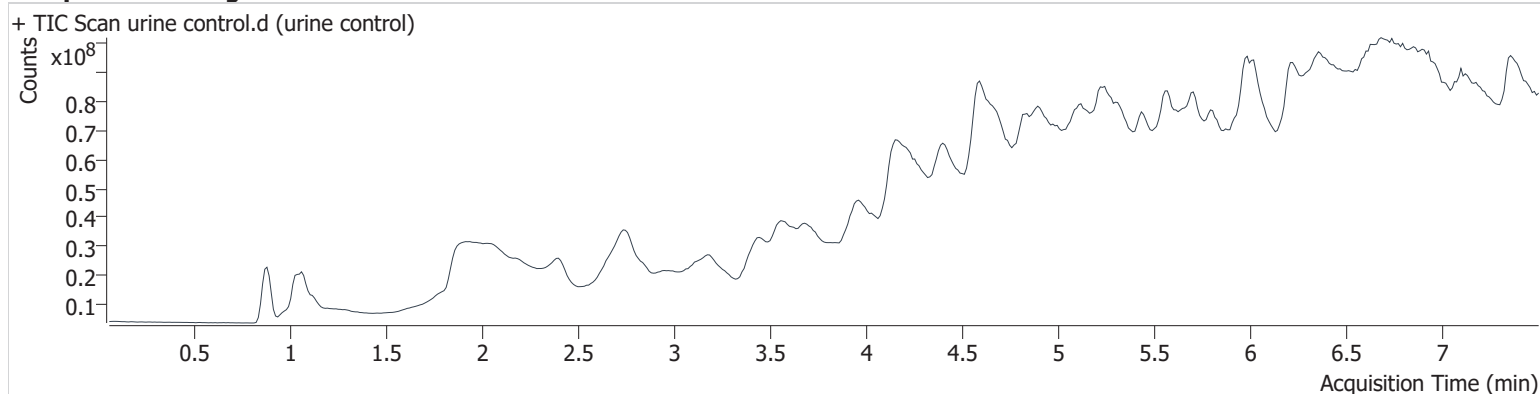


AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	urine control.d
Type	Sample	Sample	urine control
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-B4	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 6:07:57 PM		

Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.699	558553	1.90	76.7	3022250	12.9415 ng/ml

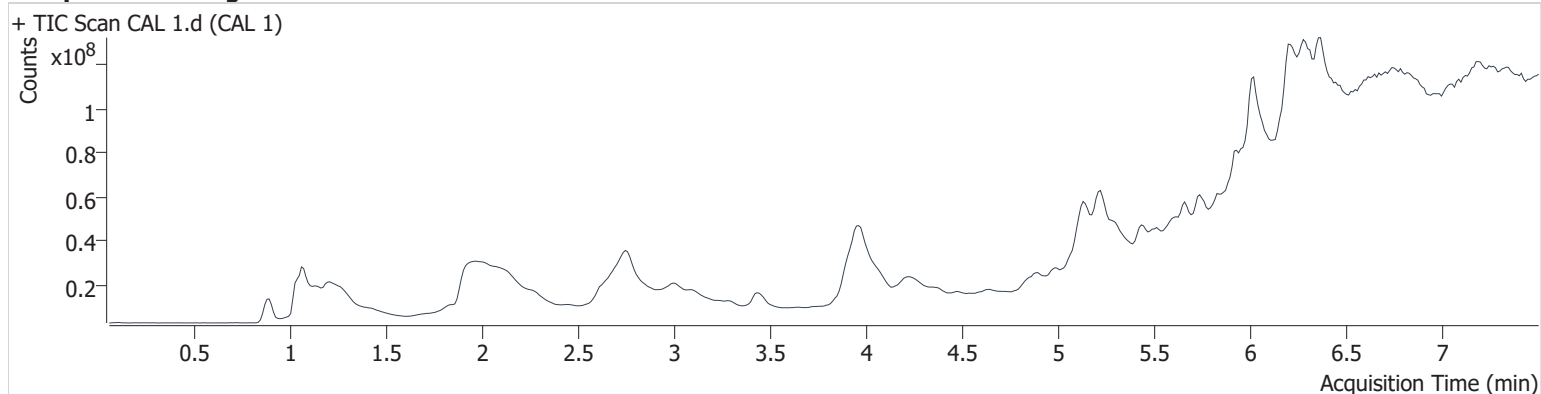
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AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	CAL 1.d
Type	Cal	Sample	CAL 1
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-A1	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 2:03:26 PM		

Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.687	144468	-0.73	71.6	1547496	6.2229 ng/ml

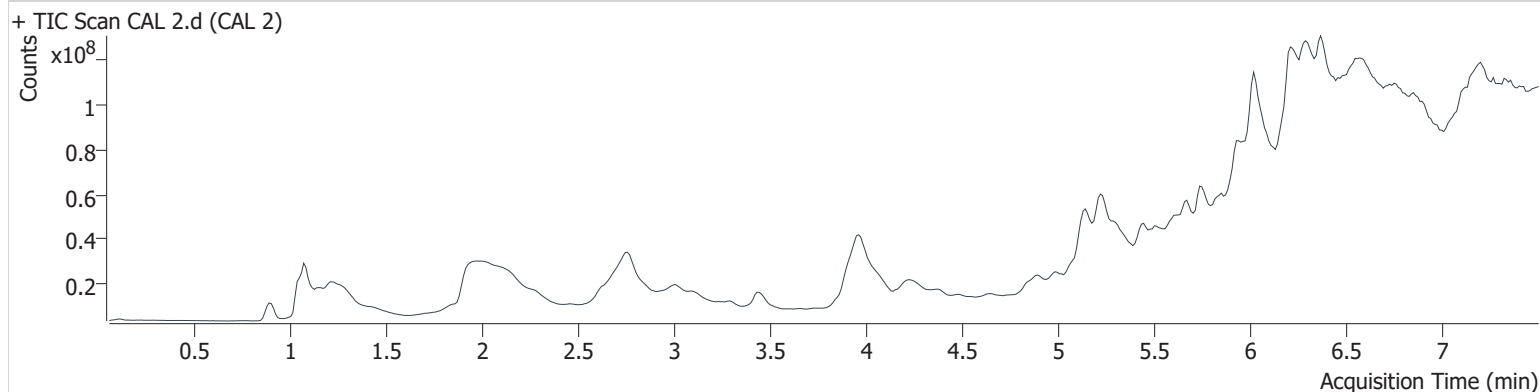
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AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	CAL 2.d
Type	Cal	Sample	CAL 2
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-B1	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 2:13:12 PM		

Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.695	227799	0.34	13.3	1488681	10.6059 ng/ml

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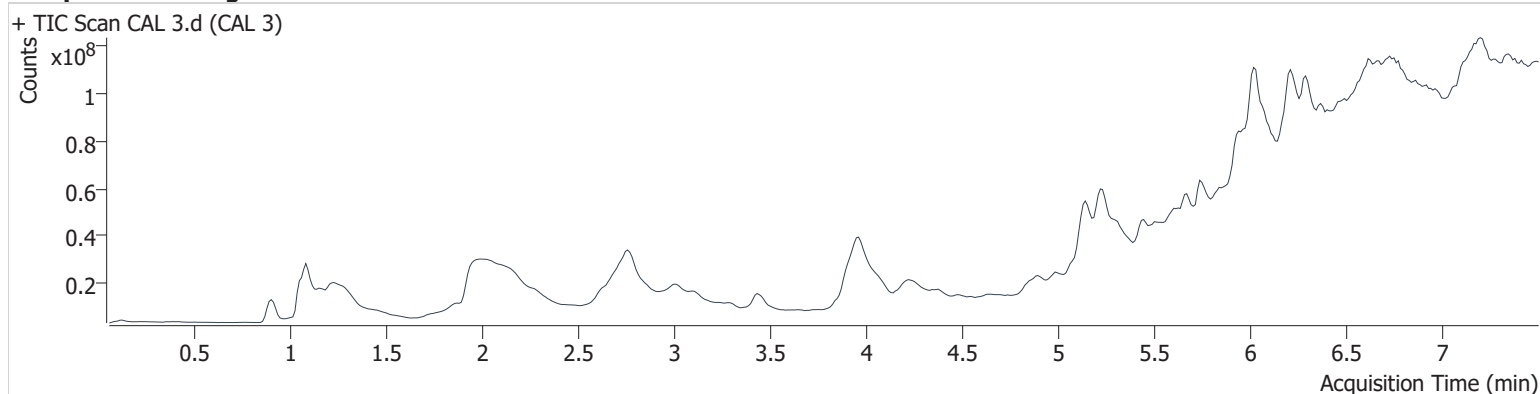
AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	CAL 3.d
Type	Cal	Sample	CAL 3
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-C1	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 2:22:45 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.702	625953	2.67	88.0	2559014	17.3340 ng/ml

