

1/13/2022

**Worklist: 5512****REVIEWED**

By Britany Wylie at 3:13 pm, Jan 13, 2022

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>	
C2021-2770	3	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2021-2790		UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2021-2793		BCK	AM 27 Blood THC Quant by LC-QQQ	
C2022-0006		BCK	AM 27 Blood THC Quant by LC-QQQ	
C2022-0012		BCK	AM 27 Blood THC Quant by LC-QQQ	
C2022-0032		UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2022-0045		BCK	AM 27 Blood THC Quant by LC-QQQ	

# AM# 27: Quantitation of THC and Metabolites in Blood and Urine by LC-MS/MS

Extraction Date 1/12/22  
Plate lot#: 211018

Analyst: Anne Nord  
Plate re-test: 4/18/22

**Mobile phase A:** 0.1% Formic Acid in LCMS Water

MTBE

LCMS Methanol

**Mobile phase B:** 0.1% Formic acid in Acetonitrile

Hexane

**Blank Blood Lot:** 21D52496 **Urine Blank:** 83121 **Column:** UCT Selectra DA 100 x 2.1mm 3um

**LCMS-QQQ ID:** 69679

## 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.

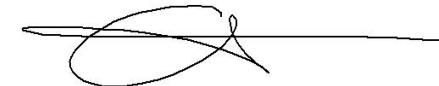
## Analytic:

- 1. Remove standards, plate, controls, and samples from cold storage. Allow to reach room temperature.
- 2. Urine hydrolysis: add 1.5 ml urine to blank plate, add 250 ul 1N KOH mix and incubate at 40 degrees for 15 minutes.  
Pipette **1000 $\mu$ L blood (calibrated pipette)** Pipette ID: k52558g in wells of analytical (standards) plate.
- 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 4. Pipette **500 $\mu$ L 0.1% formic acid in water** **blood sample**, **500 ul saturated phosphate buffer in urine** in wells of analytical plate.
- 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 6. Transfer **800 $\mu$ L 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 750 $\mu$ L)**
- 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 750 $\mu$ L)**
- 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 **100 $\mu$ L 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, Curve weighting of Linear 1/x with  $r^2$  values  $\geq 0.98$  for each analyte
- 3. RT +/- 3% or 0.100 min, whichever is greater, +/- 20% Accuracy for greater than (+/- 30% for 10ng/ml or less).  
Ion ratios must be within +/- 20% of the averaged calibrators
- 4. Case sample response for THC 1ng/ml, OH-THC 3ng/mL (quantitative blood), Carboxy-THC: 5 ng/mL (qualitative only) will be reported. Samples with a THC or OH-THC response over 50 ng/mL will be reported out as greater than 50 ng/mL.
- 5. Did all QC's pass for each analyte? (if not is it describe in comments section)
- 6. Enter QC's into control charting.
- 7. Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS: *THC-OH not evaluated in this run, ratios indicate an interferant.*



	1	2	3	4	5	6
a	cal 1	Internal control (urine)	c2021-2790-1			
b	cal 2	negative blood	c2022-0032-1			
c	cal 3	c2021-2793-1				
d	cal 4	c2022-0006-1				
e	Cal 5	c2022-0012-1				
f	cal 6	c2022-0045-1				
g	cal 7	negative urine				
h	Internal control (blood)	c2021-2770-3				



Added to central file on 3/3/2022.



## IDAHO STATE POLICE

### MEMORANDUM

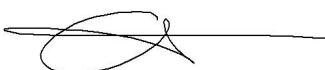
DATE: 3/3/2022

TO: Toxicology Discipline/ Jason Crowe

FROM: Celena Shrum- Toxicology Discipline lead

SUBJECT: Use of internal control in lieu of external control

Toxicology Analytical Methods #25, 26, and 27 specify that if a run contains urine samples, a negative control and **external** urine control must also be included in the run. The purpose of this control is to demonstrate that the extraction worked as intended and to ensure that the results and concentrations obtained are accurate. It was decided in October 2021 that extra QC's would be included on the analytical plates so that they could be used as an internal control for runs with urine cases instead of continuing with including an external control. An internal control serves the same purpose as an external control but is prepared and placed on the analytical plate rather than being prepared in-house and placed on the plate at the time of testing. Utilizing internal controls versus external increases the efficacy of the controls used by ensuring consistent spiking and preparation, eliminating evaporation of compounds, etc. There is no quality issue with any of the cases, since an additional urine control was used that served the same purpose as the external control, but it was a violation of the wording specified in the method.



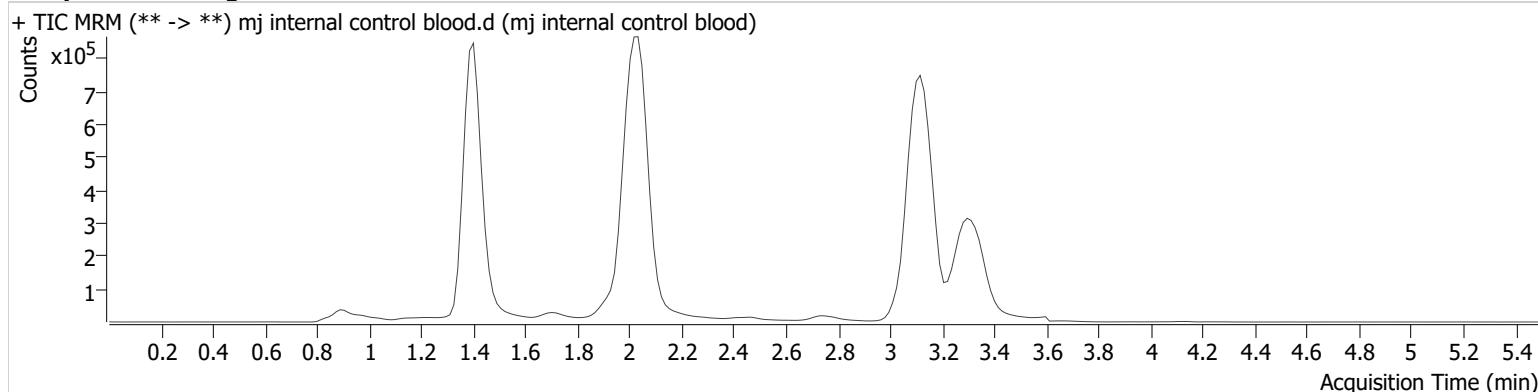
# AM #27 Cannabinoids

**Batch results** D:\MassHunter\Data\2022\am 27-28\011222\QuantResults\cann.batch.bin  
**Calibration Last Update** 1/12/2022 3:20:10 PM

