

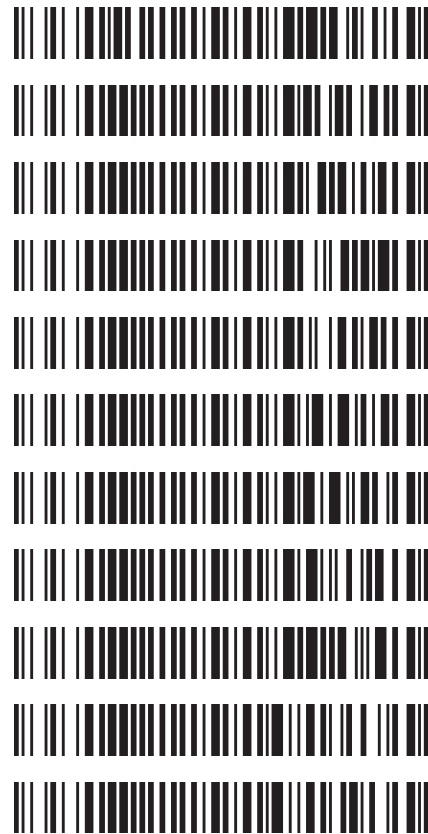
**REVIEWED**  
By Tamara Salazar at 2:06 pm, Jun 06, 2023

6/5/2023

CS

**Worklist: 6391**

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
M2023-2027	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2023-1118	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2023-1147	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2023-1305	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2023-1322	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2023-1387	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2023-1397	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2023-1401	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2023-1423	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2023-1437	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2023-1438	1	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: 05/26/2023

Analyst: Celena Shrum

Plate lot#: 220802

Plate Retest Date: 07/23/2023

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

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

**Blank Blood Lot:** Lampire 23A52594

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

**LCMS-QQQ ID:** 069901

### 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 (if applicable): add 1.5mL urine to blank plate, add 250µl 1N KOH. Shake and incubate at 40 degrees for 15 minutes.
- 3. Using a calibrated pipette, add **1000µl blood or 1000µl hydrolyzed urine** into the appropriate wells of the analytical (standards) plate. **Pipette ID: #42**
- 4. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 5. Add **500µL of 0.1% formic acid in water to blood samples or 500µl of saturated phosphate buffer to urine samples** to the appropriate wells of the analytical plate.
- 6. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 7. Transfer **800µL of blood+acid mixture or urine+acid** to corresponding wells of SLE+ plate.
- 8. 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: 067104
- 9. Wait 5 minutes.
- 10. Add **2.25mL MTBE. (Add in 3 increments of 750uL)**
- 11. Wait 5 minutes.
- 12. Apply positive pressure for approx. 15 seconds. **(10-15 PSI- Selector to the left).**
- 13. Add **2.25mL Hexane. (Add in 3 increments of 750uL)**
- 14. Wait 5 minutes.
- 15. Apply positive pressure for approx. 15 seconds. **(10-15 PSI- Selector to the left).**
- 16. Remove plate containing eluate. Place on SPE Dry and evaporate to dryness at approx. 35°C. **SPE Dry ID: 067103**
- 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 and OH-THC 3ng/mL (quantitative), 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. THC concentrations of 1-3ng/mL will be reported qualitatively.
- 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:

	1	2	3	4	5	6
a					P2023-1387-1	QC 1
b					P2023-1322-2	cal 100 ng
c					P2023-1305-1	cal 50 ng
d				P2023-1438-1	P2023-1147-1	cal 25 ng
e				P2023-1437-1	P2023-1118-2	cal 10ng
f				P2023-1423-1	M2023-2027-2	cal 5 ng
g				P2023-1401-1	NEG Blood	cal 3 ng
h				P2023-1397-1	QC 2	cal 1ng

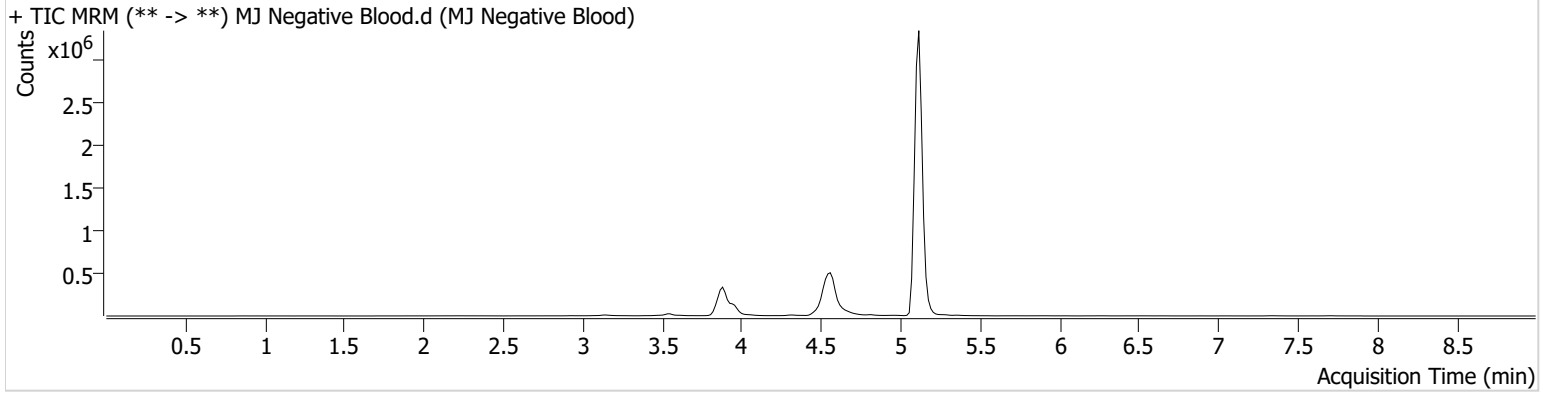


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 6/6/2023 12:04:56 PM

<b>Instrument</b>	Falco (069901)	<b>Data File</b>	MJ Negative Blood.d
<b>Type</b>	Sample	<b>Sample</b>	MJ Negative Blood
<b>Acq. Method</b>	AM 27 Agilent Method.m	<b>Operator</b>	Celena Shrum
<b>Sample Position</b>	P1-G5	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	5/26/2023 4:42:56 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



