


















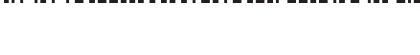


**Worklist: 4584**

REVIEWED

By Brittany Wylie at 11:20 am, Nov 03, 2020

<u>LAB_CASE</u>	<u>ITEM</u>	<u>ITEM_TYPE</u>	<u>DESCRIPTION</u>	
C2020-1979	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-1992	1	AVK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2018	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2069	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2088	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2089	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2094	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2095	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
C2020-2145	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-2822	2	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-2927	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-2928	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-2929	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-2930	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-2931	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-2931	2	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-3017	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-3074	1	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-3074	2	BCK	AM 30 Blood THC Screen by LC-QTOF	
P2020-3075	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: 10/30/20

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:** 20G20792

**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  $\geq 0.98$  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.

~~A~~

	1	2	3	4	5	6
a	cal 100 ng	neg blood	c2095-1	p2931-2		QC 1
b	cal 50 ng	c1979-1	c2145-1	p3017-1		cal 100 ng
c	cal 25 ng	c1992-1	p2822-2	p3074-1		cal 50 ng
d	cal 10ng	c2018-1	p2927-1	p3074-2		cal 25 ng
e	cal 5 ng	c2069-1	p2928-1	p3075-1		cal 10ng
f	cal 3 ng	c2088-1	p2929-1	c2094-1		cal 5 ng
g	cal 1ng	c2089-1	p2930-1			cal 3 ng
h	QC 1		p2931-1			cal 1ng

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QA

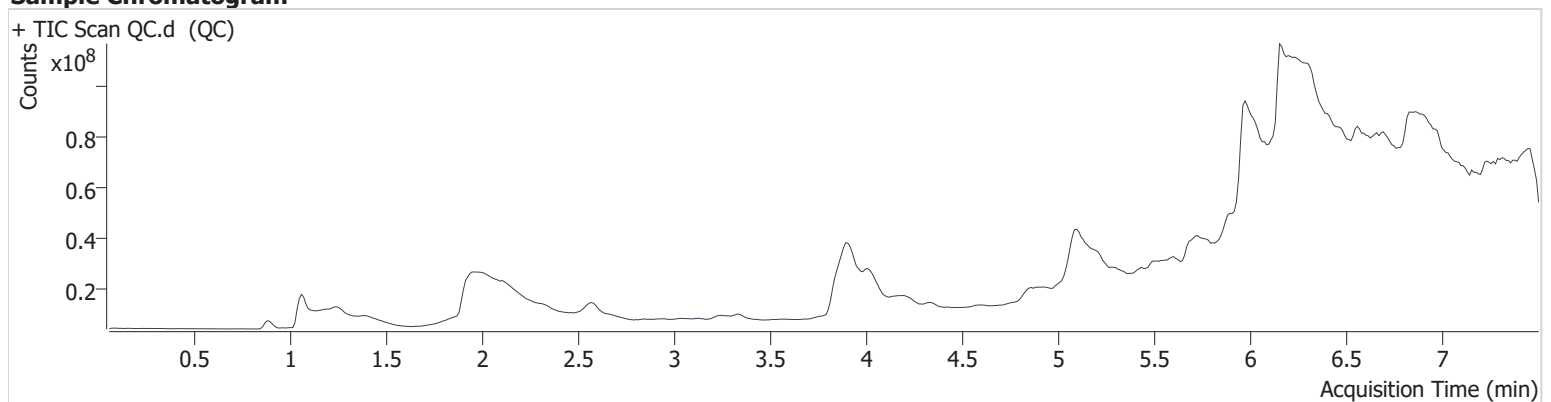
# AM #30 Cannabinoids

**Batch results** D:\MassHunter\Data\2020\am 30\103020\QuantResults\cann.batch.bin  
**Calibration Last Update** 11/3/2020 10:26:24 AM

<b>Instrument</b>	69679	<b>Data File</b>	QC.d
<b>Type</b>	QC	<b>Sample</b>	QC
<b>Acq. Method</b>	THC Screen 1122.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P2-H1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	10/30/2020 2:07:41 PM		