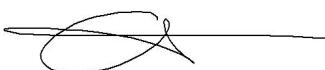
<b>Instrument</b>	69679	<b>Data File</b>	mj internal control blood.d
<b>Type</b>	QC	<b>Sample</b>	mj internal control blood
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-H1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 12:35:21 PM		

**Sample Info.**

**Sample Chromatogram**



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	46813	$\infty$	1191.8	$\infty$	2437701	5.355 ng/ml
THC-COOH	1.416	167772	466.5	34.0	238.3	751463	16.170 ng/ml
THC	3.137	418729	$\infty$	24.6	50842 68000 1341.8	3564313	4.656 ng/ml

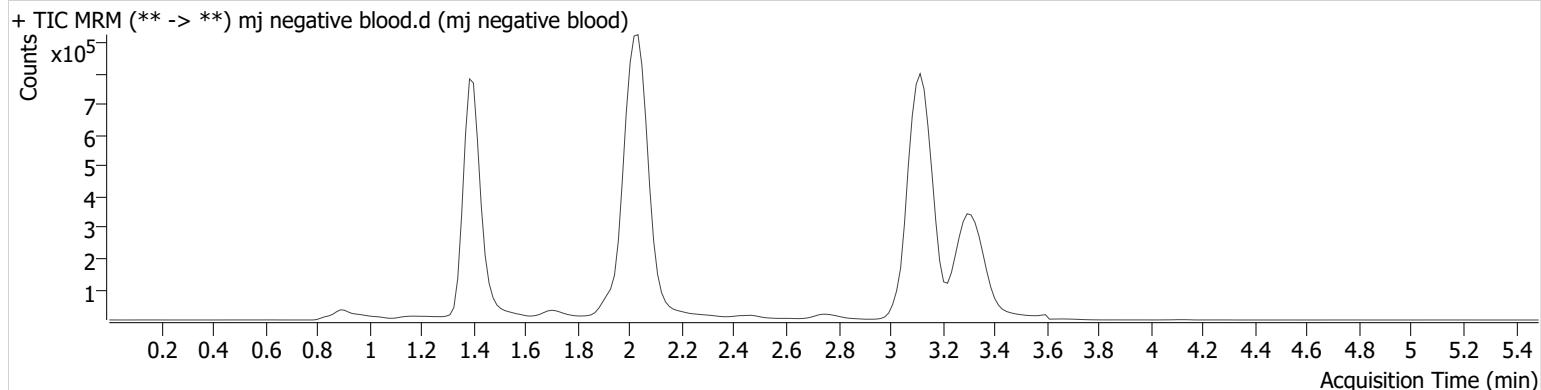


# AM #27 Cannabinoids

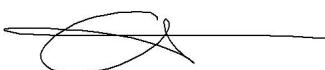
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**Calibration Last Update** 1/12/2022 3:20:10 PM

<b>Instrument</b>	69679	<b>Data File</b>	mj negative blood.d
<b>Type</b>	Sample	<b>Sample</b>	mj negative blood
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-B2	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 12:42:05 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.454	6801	∞	1600.3 High	∞	2714699	0.468 ng/ml Low



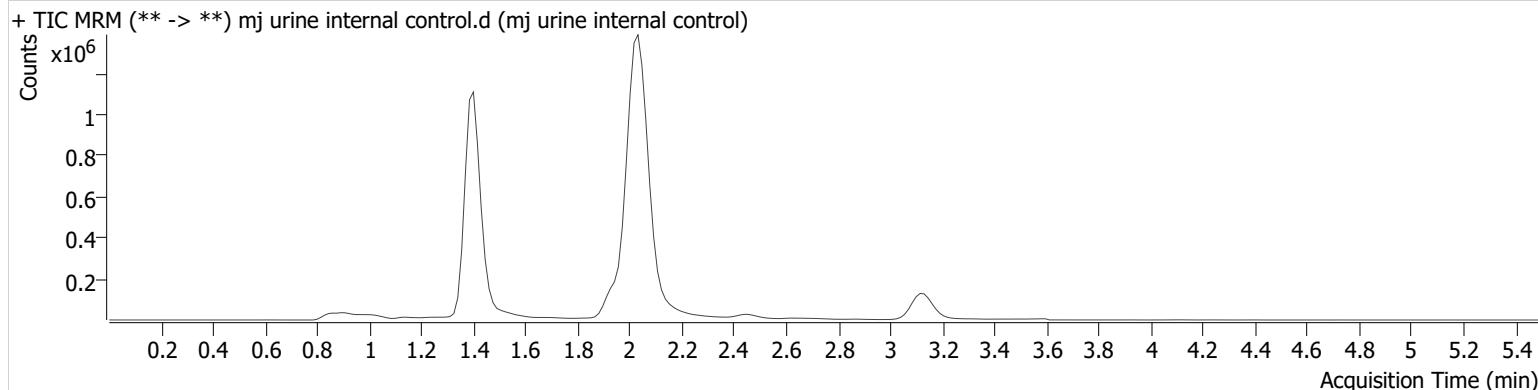
# AM #27 Cannabinoids

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**Calibration Last Update** 1/12/2022 3:20:10 PM

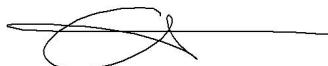
<b>Instrument</b>	69679	<b>Data File</b>	mj urine internal control.d
<b>Type</b>	Sample	<b>Sample</b>	mj urine internal control
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-A2	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 1:42:16 PM		

## Sample Info.

### Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	60185	$\infty$	868.6	$\infty$	3389819	4.931 ng/ml
THC-COOH	1.431	141706	446.7	32.8	378.7	578276	17.668 ng/ml
THC	3.137	75725	1695957797	26.5	488.5	664892	4.524 ng/ml