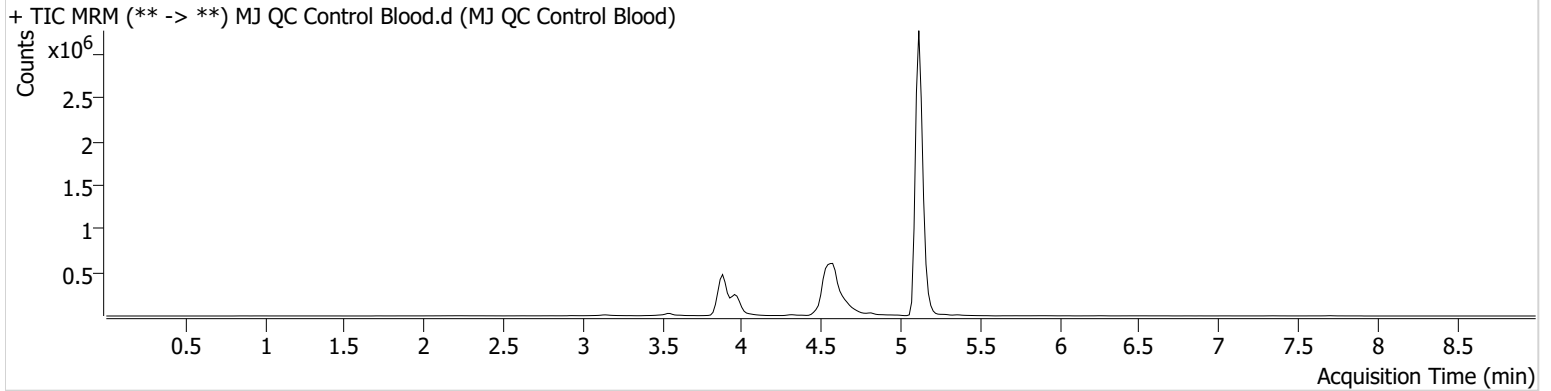


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 6/6/2023 12:04:56 PM

**Instrument** Falco (069901) **Data File** MJ QC Control Blood.d  
**Type** QC **Sample** MJ QC Control Blood  
**Acq. Method** AM 27 Agilent Method.m **Operator** Celena Shrum  
**Sample Position** P1-A6 **Comment**  
**Injection Volume** 10  
**Acq. Date-Time** 5/26/2023 4:16:44 PM  
**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	3.896	136975	∞	12.4	143.57	1749080	4.8275 ng/ml
THC-COOH	3.969	59696	2003.14	213.0	∞	518829	14.7907 ng/ml
THC	5.120	466991	∞	24.0	∞	9987359	5.3365 ng/ml

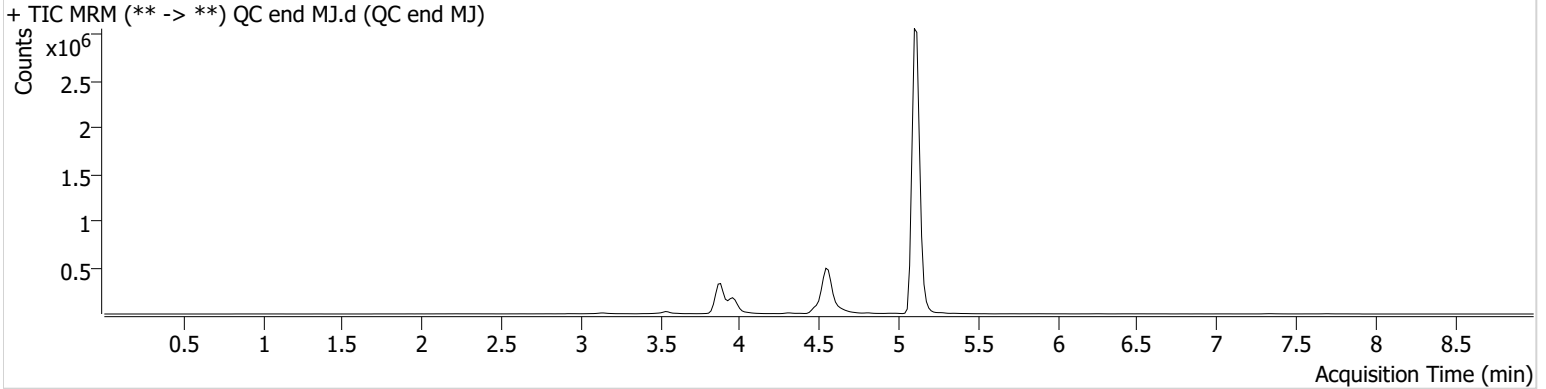


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 6/6/2023 12:04:56 PM

**Instrument** Falco (069901) **Data File** QC end MJ.d  
**Type** QC **Sample** QC end MJ  
**Acq. Method** AM 27 Agilent Method.m **Operator** Celena Shrum  
**Sample Position** P1-H5 **Comment**  
**Injection Volume** 10  
**Acq. Date-Time** 5/26/2023 9:57:23 PM  
**Sample Info.**

