








**Worklist: 4697**

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>	
M2020-3796	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
M2020-4827	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
M2020-4999	3	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3619	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3656	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3657	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3672	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3707	2	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3710	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3742	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3754	2	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3762	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3765	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3766	1	BCK	AM 27 Blood THC Quant by LC-QQQ	

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

Extraction Date: 12/30/2020

Plate lot#: IDP-108-2-200723

Mobile phase A: 0.1% Formic Acid in LCMS Water

Blank Blood Lot: Lampire 20L20725

LCMS-QQQ ID: 069901

Analyst: Sophia Jackson

Plate Expiration: 01/23/2021

Mobile phase B: 0.1% Formic acid in Acetonitrile

Column: UCT Selectra DA 100 x 2.1mm 3um

## 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: 42**
- 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 700-800µL 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)*
- 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.
- 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, Curve weighting of Linear 1/x with r<sup>2</sup> values ≥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 (quantitative), OH-THC 3ng/mL (quantitative), and Carboxy-THC 5ng/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 QCs pass for each analyte? (if not, describe in comments section)
- 6. Enter QCs into control charting.
- 7. Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

### COMMENTS:

Samples P2020-3657-1 and P2020-3742-1 were reconstituted and re-injected due to poor ISTD response. Re-inject data analyzed for both

SJ

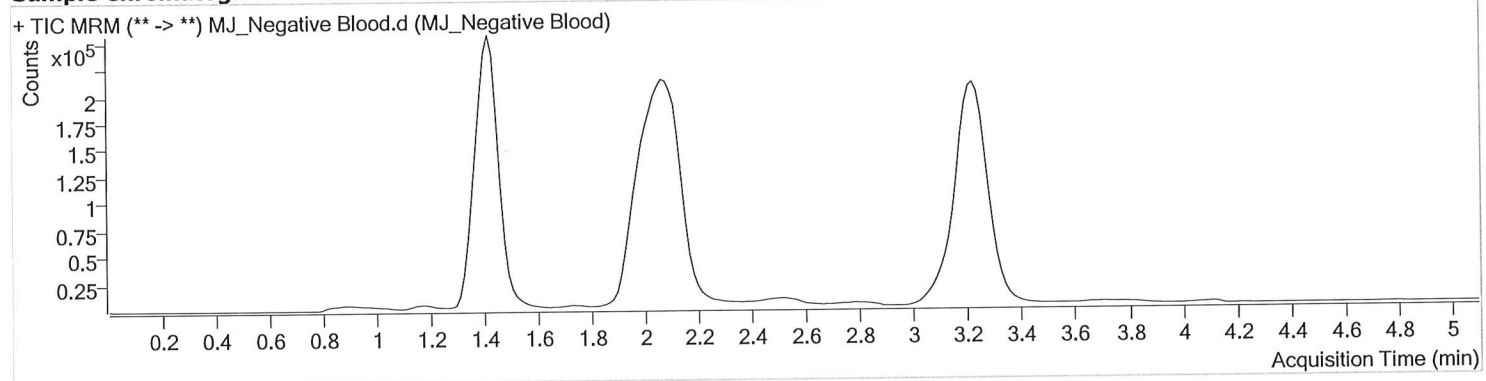


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 1/4/2021 10:40:44 AM

<b>Instrument</b>	Instrument 1	<b>Data File</b>	MJ_Negative Blood.d
<b>Type</b>	Sample	<b>Sample</b>	MJ_Negative Blood
<b>Acq. Method</b>	AM 27 THCQ.m	<b>Operator</b>	Sophia Jackson
<b>Sample Position</b>	P1-A2	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	12/30/2020 2:10:59 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