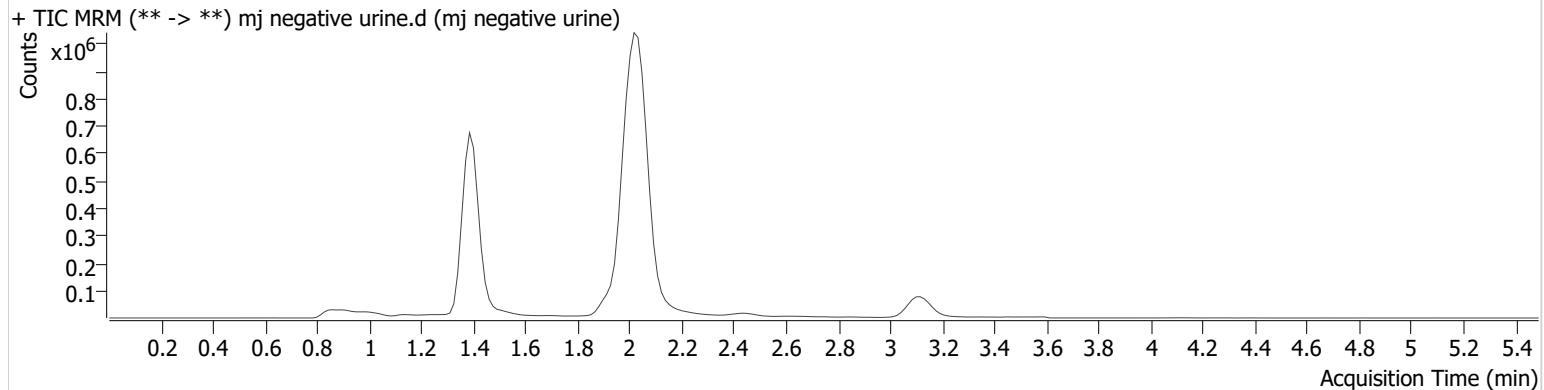
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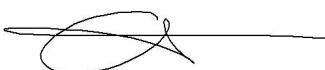
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**Calibration Last Update** 1/12/2022 3:20:10 PM

<b>Instrument</b>	69679	<b>Data File</b>	mj negative urine.d
<b>Type</b>	Sample	<b>Sample</b>	mj negative urine
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-G2	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 1:48:58 PM		

## Sample Info.

### Sample Chromatogram





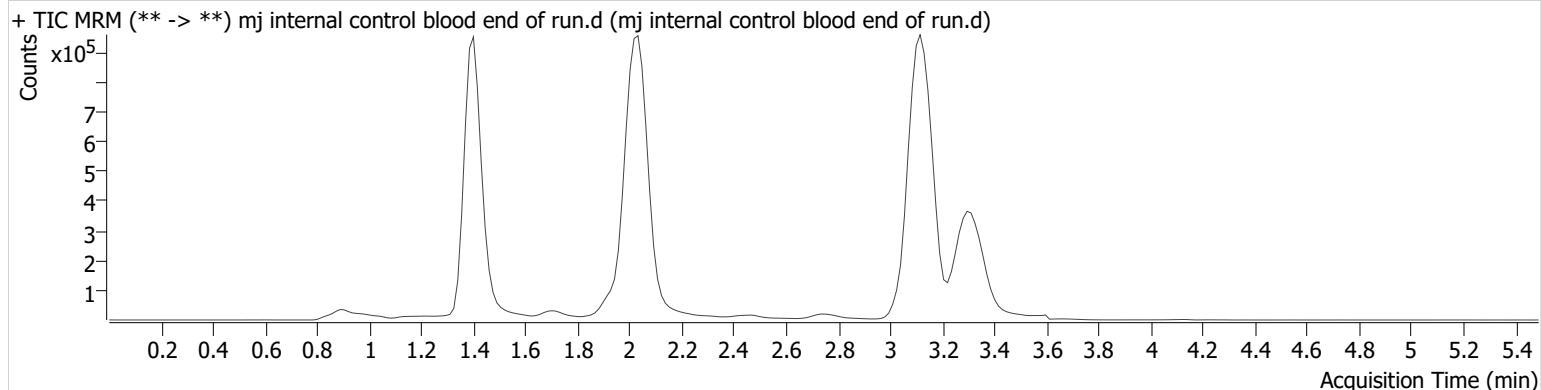
# AM #27 Cannabinoids

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**Calibration Last Update** 1/12/2022 3:20:10 PM

<b>Instrument</b>	69679	<b>Data File</b>	mj internal control blood end of run.d
<b>Type</b>	QC	<b>Sample</b>	mj internal control blood end of run.d
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-H1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 3:08:58 PM		

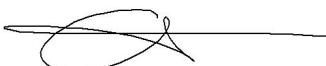
## Sample Info.

### Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	48096	∞	1208.2 High	∞	2604917	5.138 ng/ml
THC-COOH	1.416	175926	206.3	35.4	244.8	832862	15.343 ng/ml
THC	3.137	542134	∞	24.3	3191.1	4548848	4.718 ng/ml