## Sample Chromatogram



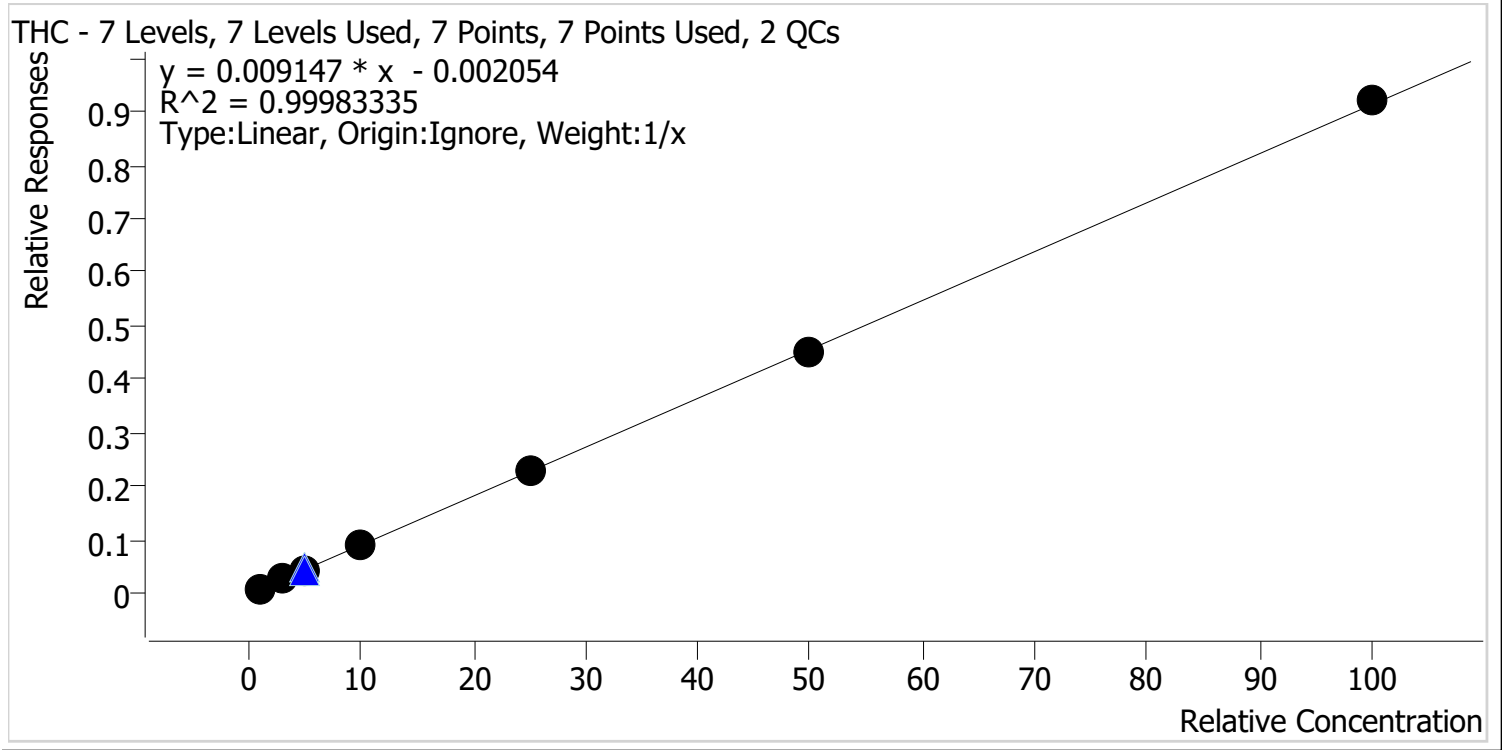
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	3.881	91207	∞	12.8	244.43	1194984	4.7107 ng/ml
THC-COOH	3.969	40476	557.44	216.9	∞	369675	14.1162 ng/ml
THC	5.120	420773	9507.60	24.9	∞	9907235	4.8678 ng/ml

cg



# AM #27 Cannabinoids Quant. Calibration Curve Report

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Last Cal. Update** 6/6/2023 12:04 PM  
**Analyst Name** ISP\Datastor  
**Analyte** THC **Internal Standard** THC-D3