SJ

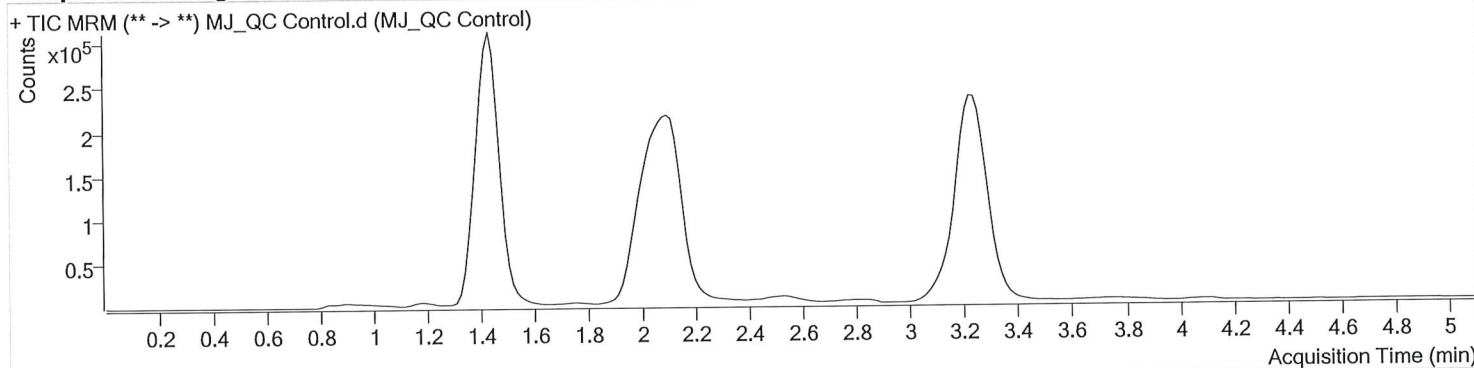


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
Calibration Last Update 1/4/2021 10:40:44 AM

<b>Instrument</b>	Instrument 1	<b>Data File</b>	MJ_QC Control.d
<b>Type</b>	Sample	<b>Sample</b>	MJ_QC Control
<b>Acq. Method</b>	AM 27 THCQ.m	<b>Operator</b>	Sophia Jackson
<b>Sample Position</b>	P1-H1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	12/30/2020 1:55:46 PM		
<b>Sample Info.</b>			

### Sample Chromatogram

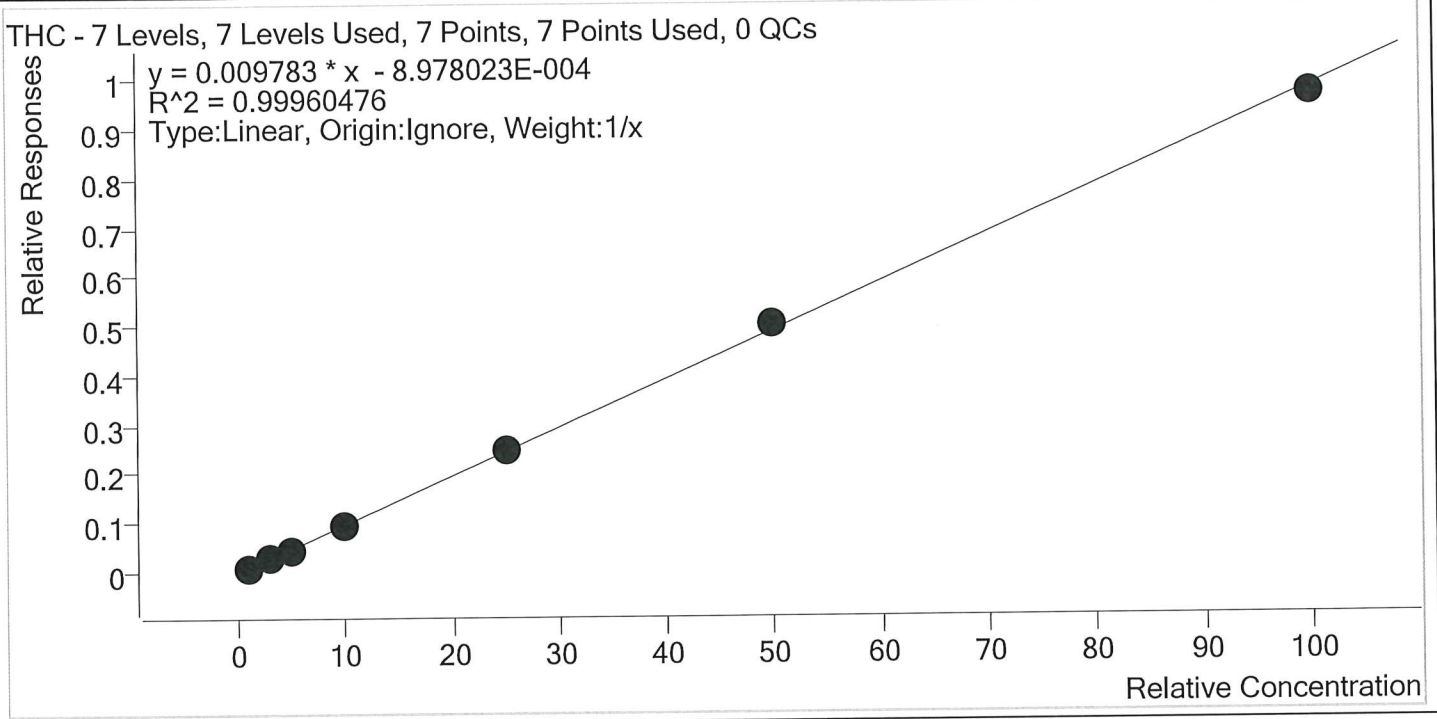


Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	98777	∞	11.6	241.69	1233327	4.6731 ng/ml
THC-COOH	1.459	121473	∞	57.6	830.52	333327	15.3256 ng/ml
THC	3.239	81640	∞	30.4	79.68	1805752	4.7133 ng/ml