**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.628	276685	-3.12	67.3	1430316	18.0891 ng/ml

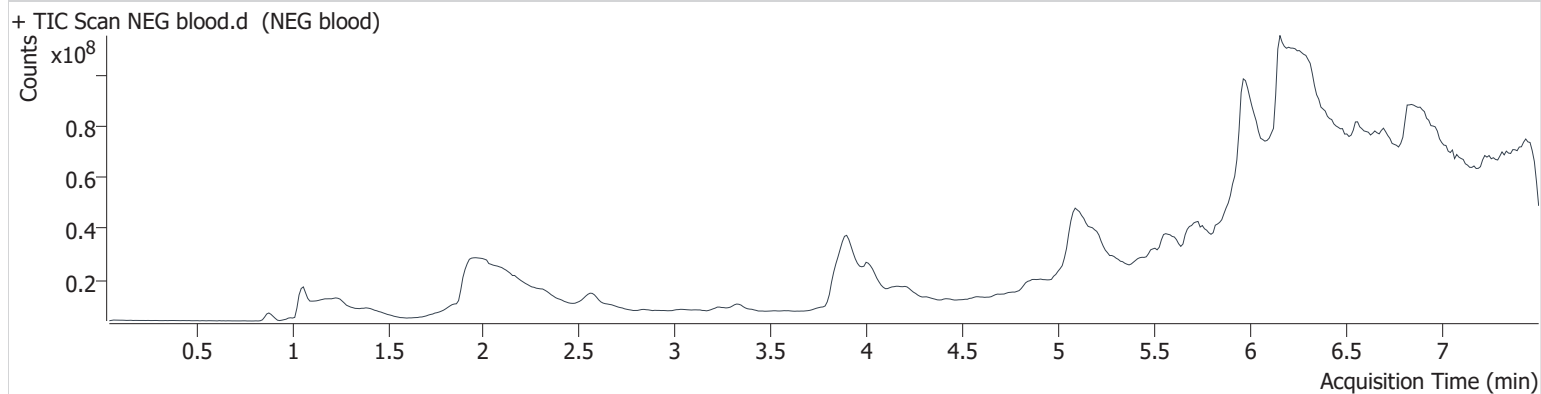
GA

# AM #30 Cannabinoids

**Batch results** D:\MassHunter\Data\2020\am 30\103020\QuantResults\cann.batch.bin  
**Calibration Last Update** 11/3/2020 10:26:24 AM

<b>Instrument</b>	69679	<b>Data File</b>	NEG blood.d
<b>Type</b>	Sample	<b>Sample</b>	NEG blood
<b>Acq. Method</b>	THC Screen 1122.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P2-A2	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	10/30/2020 2:17:15 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