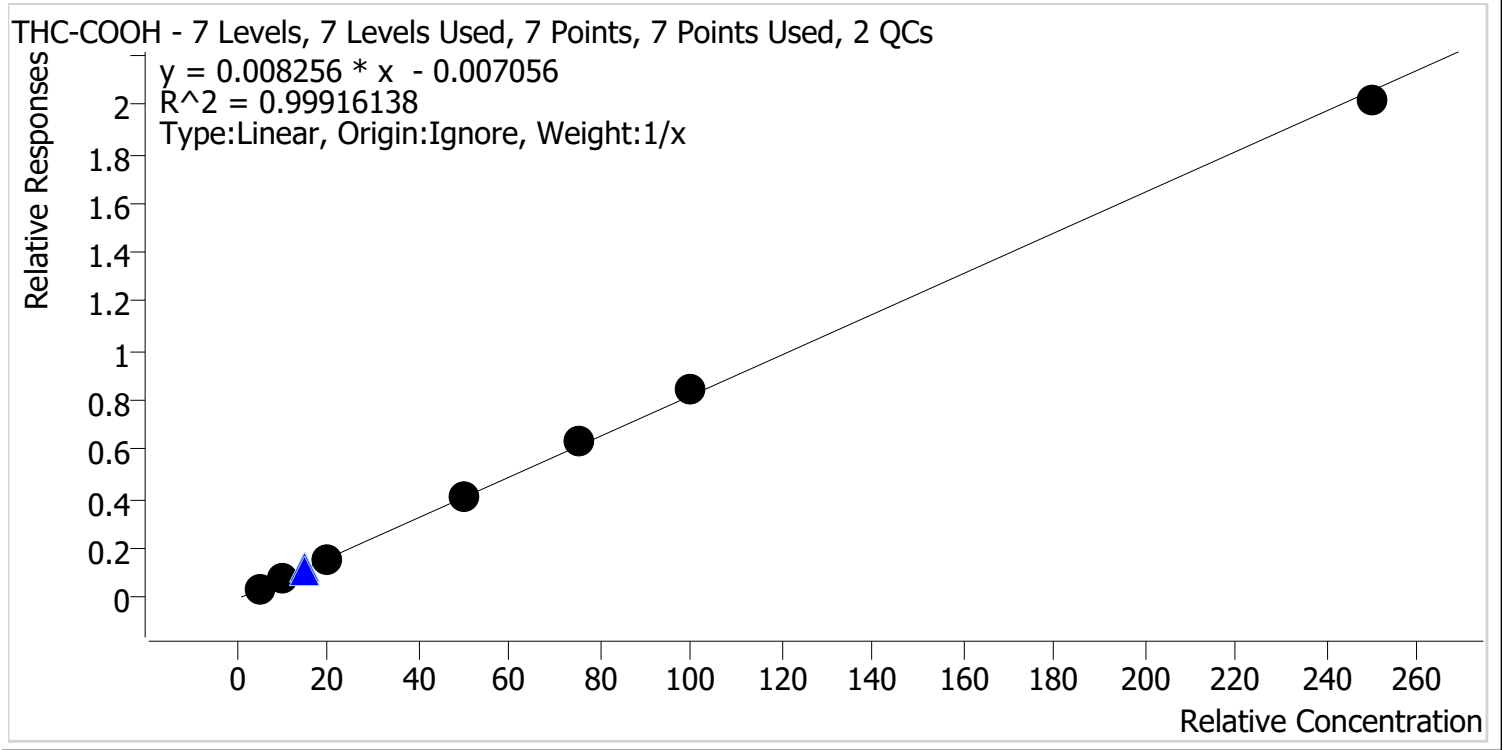


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	✓	1.0	1.1	106.9
Cal 2 MJ	2	✓	3.0	3.0	99.1
Cal 3 MJ	3	✓	5.0	4.8	95.3
Cal 4 MJ	4	✓	10.0	9.9	99.2
Cal 5 MJ	5	✓	25.0	24.9	99.6
Cal 6 MJ	6	✓	50.0	49.6	99.1
Cal 7 MJ	7	✓	100.0	100.8	100.8



# AM #27 Cannabinoids Quant. Calibration Curve Report

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Last Cal. Update** 6/6/2023 12:04 PM  
**Analyst Name** ISP\Datastor  
**Analyte** THC-COOH **Internal Standard** THC-COOH-D9