# AM #27 Cannabinoids Quant. Calibration Curve Report

**Batch results** D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
**Last Cal. Update** 1/4/2021 10:40 AM  
**Analyst Name** ISP\datastor  
**Analyte** THC **Internal Standard** THC-D3



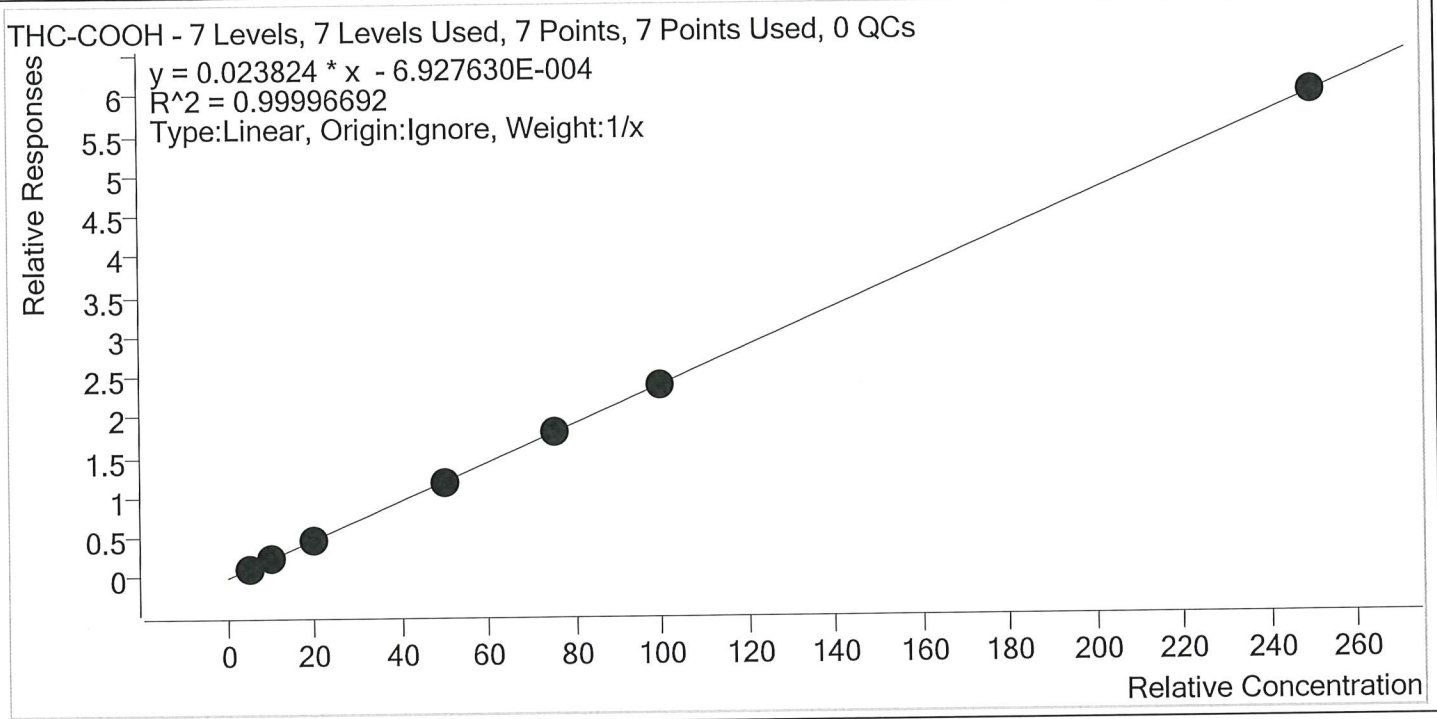
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ Cal 1	1	✓	1.0	1.0	101.6
MJ Cal 2	2	✓	3.0	3.0	99.3
MJ Cal 3	3	✓	5.0	4.9	97.8
MJ Cal 4	4	✓	10.0	9.9	99.3
MJ Cal 5	5	✓	25.0	25.1	100.6
MJ Cal 6	6	✓	50.0	51.4	102.9
MJ Cal 7	7	✓	100.0	98.6	98.6