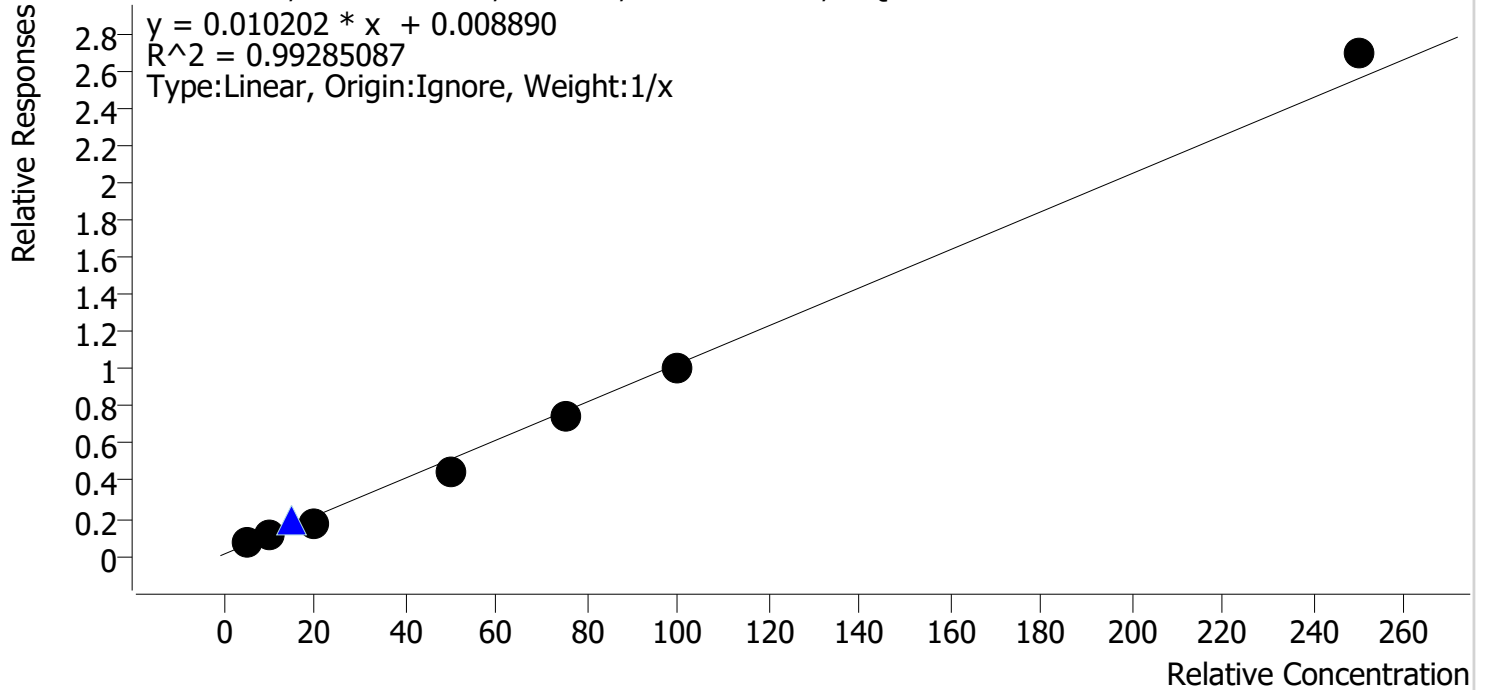
# Compound Calibration Report



**Batch results** D:\MassHunter\Data\2020\am 30\103020\QuantResults\cann.batch.bin  
**Last Cal. Update** 11/3/2020 10:26 AM  
**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	6.5	129.6
CAL 2	2	✓	10.0	10.2	102.0
CAL 3	3	✓	20.0	16.3	81.6
CAL 4	4	✓	50.0	43.4	86.8
CAL 5	5	✓	75.0	72.4	96.6
CAL 6	6	✓	100.0	98.1	98.1
CAL 7	7	✓	250.0	263.0	105.2

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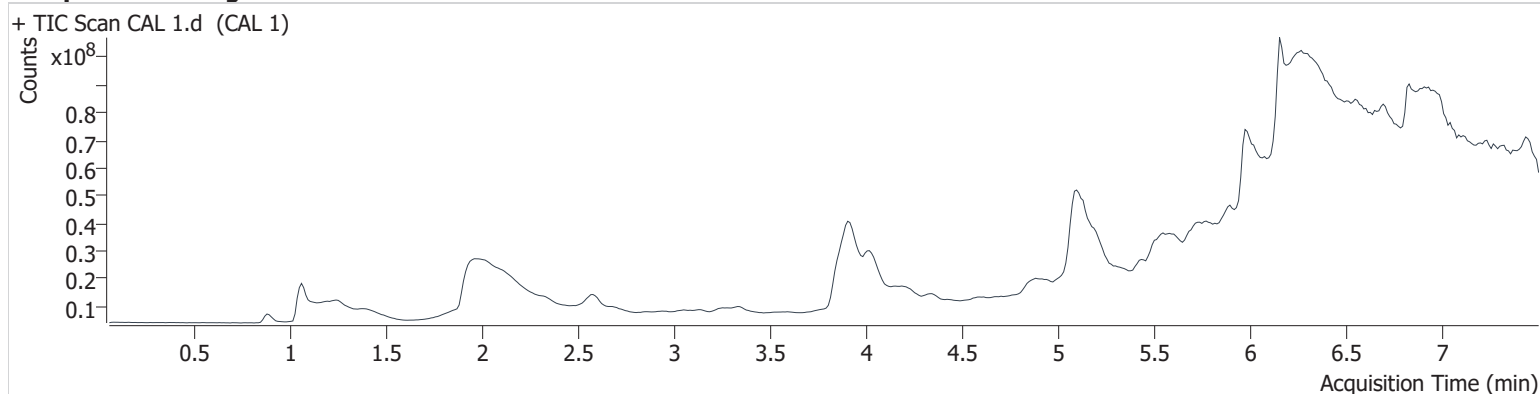
# AM #30 Cannabinoids