# Compound Calibration Report

**Batch results**

D:\MassHunter\Data\2022\am 27-28\011222\QuantResults\cann.batch.bin

**Last Cal. Update**

1/12/2022 3:20 PM

**Analyst Name**

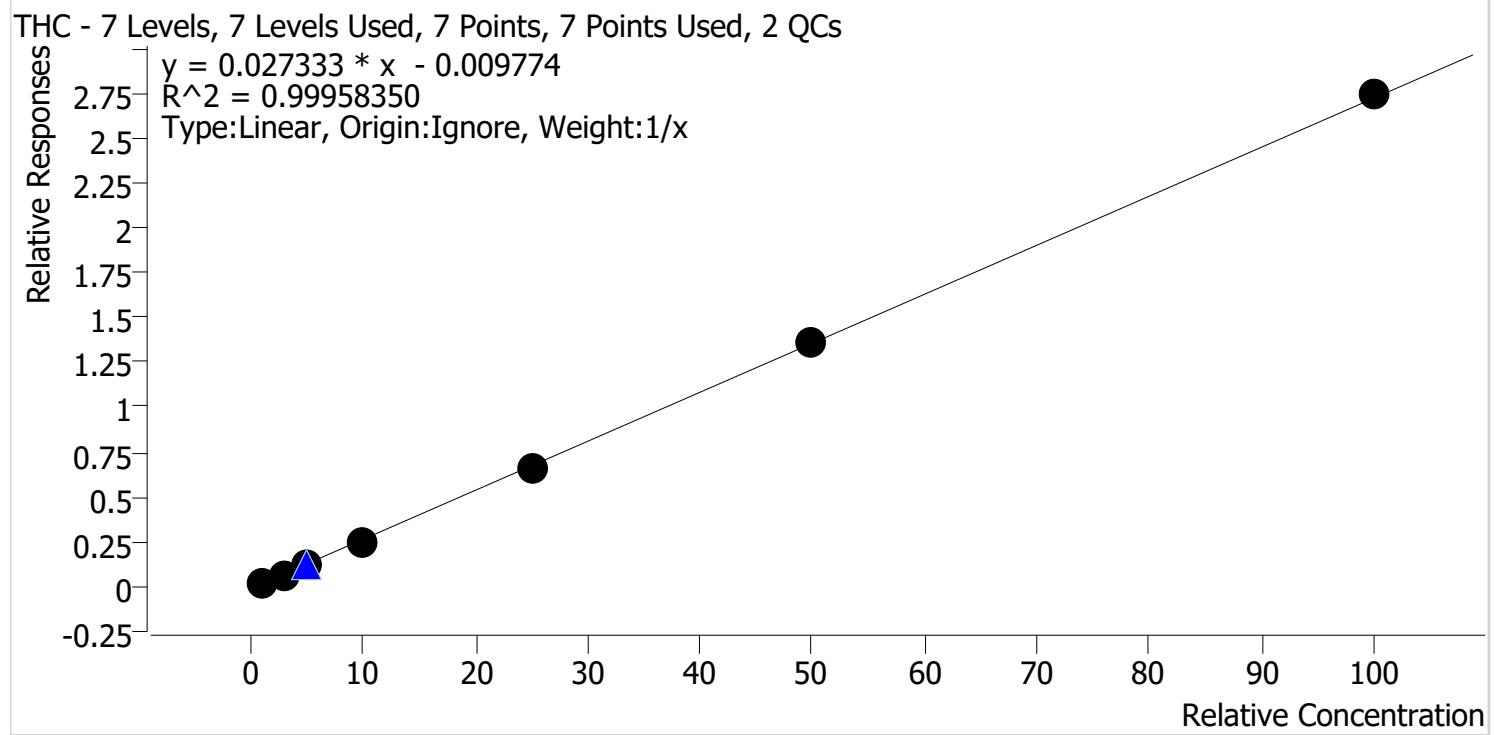
ISP\datastor

**Analyte**

THC

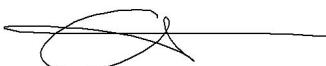
**Internal Standard**

THC-d3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✓	1.0	1.1	114.5
mj cal2	2	✓	3.0	2.9	95.2
mj cal 3	3	✓	5.0	4.7	94.0
mj cal 4	4	✓	10.0	9.6	96.1
mj cal 5	5	✓	25.0	24.8	99.1
mj cal 6	6	✓	50.0	50.2	100.4
mj cal 7	7	✓	100.0	100.7	100.7