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# AM #27 Cannabinoids Quant. Calibration Curve Report

**Batch results** D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
**Last Cal. Update** 1/4/2021 10:40 AM  
**Analyst Name** ISP\datastor  
**Analyte** THC-COOH **Internal Standard** THC-COOH-D9



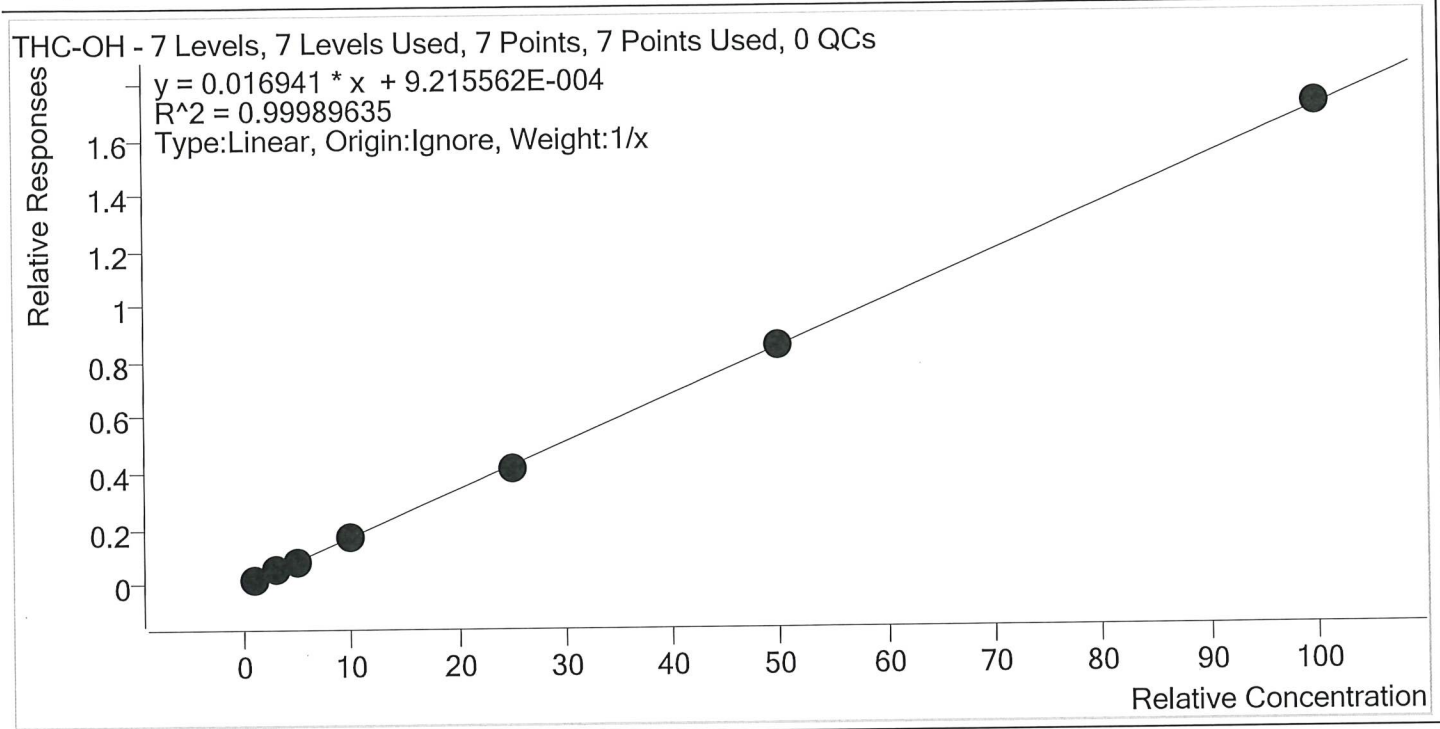
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ Cal 1	1	✓	5.0	4.9	99.0
MJ Cal 2	2	✓	10.0	10.0	100.3
MJ Cal 3	3	✓	20.0	20.4	101.8
MJ Cal 4	4	✓	50.0	49.6	99.2
MJ Cal 5	5	✓	75.0	74.6	99.5
MJ Cal 6	6	✓	100.0	100.0	100.0
MJ Cal 7	7	✓	250.0	250.4	100.2

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# AM #27 Cannabinoids Quant. Calibration Curve Report