**Batch results** D:\MassHunter\Data\2020\am 30\103020\QuantResults\cann.batch.bin  
**Calibration Last Update** 11/3/2020 10:26:24 AM

<b>Instrument</b>	69679	<b>Data File</b>	CAL 1.d
<b>Type</b>	Cal	<b>Sample</b>	CAL 1
<b>Acq. Method</b>	THC Screen 1122.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P2-A1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	10/30/2020 1:00:27 PM		

**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.641	90503	-3.31	83.0	1206372	6.4818 ng/ml

GA

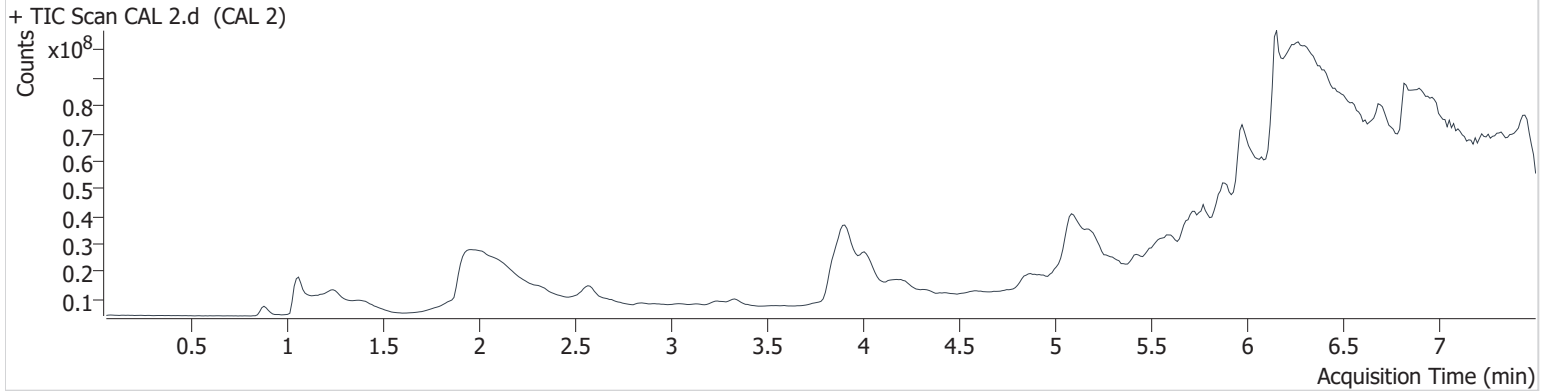
# AM #30 Cannabinoids

**Batch results** D:\MassHunter\Data\2020\am 30\103020\QuantResults\cann.batch.bin  
**Calibration Last Update** 11/3/2020 10:26:24 AM

<b>Instrument</b>	69679	<b>Data File</b>	CAL 2.d
<b>Type</b>	Cal	<b>Sample</b>	CAL 2
<b>Acq. Method</b>	THC Screen 1122.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P2-B1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	10/30/2020 1:10:11 PM		