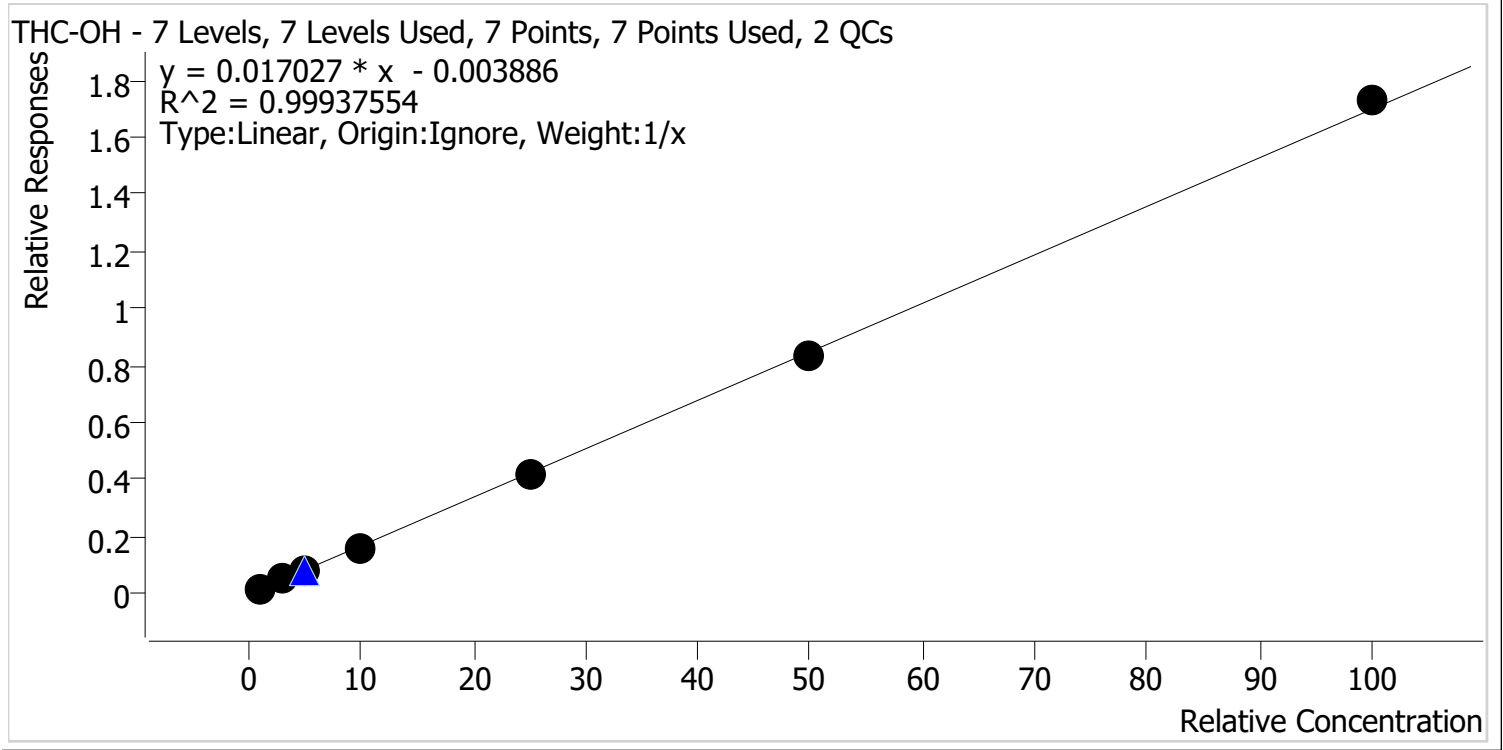
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	✓	5.0	5.0	100.3
Cal 2 MJ	2	✓	10.0	9.7	97.5
Cal 3 MJ	3	✓	20.0	19.7	98.4
Cal 4 MJ	4	✓	50.0	49.5	99.0
Cal 5 MJ	5	✓	75.0	77.4	103.2
Cal 6 MJ	6	✓	100.0	103.6	103.6
Cal 7 MJ	7	✓	250.0	245.0	98.0





# AM #27 Cannabinoids Quant. Calibration Curve Report

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Last Cal. Update** 6/6/2023 12:04 PM  
**Analyst Name** ISP\Datastor  
**Analyte** THC-OH **Internal Standard** THC-OH-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	✓	1.0	1.1	113.6
Cal 2 MJ	2	✓	3.0	2.9	97.5
Cal 3 MJ	3	✓	5.0	4.7	94.9
Cal 4 MJ	4	✓	10.0	9.5	95.2
Cal 5 MJ	5	✓	25.0	24.6	98.3
Cal 6 MJ	6	✓	50.0	49.3	98.7
Cal 7 MJ	7	✓	100.0	101.8	101.8

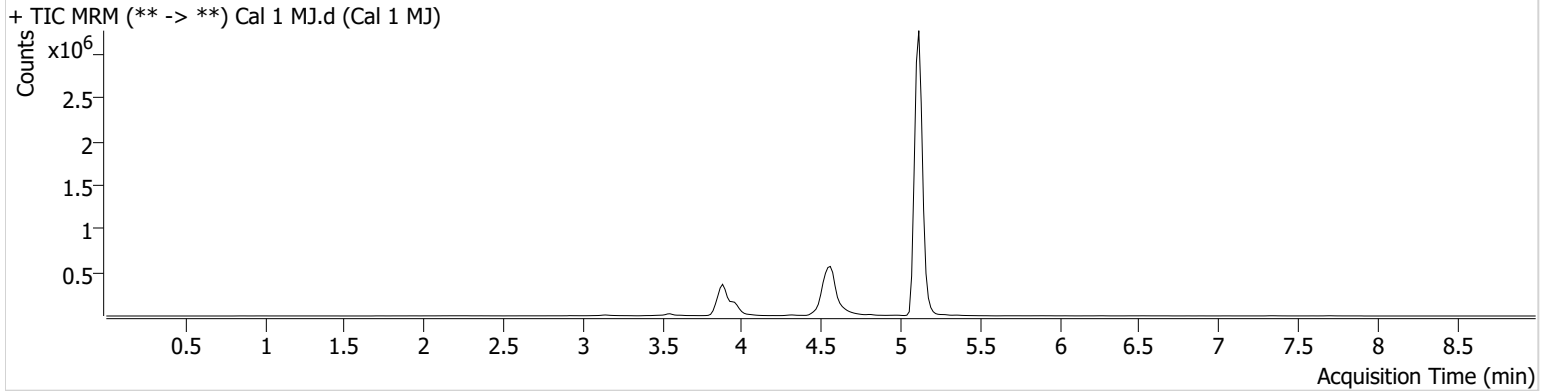


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 6/6/2023 12:04:56 PM

<b>Instrument</b>	Falco (069901)	<b>Data File</b>	Cal 1 MJ.d
<b>Type</b>	Cal	<b>Sample</b>	Cal 1 MJ
<b>Acq. Method</b>	AM 27 Agilent Method.m	<b>Operator</b>	Celena Shrum
<b>Sample Position</b>	P1-H6	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	5/26/2023 2:31:45 PM		

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	3.881	23233	∞	15.6	23.66	1503015	1.1361 ng/ml <b>Low</b>
THC-COOH	3.969	14939	75.32	242.8	244.62	434801	5.0160 ng/ml
THC	5.120	87133	∞	26.5	∞	11286302	1.0686 ng/ml