**Batch results** D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
**Last Cal. Update** 1/4/2021 10:40 AM  
**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	1.0	103.2
MJ Cal 2	2	✓	3.0	3.0	99.4
MJ Cal 3	3	✓	5.0	5.0	100.7
MJ Cal 4	4	✓	10.0	9.8	97.8
MJ Cal 5	5	✓	25.0	24.6	98.5
MJ Cal 6	6	✓	50.0	49.8	99.7
MJ Cal 7	7	✓	100.0	100.7	100.7

SJ

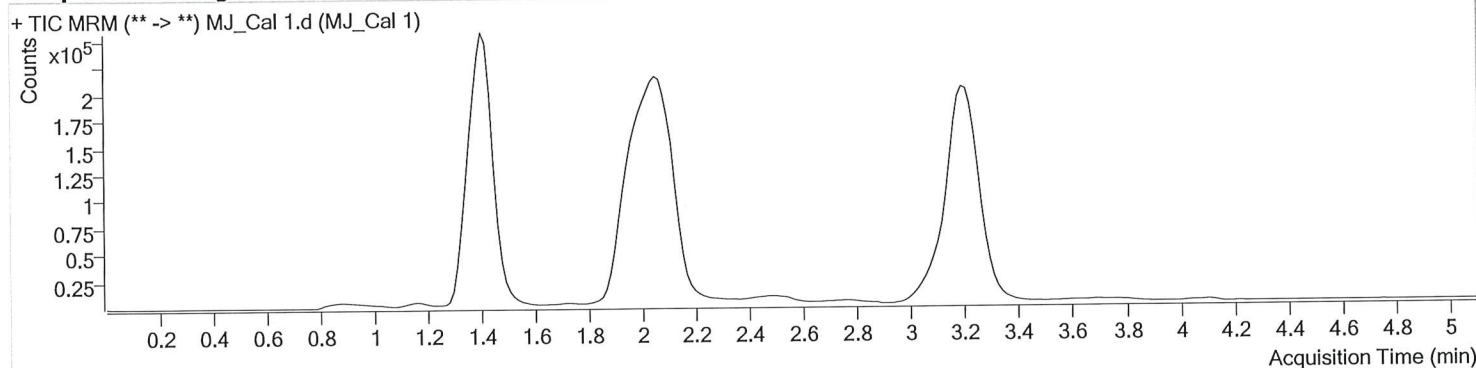


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
 Calibration Last Update 1/4/2021 10:40:44 AM

<b>Instrument</b>	Instrument 1	<b>Data File</b>	MJ_Cal 1.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 1
<b>Acq. Method</b>	AM 27 THCQ.m	<b>Operator</b>	Sophia Jackson
<b>Sample Position</b>	P1-A1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	12/30/2020 12:54:50 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.423	22336	∞	11.3	18.35	1213862	1.0317 ng/ml <b>Low</b>
THC-COOH	1.429	38142	543.70	55.9	∞	325509	4.9475 ng/ml <b>Low</b>
THC	3.224	16154	63.42	29.6	18.96	1787266	1.0157 ng/ml



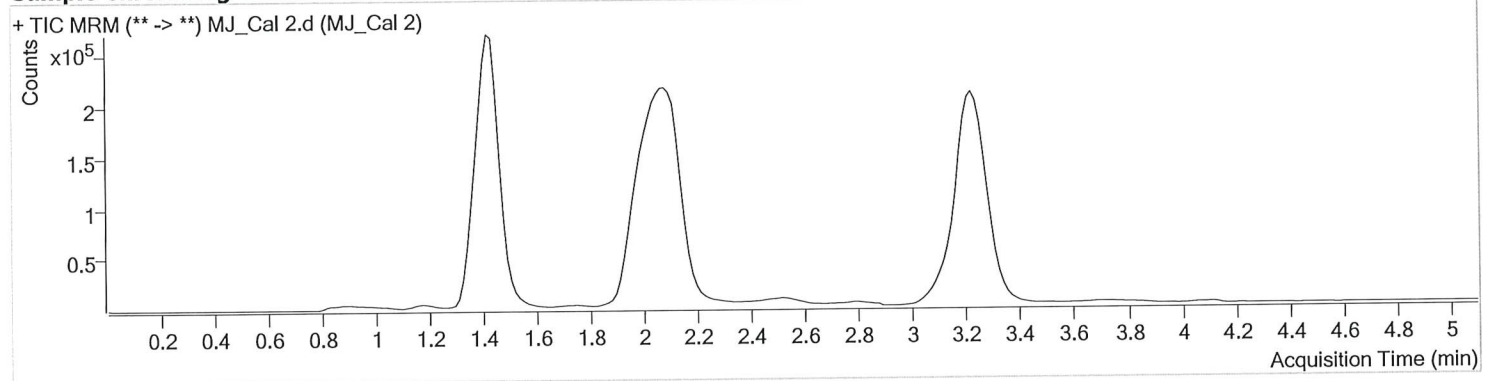


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
 Calibration Last Update 1/4/2021 10:40:44 AM