# Compound Calibration Report

**Batch results**

D:\MassHunter\Data\2022\am 27-28\011222\QuantResults\cann.batch.bin

**Last Cal. Update**

1/12/2022 3:20 PM

**Analyst Name**

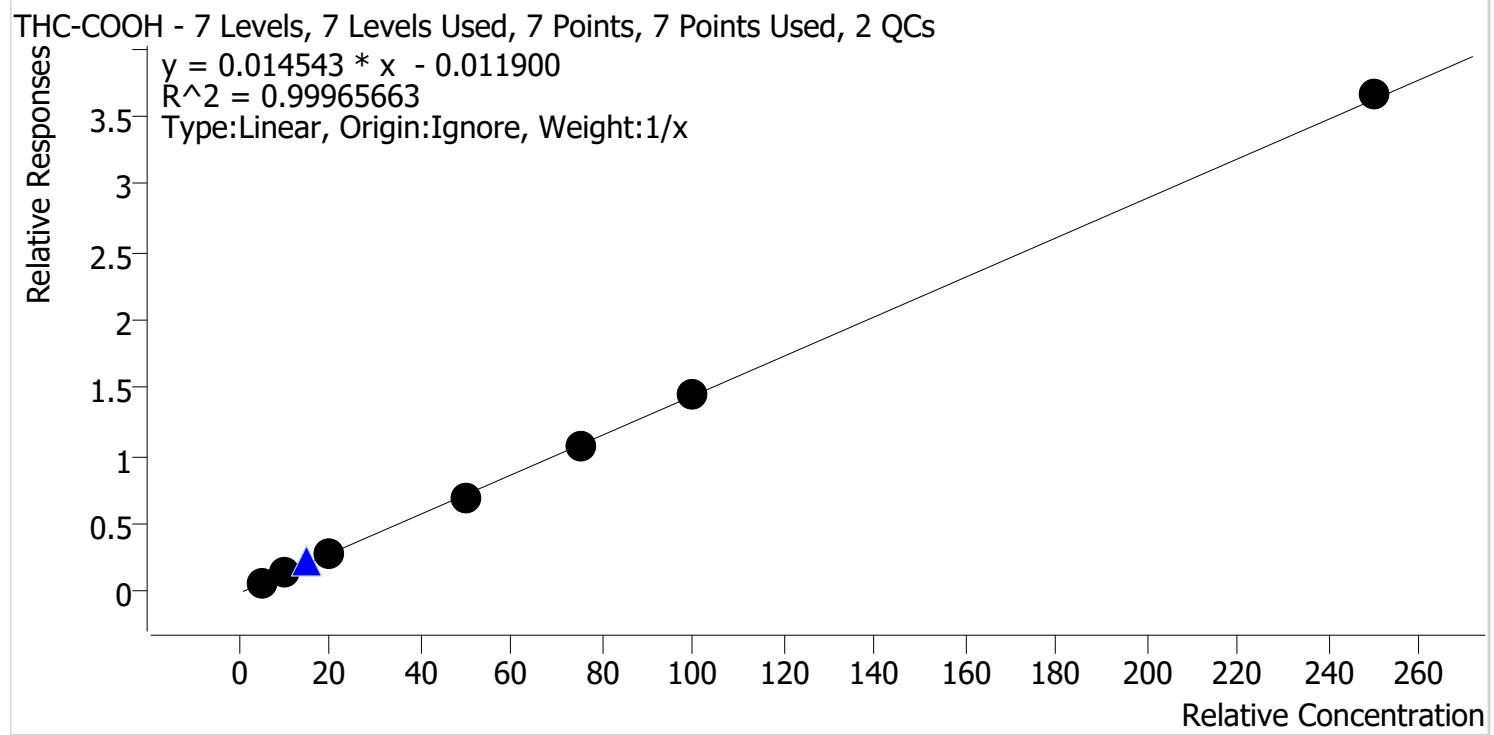
ISP\datastor

**Analyte**

THC-COOH

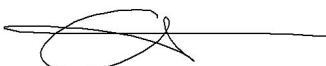
**Internal Standard**

THC-COOH-d9



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✓	5.0	5.3	105.6
mj cal2	2	✓	10.0	9.9	99.3
mj cal 3	3	✓	20.0	19.7	98.7
mj cal 4	4	✓	50.0	48.1	96.2
mj cal 5	5	✓	75.0	74.0	98.6
mj cal 6	6	✓	100.0	100.5	100.5
mj cal 7	7	✓	250.0	252.5	101.0

# Compound Calibration Report

**Batch results**

D:\MassHunter\Data\2022\am 27-28\011222\QuantResults\cann.batch.bin

**Last Cal. Update**

1/12/2022 3:20 PM

**Analyst Name**

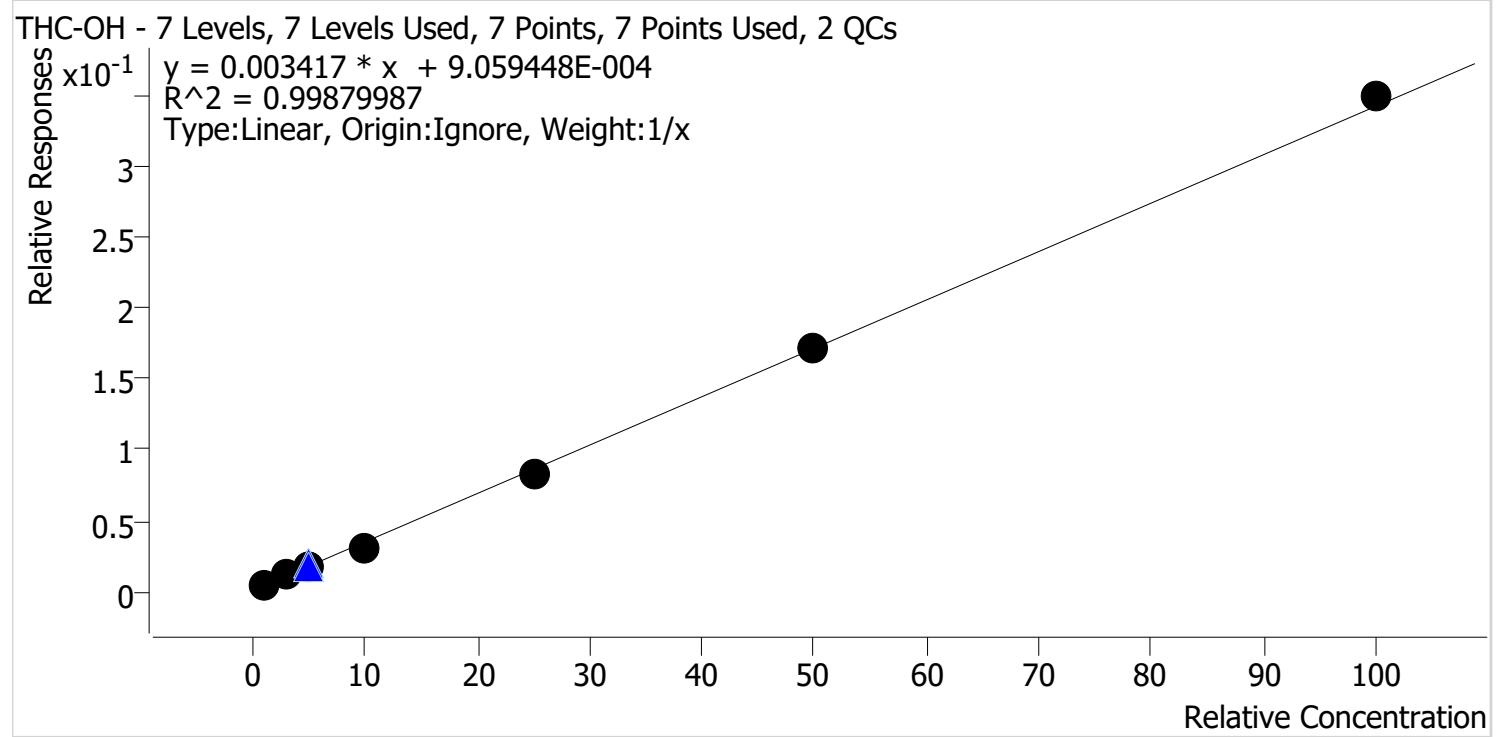
ISP\datastor

**Analyte**

THC-OH

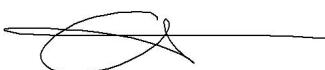
**Internal Standard**

THC-OH-d3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✓	1.0	1.1	114.9
mj cal2	2	✓	3.0	3.0	101.3
mj cal 3	3	✓	5.0	4.8	95.2
mj cal 4	4	✓	10.0	9.0	90.1
mj cal 5	5	✓	25.0	24.2	96.7
mj cal 6	6	✓	50.0	49.9	99.8
mj cal 7	7	✓	100.0	102.0	102.0