**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.634	184334	-2.03	51.7	1632233	10.1979 ng/ml



GA

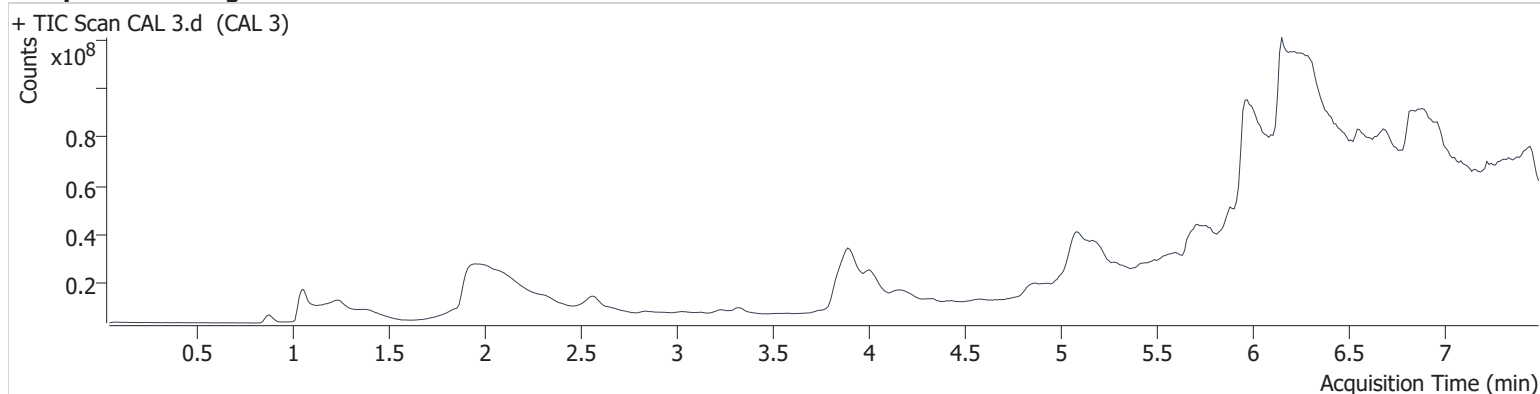
# AM #30 Cannabinoids

**Batch results** D:\MassHunter\Data\2020\am 30\103020\QuantResults\cann.batch.bin  
**Calibration Last Update** 11/3/2020 10:26:24 AM

<b>Instrument</b>	69679	<b>Data File</b>	CAL 3.d
<b>Type</b>	Cal	<b>Sample</b>	CAL 3
<b>Acq. Method</b>	THC Screen 1122.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P2-C1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	10/30/2020 1:19:47 PM		

**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.624	303469	0.55	64.9	1729473	16.3273 ng/ml

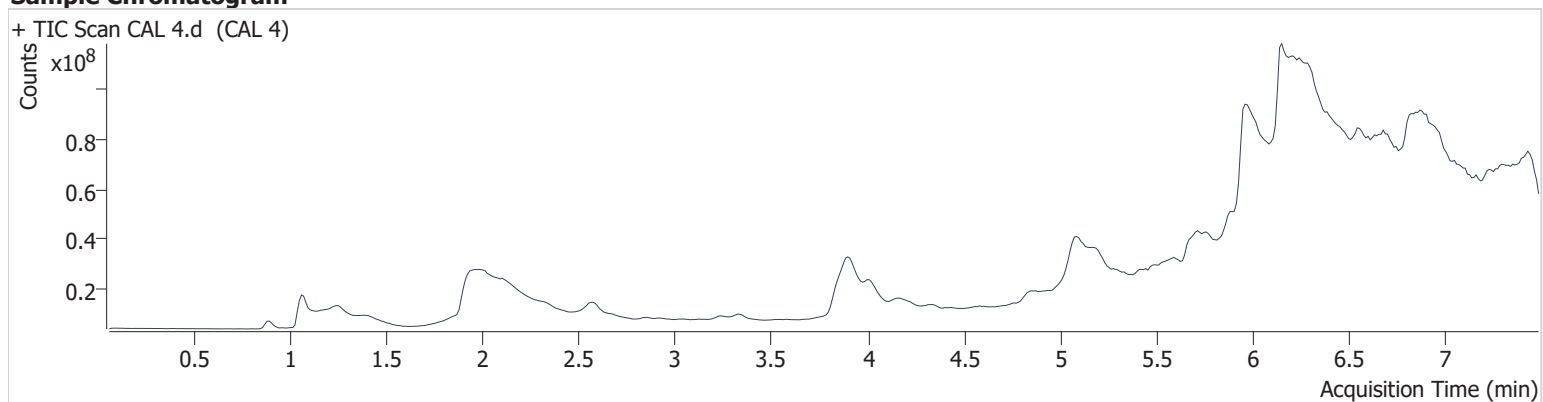
GA

# AM #30 Cannabinoids

**Batch results** D:\MassHunter\Data\2020\am 30\103020\QuantResults\cann.batch.bin  
**Calibration Last Update** 11/3/2020 10:26:24 AM