<b>Instrument</b>	Instrument 1	<b>Data File</b>	MJ_Cal 2.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 2
<b>Acq. Method</b>	AM 27 THCQ.m	<b>Operator</b>	Sophia Jackson
<b>Sample Position</b>	P1-B1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	12/30/2020 1:02:35 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.423	60289	∞	12.0	176.30	1172206	2.9815 ng/ml <b>Low</b>
THC-COOH	1.459	74687	∞	56.3	704.38	313432	10.0311 ng/ml
THC	3.224	49559	130.90	31.0	∞	1754027	2.9799 ng/ml

SJ

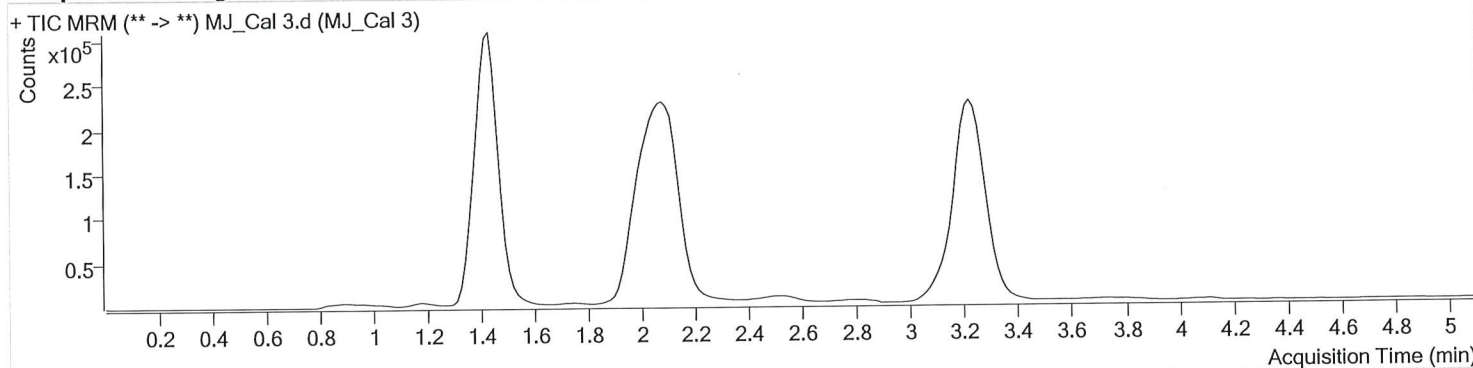


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 1/4/2021 10:40:44 AM

<b>Instrument</b>	Instrument 1	<b>Data File</b>	MJ_Cal 3.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 3
<b>Acq. Method</b>	AM 27 THCQ.m	<b>Operator</b>	Sophia Jackson
<b>Sample Position</b>	P1-C1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	12/30/2020 1:10:11 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	102882	∞	11.9	121.70	1192840	5.0367 ng/ml
THC-COOH	1.459	155319	581.85	56.9	967.94	320546	20.3675 ng/ml
THC	3.239	86322	775.90	30.0	247.43	1839590	4.8885 ng/ml

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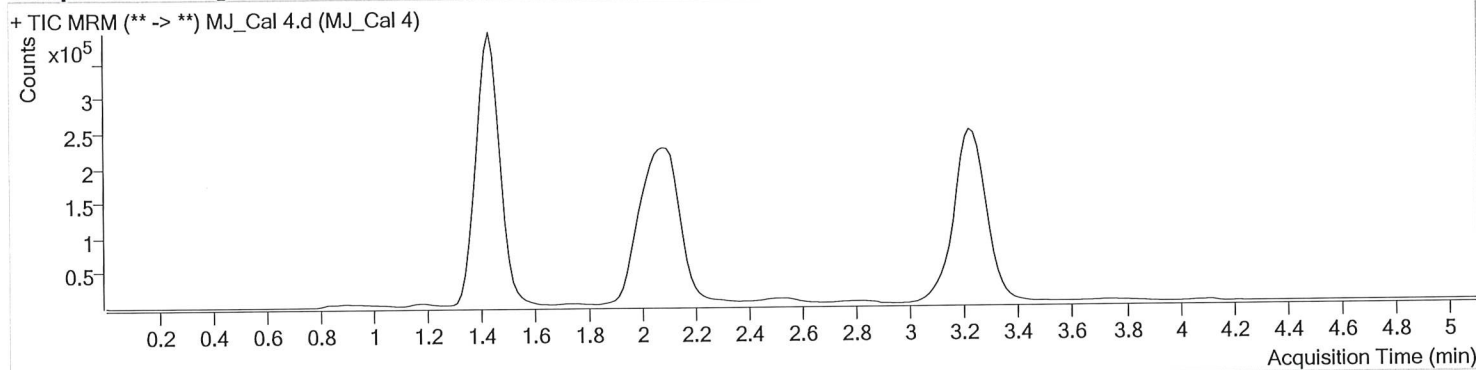