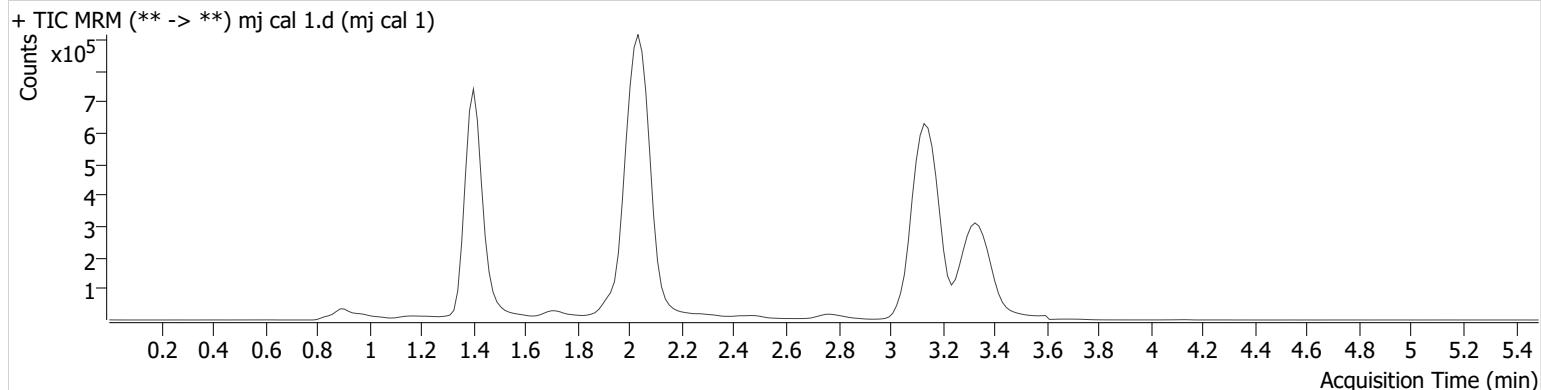
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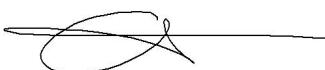
# AM #27 Cannabinoids

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**Calibration Last Update** 1/12/2022 3:20:10 PM

<b>Instrument</b>	69679	<b>Data File</b>	mj cal 1.d
<b>Type</b>	Cal	<b>Sample</b>	mj cal 1
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-A1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 11:41:46 AM		

**Sample Info.****Sample Chromatogram**

Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.409	11877	$\infty$	2236.1 High	$\infty$	2458138	1.149 ng/ml	Low
THC-COOH	1.431	46172	135.5	37.1	370.4	711232	5.282 ng/ml	
THC	3.167	73850	$\infty$	25.1	294.1	3433497	1.145 ng/ml	



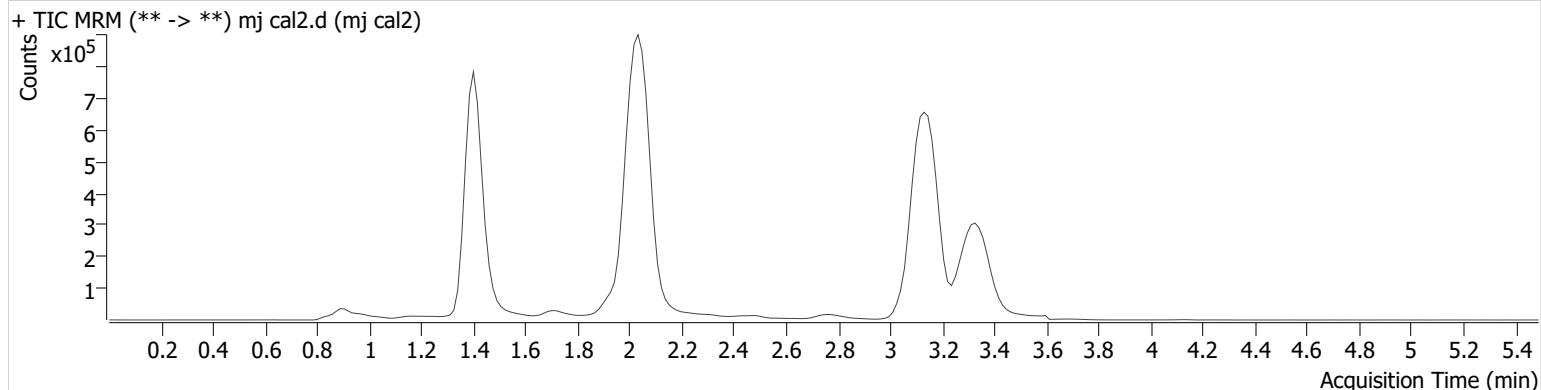
# AM #27 Cannabinoids

**Batch results** D:\MassHunter\Data\2022\am 27-28\011222\QuantResults\cann.batch.bin  
**Calibration Last Update** 1/12/2022 3:20:10 PM

<b>Instrument</b>	69679	<b>Data File</b>	mj cal2.d
<b>Type</b>	Cal	<b>Sample</b>	mj cal2
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-B1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 11:48:31 AM		

**Sample Info.**

**Sample Chromatogram**



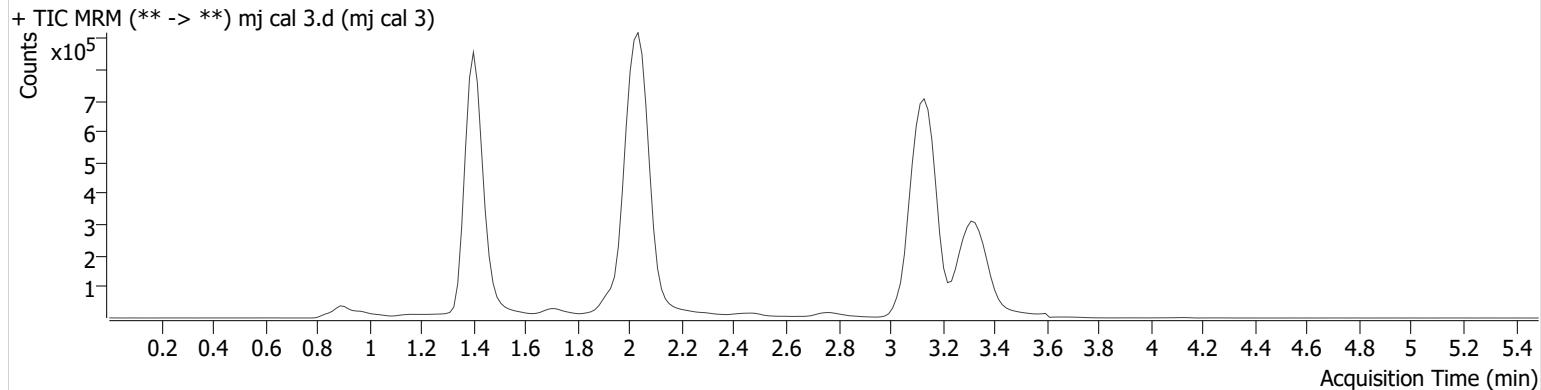
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.409	27587	$\infty$	1439.3 High	$\infty$	2442479	3.040 ng/ml
THC-COOH	1.431	93968	337.0	36.4	55.8	708809	9.934 ng/ml
THC	3.152	231009	36866.5	25.1	1076.7	3382812	2.856 ng/ml