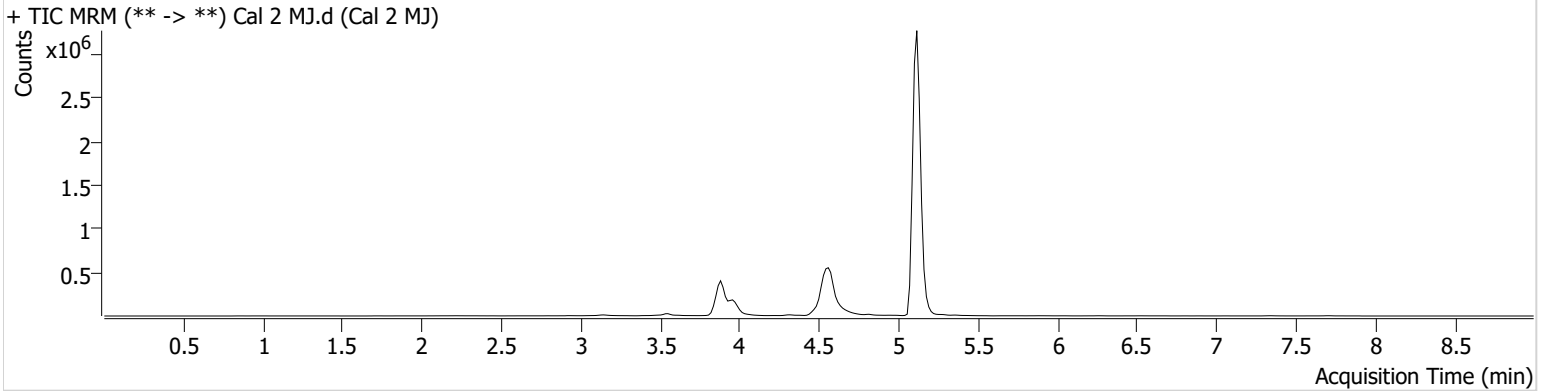


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 6/6/2023 12:04:56 PM

**Instrument** Falco (069901) **Data File** Cal 2 MJ.d  
**Type** Cal **Sample** Cal 2 MJ  
**Acq. Method** AM 27 Agilent Method.m **Operator** Celena Shrum  
**Sample Position** P1-G6 **Comment**  
**Injection Volume** 10  
**Acq. Date-Time** 5/26/2023 2:45:01 PM  
**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	3.896	69616	∞	13.1	63.30	1516118	2.9249 ng/ml <b>Low</b>
THC-COOH	3.969	31637	∞	224.8	∞	430882	9.7477 ng/ml
THC	5.120	276195	∞	24.4	∞	10985694	2.9732 ng/ml

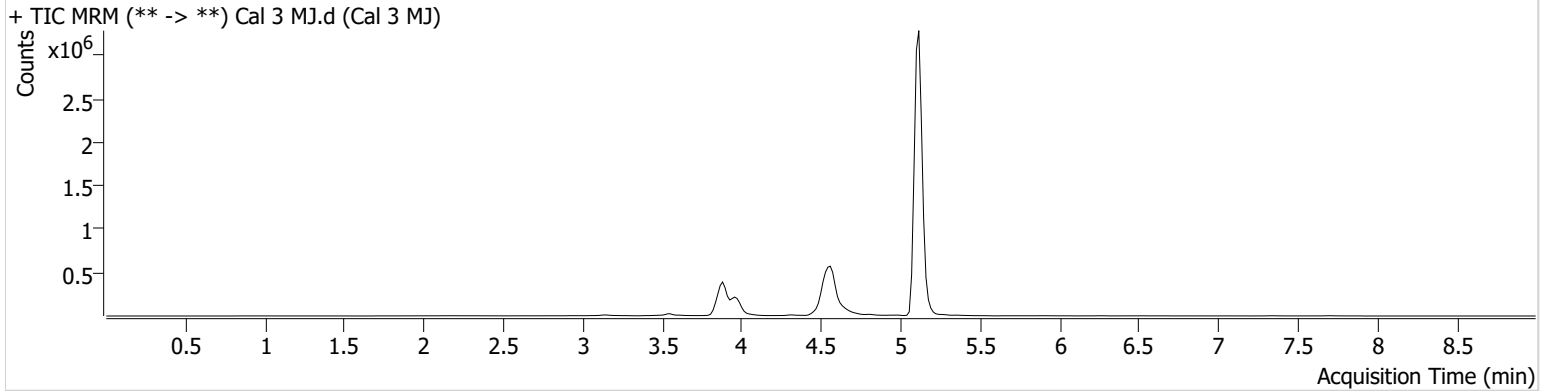


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 6/6/2023 12:04:56 PM

**Instrument** Falco (069901) **Data File** Cal 3 MJ.d  
**Type** Cal **Sample** Cal 3 MJ  
**Acq. Method** AM 27 Agilent Method.m **Operator** Celena Shrum  
**Sample Position** P1-F6 **Comment**  
**Injection Volume** 10  
**Acq. Date-Time** 5/26/2023 2:58:07 PM  
**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	3.881	117188	∞	14.9	∞	1523054	4.7470 ng/ml
THC-COOH	3.969	65816	3693.04	208.1	∞	423642	19.6718 ng/ml
THC	5.120	451053	∞	24.3	∞	10855171	4.7673 ng/ml