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 1/4/2021 10:40:44 AM

<b>Instrument</b>	Instrument 1	<b>Data File</b>	MJ_Cal 4.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 4
<b>Acq. Method</b>	AM 27 THCQ.m	<b>Operator</b>	Sophia Jackson
<b>Sample Position</b>	P1-D1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	12/30/2020 1:17:46 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	198915	∞	11.7	433.80	1193718	9.7816 ng/ml
THC-COOH	1.459	373082	∞	59.1	1875.30	315797	49.6175 ng/ml
THC	3.239	171410	2004.94	27.5	146.36	1781750	9.9257 ng/ml

SJ

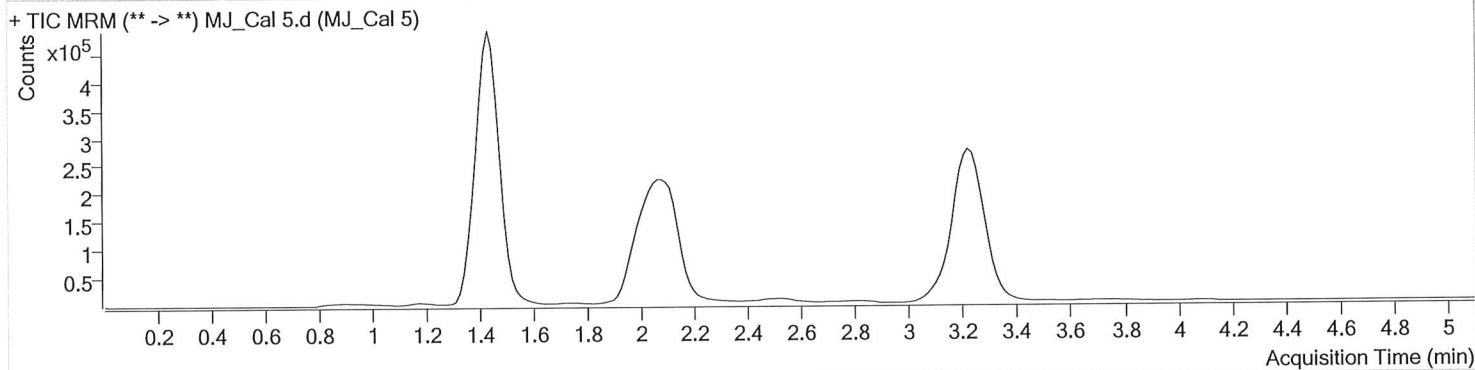


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
 Calibration Last Update 1/4/2021 10:40:44 AM

<b>Instrument</b>	Instrument 1	<b>Data File</b>	MJ_Cal 5.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 5
<b>Acq. Method</b>	AM 27 THCQ.m	<b>Operator</b>	Sophia Jackson
<b>Sample Position</b>	P1-E1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	12/30/2020 1:25:22 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.423	494454	∞	11.9	1562.75	1182404	24.6294 ng/ml
THC-COOH	1.459	553475	∞	59.2	2236.01	311394	74.6348 ng/ml
THC	3.239	418434	1366.45	26.6	∞	1707045	25.1483 ng/ml