<b>Instrument</b>	69679	<b>Data File</b>	CAL 4.d
<b>Type</b>	Cal	<b>Sample</b>	CAL 4
<b>Acq. Method</b>	THC Screen 1122.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P2-D1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	10/30/2020 1:29:20 PM		

## Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.626	765610	3.53	95.1	1694374	43.4174 ng/ml

OA

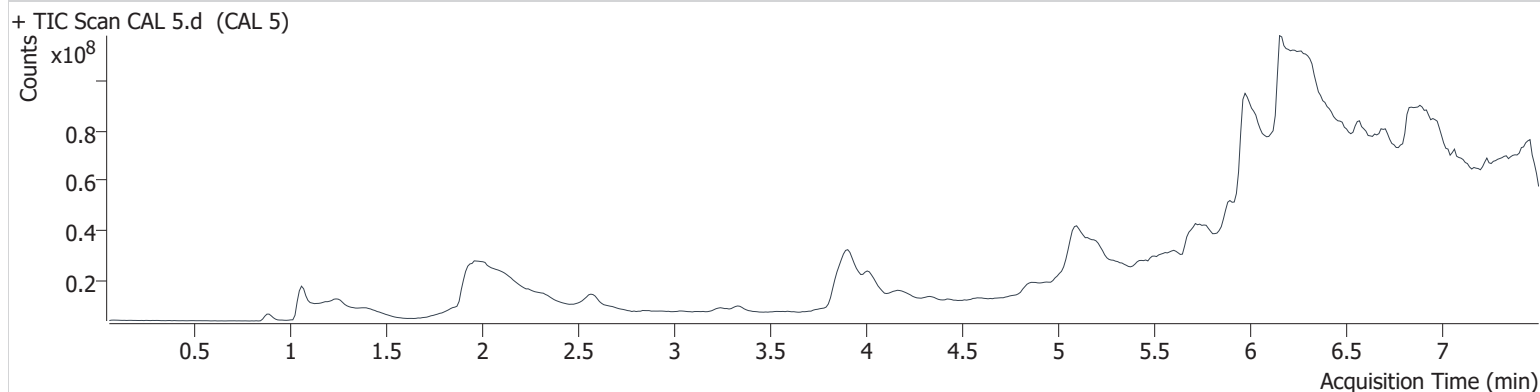
# AM #30 Cannabinoids

**Batch results** D:\MassHunter\Data\2020\am 30\103020\QuantResults\cann.batch.bin  
**Calibration Last Update** 11/3/2020 10:26:24 AM

<b>Instrument</b>	69679	<b>Data File</b>	CAL 5.d
<b>Type</b>	Cal	<b>Sample</b>	CAL 5
<b>Acq. Method</b>	THC Screen 1122.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P2-E1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	10/30/2020 1:38:54 PM		