# AM #27 Cannabinoids

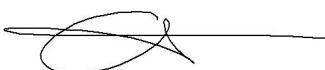
Batch results D:\MassHunter\Data\2022\am 27-28\011222\QuantResults\cann.batch.bin  
Calibration Last Update 1/12/2022 3:20:10 PM

Instrument	69679	Data File	mj cal 3.d
Type	Cal	Sample	mj cal 3
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-C1	Comment	
Injection Volume	10		
Acq. Date-Time	1/12/2022 11:55:13 AM		
Sample Info.			

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.409	41147	$\infty$	1281.9 High	$\infty$	2396011	4.761 ng/ml
THC-COOH	1.431	197446	169.4	34.7	151.2	717544	19.739 ng/ml
THC	3.152	407128 4618471638	55621.0	23.1	1375.3	3428688	4.702 ng/ml



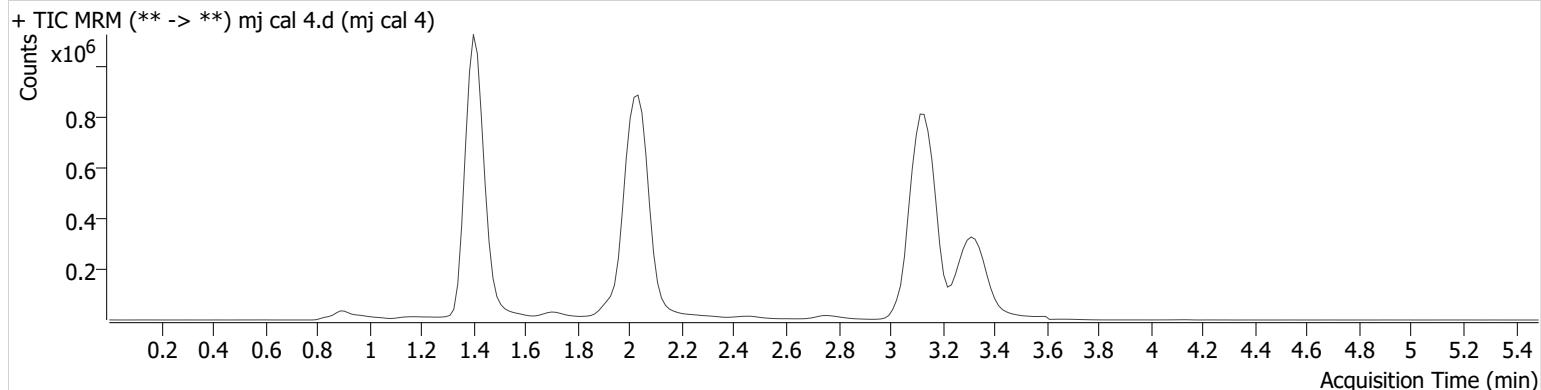
# AM #27 Cannabinoids

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**Calibration Last Update** 1/12/2022 3:20:10 PM

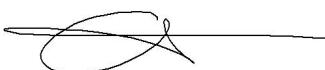
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<b>Type</b>	Cal	<b>Sample</b>	mj cal 4
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-D1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 12:01:55 PM		

**Sample Info.**

**Sample Chromatogram**



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	79535	∞	1139.1	∞	2510238	9.008 ng/ml
THC-COOH	1.431	516754	719.4	37.9	2098.4	751425	48.106 ng/ml
THC	3.152	900426	∞	22.7	∞	3560556	9.610 ng/ml



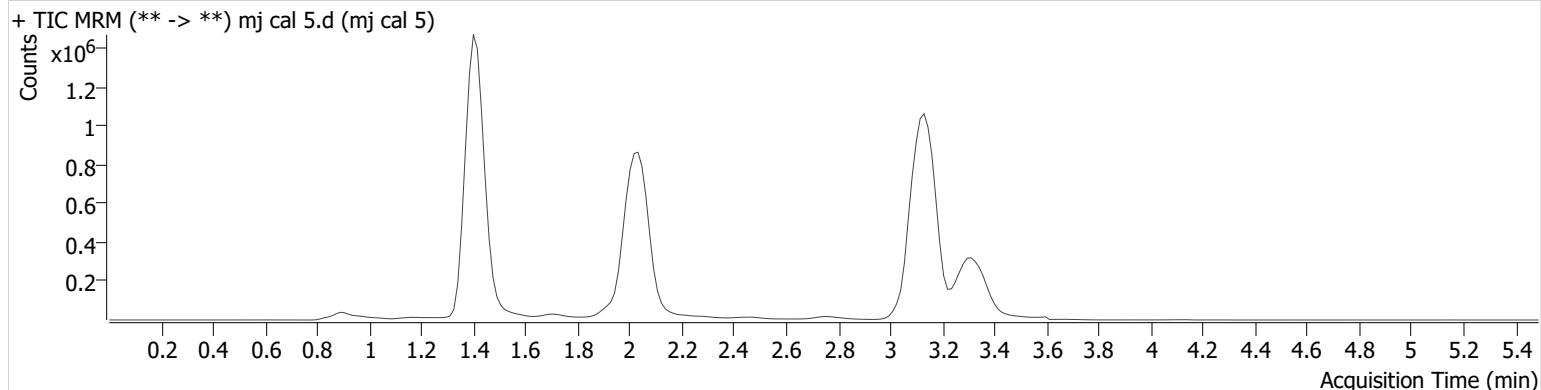
# AM #27 Cannabinoids

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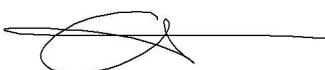
<b>Instrument</b>	69679	<b>Data File</b>	mj cal 5.d
<b>Type</b>	Cal	<b>Sample</b>	mj cal 5
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-E1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 12:08:37 PM		