SJ

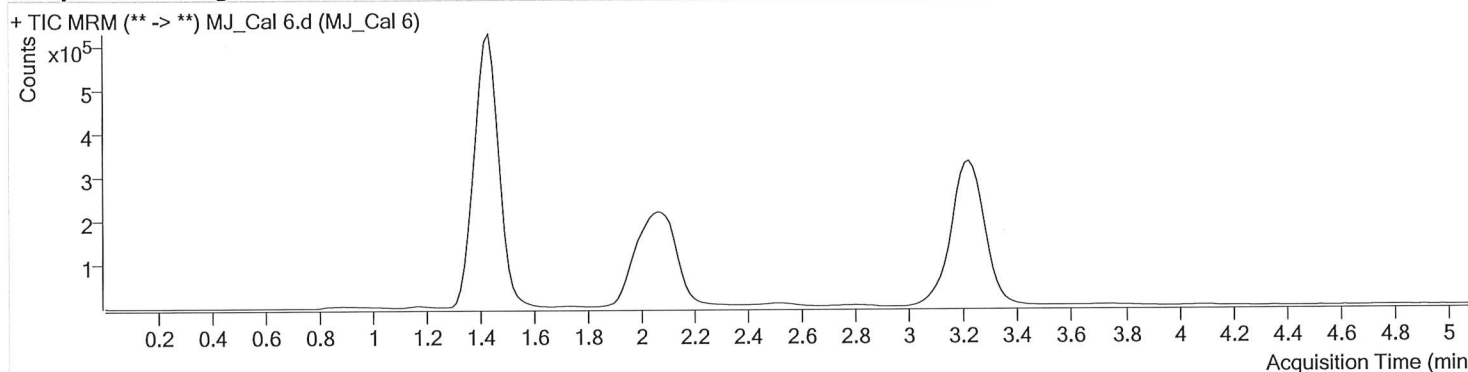


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 1/4/2021 10:40:44 AM

<b>Instrument</b>	Instrument 1	<b>Data File</b>	MJ_Cal 6.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 6
<b>Acq. Method</b>	AM 27 THCQ.m	<b>Operator</b>	Sophia Jackson
<b>Sample Position</b>	P1-F1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	12/30/2020 1:32:58 PM		
<b>Sample Info.</b>			

**Sample Chromatogram**



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.423	1000639	∞	11.9	5473.59	1183888	49.8363 ng/ml
THC-COOH	1.444	738324	3204.37	58.8	2200.62	310038	99.9870 ng/ml
THC	3.224	858891	4664.39	24.7	385.70	1709820	51.4402 ng/ml

SJ

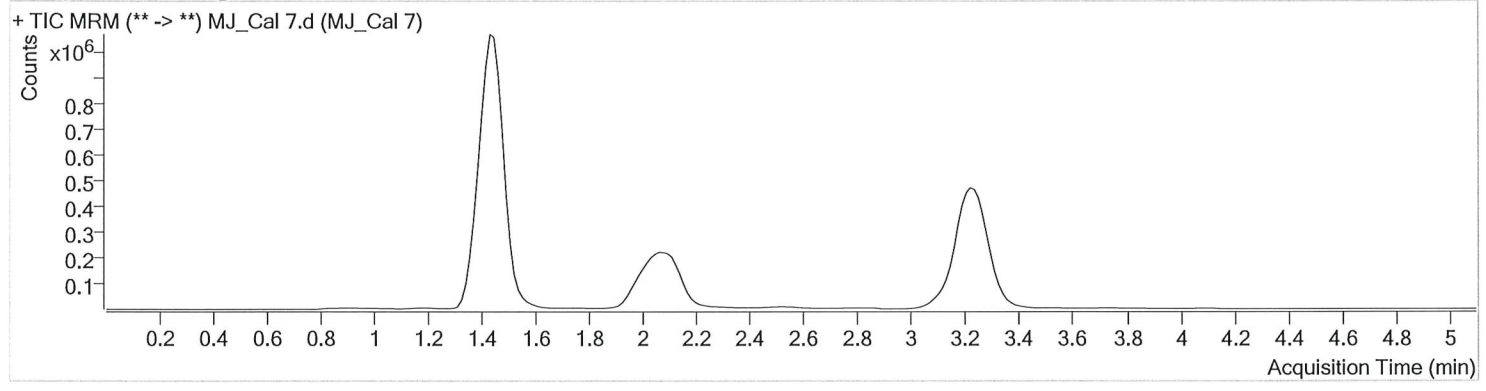


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2020\AM 27-28\122920 AM 27 28 SJ\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 1/4/2021 10:40:44 AM

<b>Instrument</b>	Instrument 1	<b>Data File</b>	MJ_Cal 7.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 7
<b>Acq. Method</b>	AM 27 THCQ.m	<b>Operator</b>	Sophia Jackson
<b>Sample Position</b>	P1-G1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	12/30/2020 1:40:34 PM		

**Sample Chromatogram**



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.423	1967521	∞	12.3	3851.15	1152646	100.7028 ng/ml
THC-COOH	1.459	1768548	2986.07	59.5	15015.8	296478	250.4146 ng/ml
THC	3.239	1789537	∞	25.1	1849.61	1856947	98.6017 ng/ml