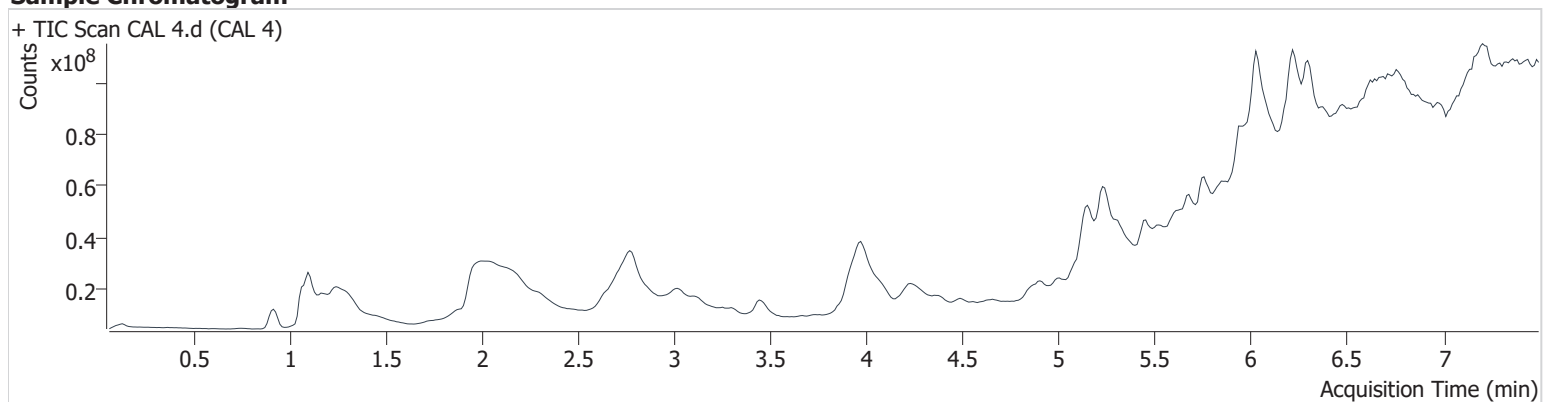
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AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	CAL 4.d
Type	Cal	Sample	CAL 4
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-D1	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 2:32:22 PM		

Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.706	1942730	3.36	97.6	3349677	41.9705 ng/ml

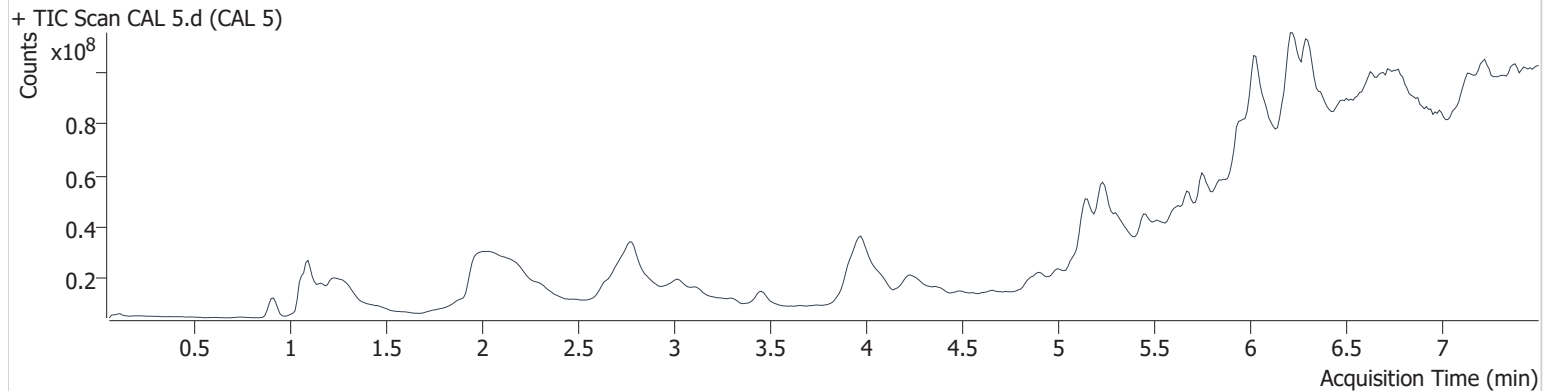
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AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	CAL 5.d
Type	Cal	Sample	CAL 5
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-E1	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 2:42:00 PM		

Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.712	2892728	3.29	99.1	2870637	73.3912 ng/ml