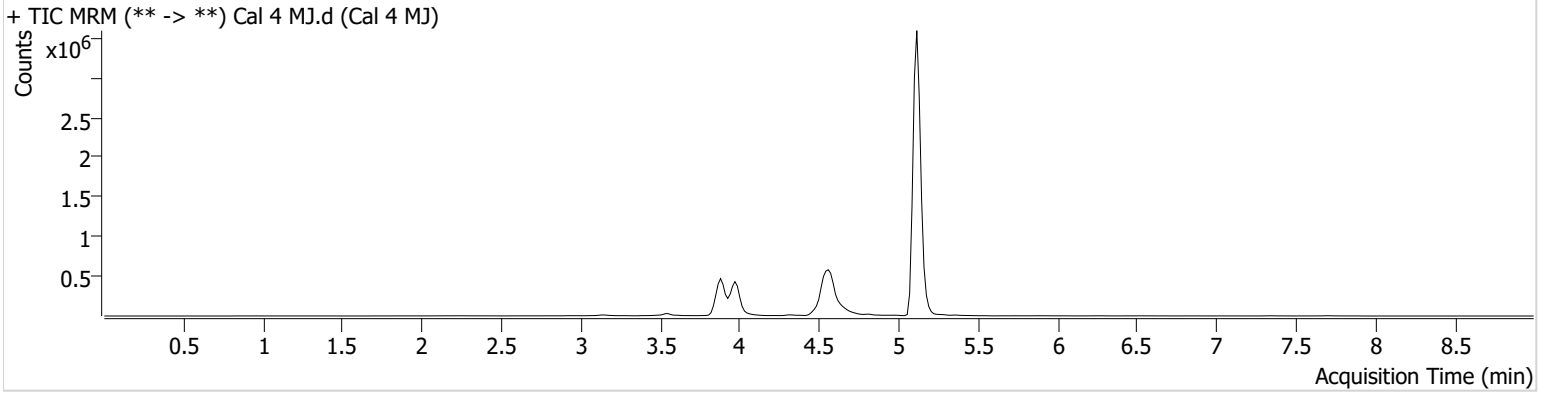


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 6/6/2023 12:04:56 PM

**Instrument** Falco (069901) **Data File** Cal 4 MJ.d  
**Type** Cal **Sample** Cal 4 MJ  
**Acq. Method** AM 27 Agilent Method.m **Operator** Celena Shrum  
**Sample Position** P1-E6 **Comment**  
**Injection Volume** 10  
**Acq. Date-Time** 5/26/2023 3:11:13 PM  
**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	3.896	253634	∞	14.1	744.02	1602813	9.5217 ng/ml
THC-COOH	3.969	185458	3392.03	211.3	∞	461848	49.4917 ng/ml
THC	5.120	966988	∞	24.2	∞	10908496	9.9159 ng/ml

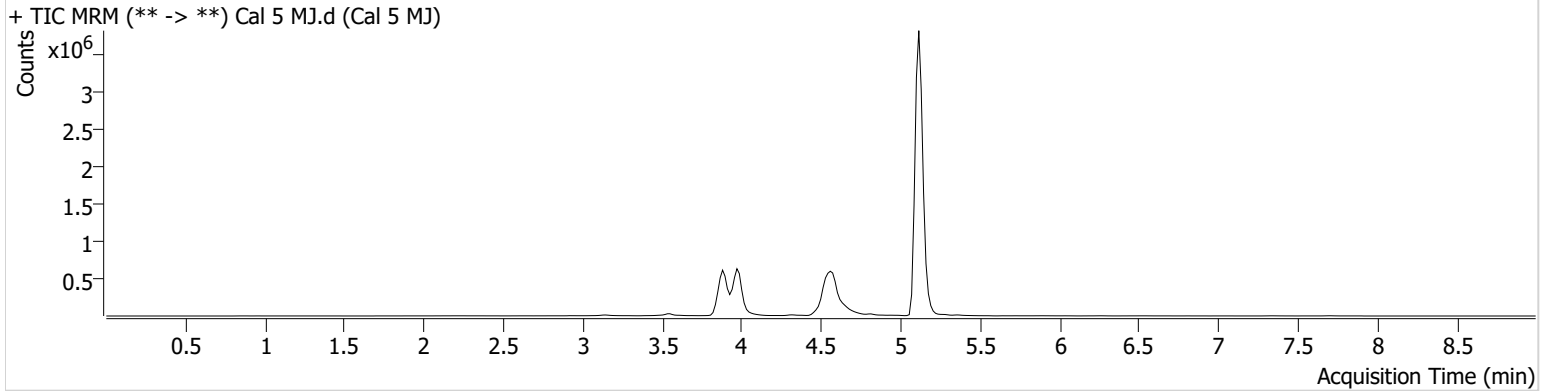


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 6/6/2023 12:04:56 PM

**Instrument** Falco (069901) **Data File** Cal 5 MJ.d  
**Type** Cal **Sample** Cal 5 MJ  
**Acq. Method** AM 27 Agilent Method.m **Operator** Celena Shrum  
**Sample Position** P1-D6 **Comment**  
**Injection Volume** 10  
**Acq. Date-Time** 5/26/2023 3:24:19 PM  
**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	3.896	701118	∞	13.7	∞	1690848	24.5805 ng/ml
THC-COOH	3.969	298319	13264.07	209.7	∞	471897	77.4239 ng/ml
THC	5.120	2305094	∞	25.2	∞	10212688	24.9006 ng/ml