**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.628	1302566	2.60	99.0	1741438	72.4427 ng/ml

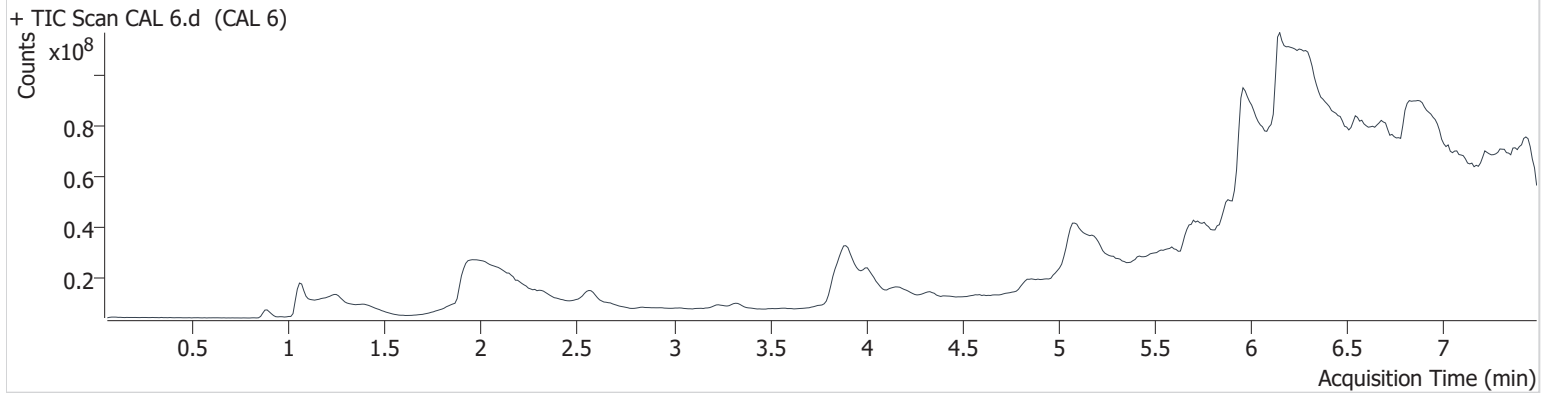
# AM #30 Cannabinoids

**Batch results** D:\MassHunter\Data\2020\am 30\103020\QuantResults\cann.batch.bin  
**Calibration Last Update** 11/3/2020 10:26:24 AM

<b>Instrument</b>	69679	<b>Data File</b>	CAL 6.d
<b>Type</b>	Cal	<b>Sample</b>	CAL 6
<b>Acq. Method</b>	THC Screen 1122.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P2-F1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	10/30/2020 1:48:31 PM		

**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.626	1686289	2.81	99.1	1669713	98.1174 ng/ml

GA

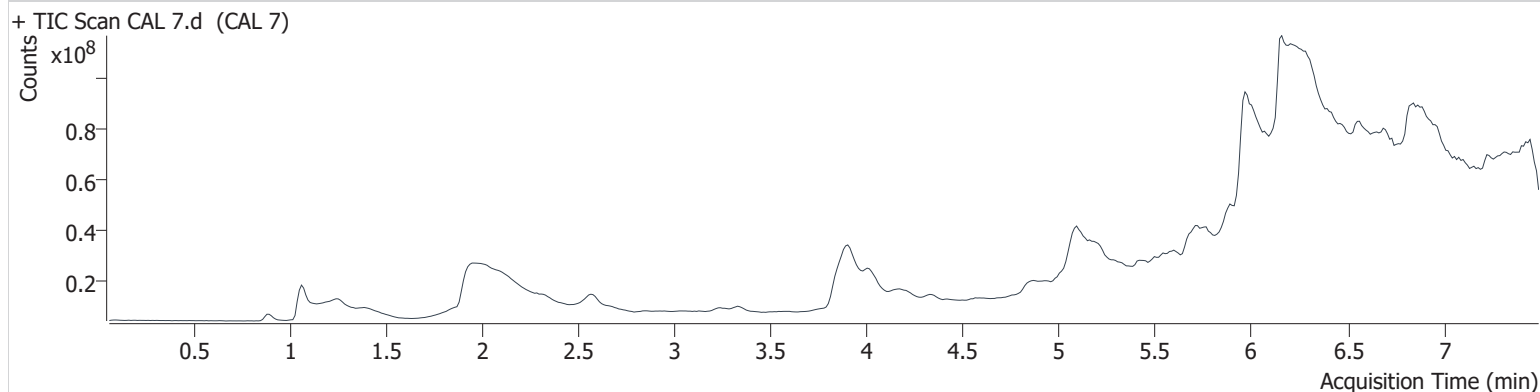
# AM #30 Cannabinoids

**Batch results** D:\MassHunter\Data\2020\am 30\103020\QuantResults\cann.batch.bin  
**Calibration Last Update** 11/3/2020 10:26:24 AM

<b>Instrument</b>	69679	<b>Data File</b>	CAL 7.d
<b>Type</b>	Cal	<b>Sample</b>	CAL 7
<b>Acq. Method</b>	THC Screen 1122.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P2-G1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	10/30/2020 1:58:04 PM		

**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	Mass Accuracy	Mass Abundance Score	ISTD Resp.	Final Conc.
THC-COOH	6.627	4431353	2.83	99.0	1645943	263.0154 ng/ml