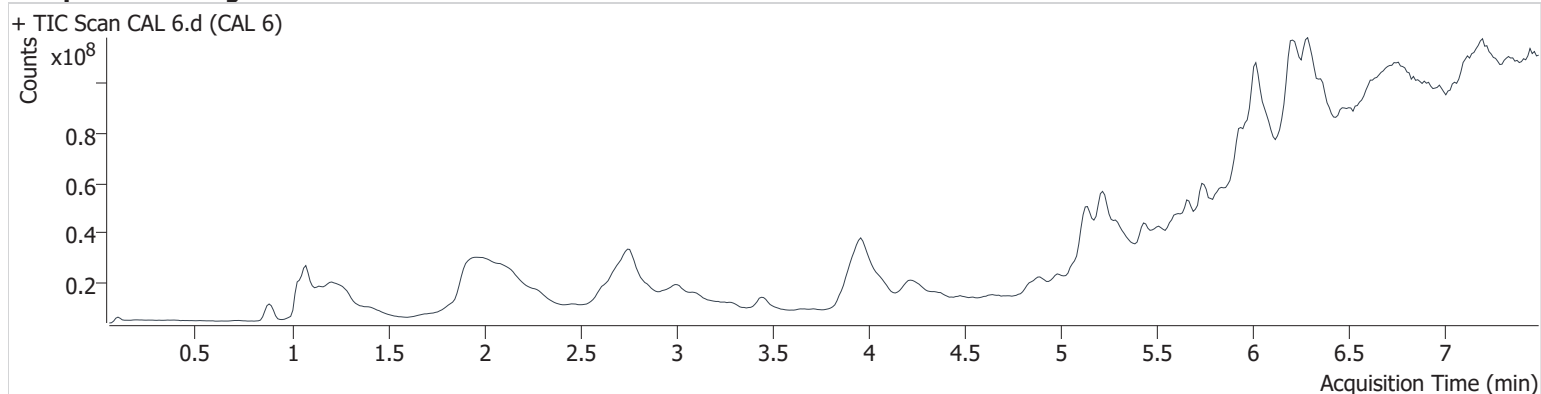
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AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	CAL 6.d
Type	Cal	Sample	CAL 6
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-F1	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 2:51:37 PM		

Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.704	3882809	-1.02	99.6	2991593	94.7104 ng/ml

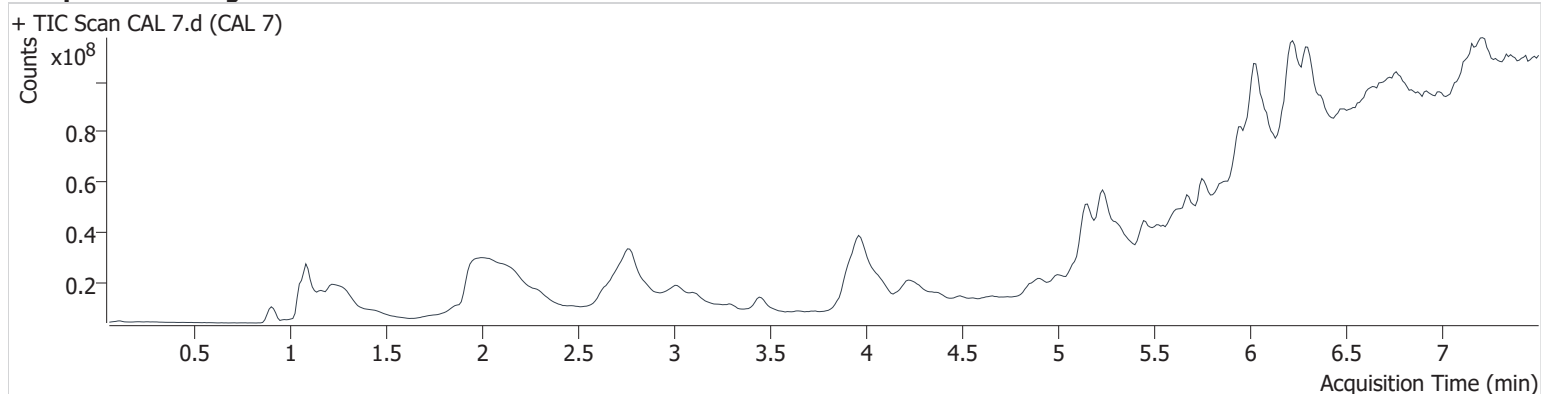
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AM #30 Cannabinoids

Batch results D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/13/2021 2:44:46 PM