**Sample Info.**

**Sample Chromatogram**



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	202323	$\infty$	969.9	$\infty$	2421822	24.184 ng/ml
THC-COOH	1.431	773516	2774.2	37.8	3371.2	727139	73.966 ng/ml
THC	3.152	2291536	$\infty$	23.7	$\infty$	3434828	24.766 ng/ml

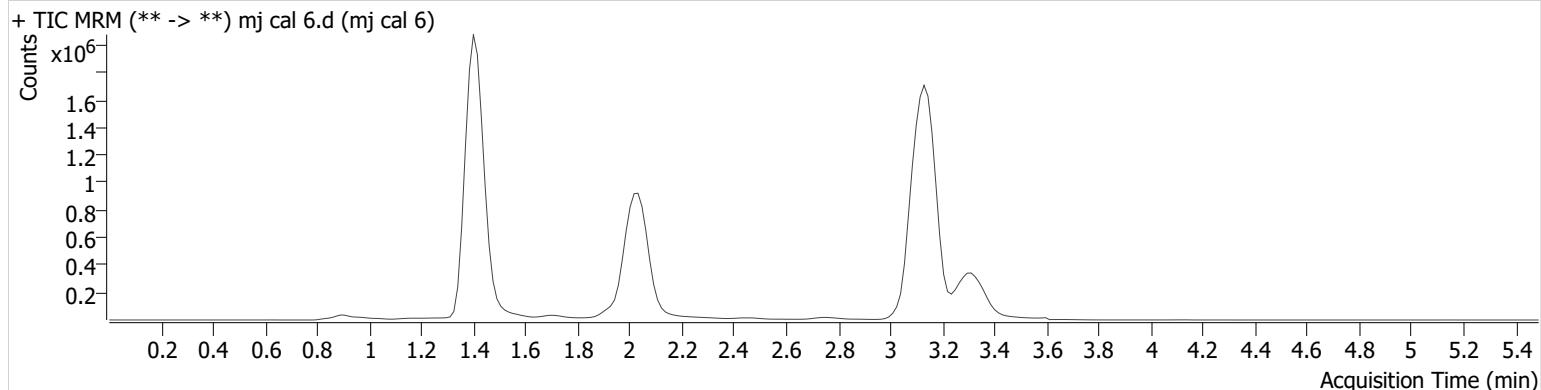


# AM #27 Cannabinoids

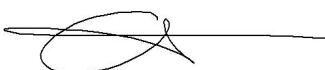
**Batch results** D:\MassHunter\Data\2022\am 27-28\011222\QuantResults\cann.batch.bin  
**Calibration Last Update** 1/12/2022 3:20:10 PM

<b>Instrument</b>	69679	<b>Data File</b>	mj cal 6.d
<b>Type</b>	Cal	<b>Sample</b>	mj cal 6
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-F1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 12:15:19 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	424473	$\infty$	900.8	$\infty$	2477158	49.883 ng/ml
THC-COOH	1.431	1043642	10625.2	37.8	302.3	719951	100.495 ng/ml
THC	3.152	5186164	$\infty$	24.7	$\infty$	3805085	50.223 ng/ml



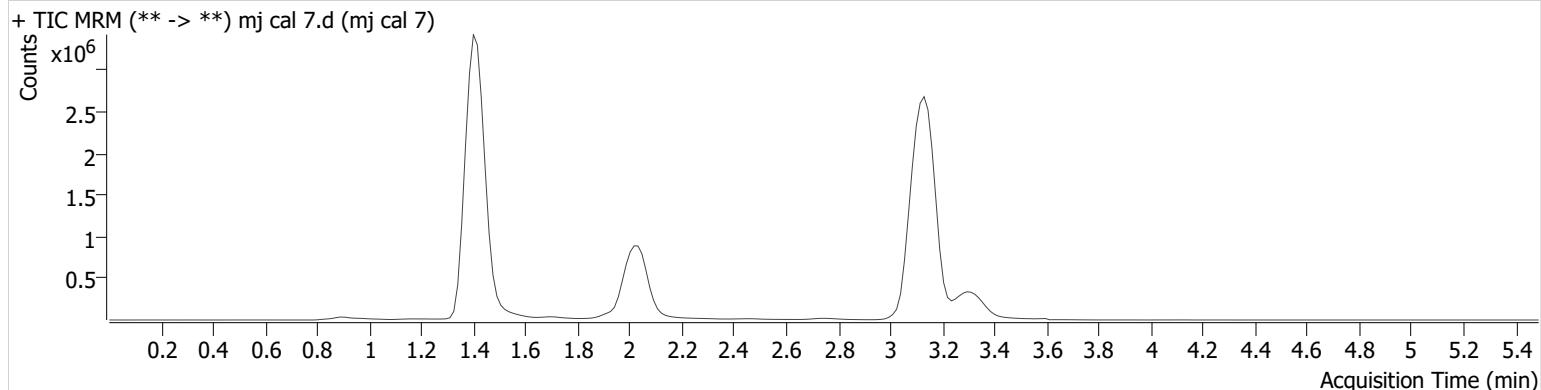
# AM #27 Cannabinoids

**Batch results** D:\MassHunter\Data\2022\am 27-28\011222\QuantResults\cann.batch.bin  
**Calibration Last Update** 1/12/2022 3:20:10 PM

<b>Instrument</b>	69679	<b>Data File</b>	mj cal 7.d
<b>Type</b>	Cal	<b>Sample</b>	mj cal 7
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Anne Nord
<b>Sample Position</b>	P3-G1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	1/12/2022 12:22:03 PM		

**Sample Info.**

**Sample Chromatogram**



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	827584	$\infty$	858.5	$\infty$	2368938	101.975 ng/ml
THC-COOH	1.431	2396980	1635.4	38.9	2037.5	654933	252.478 ng/ml
THC	3.136	10359047	$\infty$	24.4	$\infty$	3777114	100.698 ng/ml