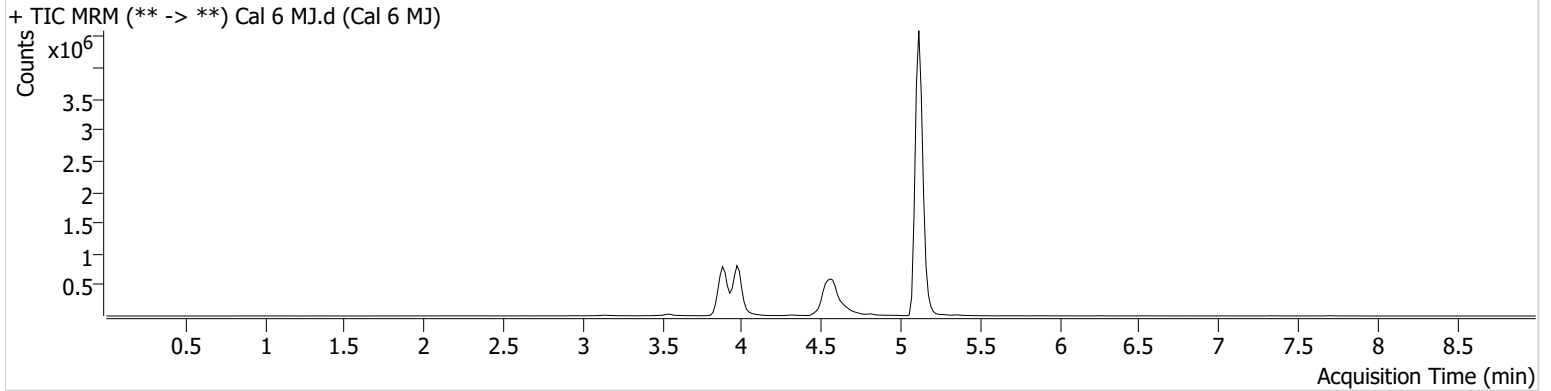
# AM #27 Cannabinoid Quant. Results



**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 6/6/2023 12:04:56 PM

<b>Instrument</b>	Falco (069901)	<b>Data File</b>	Cal 6 MJ.d
<b>Type</b>	Cal	<b>Sample</b>	Cal 6 MJ
<b>Acq. Method</b>	AM 27 Agilent Method.m	<b>Operator</b>	Celena Shrum
<b>Sample Position</b>	P1-C6	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	5/26/2023 3:37:25 PM		

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	3.881	1402714	∞	14.2	∞	1677850	49.3269 ng/ml
THC-COOH	3.969	390828	16871.57	205.7	∞	460664	103.6141 ng/ml
THC	5.120	4435180	16714.89	25.8	19797.9 3	9829222	49.5554 ng/ml

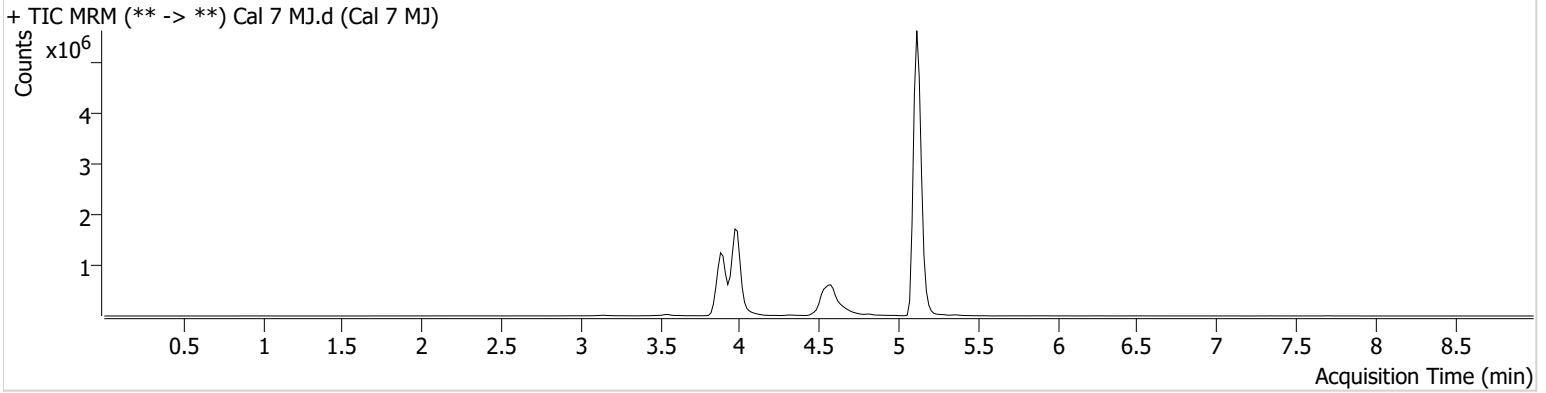


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2023\AM 27 28\052623 AM 27 28 CS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 6/6/2023 12:04:56 PM

**Instrument** Falco (069901) **Data File** Cal 7 MJ.d  
**Type** Cal **Sample** Cal 7 MJ  
**Acq. Method** AM 27 Agilent Method.m **Operator** Celena Shrum  
**Sample Position** P1-B6 **Comment**  
**Injection Volume** 10  
**Acq. Date-Time** 5/26/2023 3:50:31 PM  
**Sample Info.**

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	3.896	3064323	∞	13.9	∞	1772447	101.7629 ng/ml
THC-COOH	3.969	938070	∞	211.5	∞	465314	245.0347 ng/ml
THC	5.120	8252514	∞	26.0	∞	8968878	100.8191 ng/ml