Instrument	70044	Data File	CAL 7.d
Type	Cal	Sample	CAL 7
Acq. Method	THC Screen 1122.m	Operator	Anne Nord
Sample Position	P2-G1	Comment	
Injection Volume	10		
Acq. Date-Time	1/11/2021 3:01:15 PM		

Sample Chromatogram



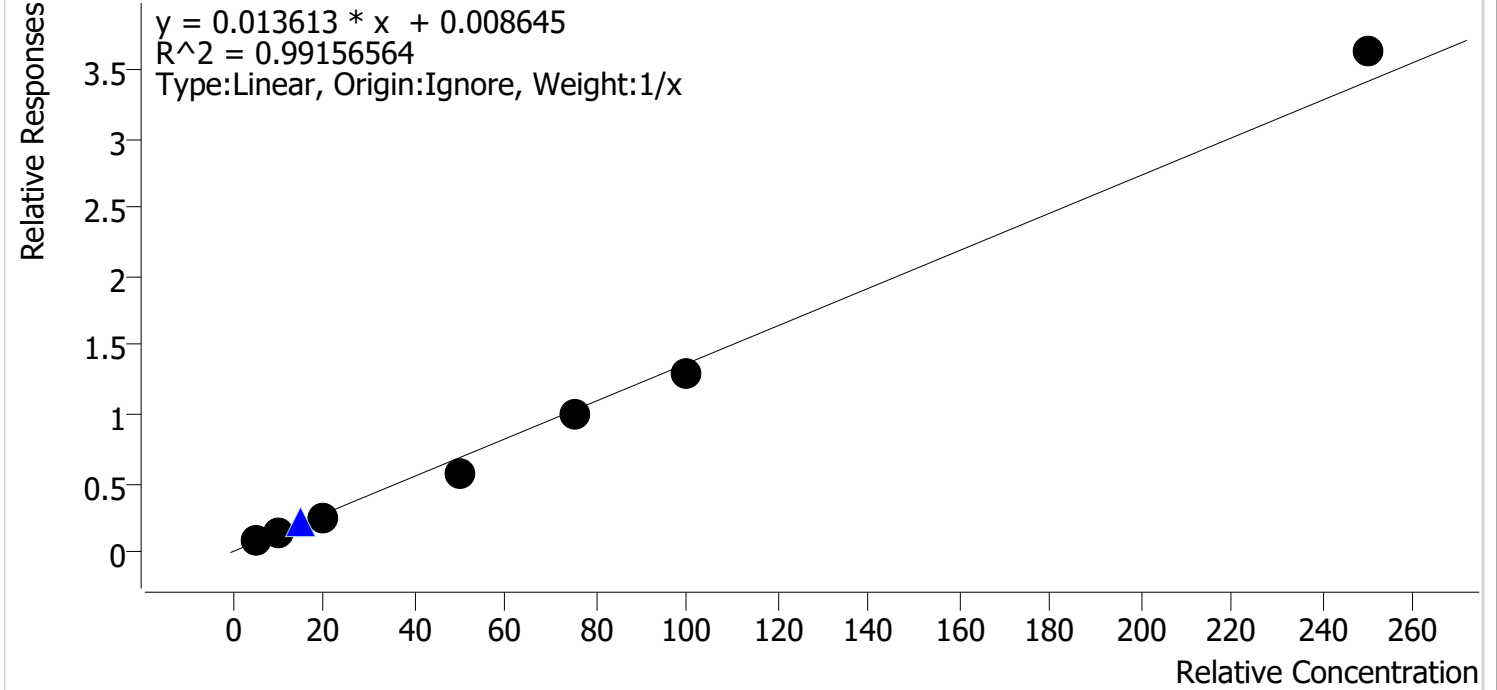
Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.708	10495770	2.53	99.7	2894255	265.7650 ng/ml

Compound Calibration Report



Batch results	D:\MassHunter\Data\2021\am 30\011121\QuantResults\cann screen.batch.bin		
Last Cal. Update	1/13/2021 2:44 PM		
Analyst Name	ISP\datastor		
Analyte	THC-COOH	Internal Standard	THC-COOH-D9

THC-COOH - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 1 QCs



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
CAL 1	1	✓	5.0	6.2	124.5
CAL 2	2	✓	10.0	10.6	106.1
CAL 3	3	✓	20.0	17.3	86.7
CAL 4	4	✓	50.0	42.0	83.9
CAL 5	5	✓	75.0	73.4	97.9
CAL 6	6	✓	100.0	94.7	94.7
CAL 7	7	✓	250.0	265.8	106.3