

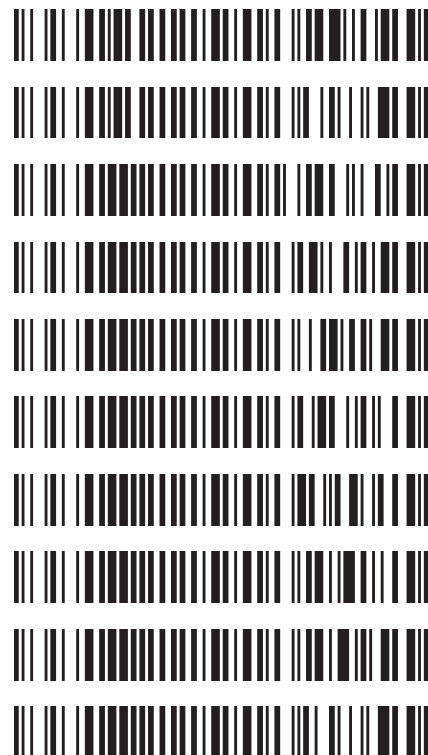
REVIEWED
By Tamara Salazar at 11:59 am, Feb 07, 2022

2/4/2022

CS

Worklist: 5562

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
M2022-0232	2	BCK	AM 27 Blood THC Quant by LC-QQQ
M2022-0298	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-3865	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-4151	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-4225	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2022-0092	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2022-0156	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2022-0161	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2022-0163	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2022-0194	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: 02/02/2022

Analyst: Celena Shrum

Plate lot#: 211018

Plate Retest Date: 04/18/2022

Mobile phase A: 0.1% Formic Acid in LCMS Water

Mobile phase B: 0.1% Formic acid in Acetonitrile

Blank Blood Lot: Lampire 20L20725

Column: UCT Selectra DA 100 x 2.1mm 3um

LCMS-QQQ ID: 069901

Blank Urine Lot: POC031319

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. 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** 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)**
- 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 r² 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.
- 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: THC-OH not evaluated due to accuracy and ratio issues.

Two urine samples (P2021-4247-2 and P2022-0015-2) from a previous batch* were also included in this run. Urine samples are being evaluated for carboxy-THC only.

*Previous batch was Worklist 5547_{CS} And also worklist 5614 for P2022-0015-2. _{CS 2/17/22}

	1	2	3	4	5	6
a	cal 1ng	QC 2	P2022-0156-1			
b	cal 3 ng	Blood NEG	P2022-0161-1			
c	cal 5 ng	M2022-0232-2	P2022-0163-1			
d	cal 10ng	M2022-0298-2	P2022-0194-1			
e	cal 25 ng	P2021-3865-1	Urine NEG			
f	cal 50 ng	P2021-4151-1	P2021-4247-2			
g	cal 100 ng	P2021-4225-1	P2022-0015-2			
h	QC 1	P2022-0092-1				

AM #27 Cannabinoid Quant. Results

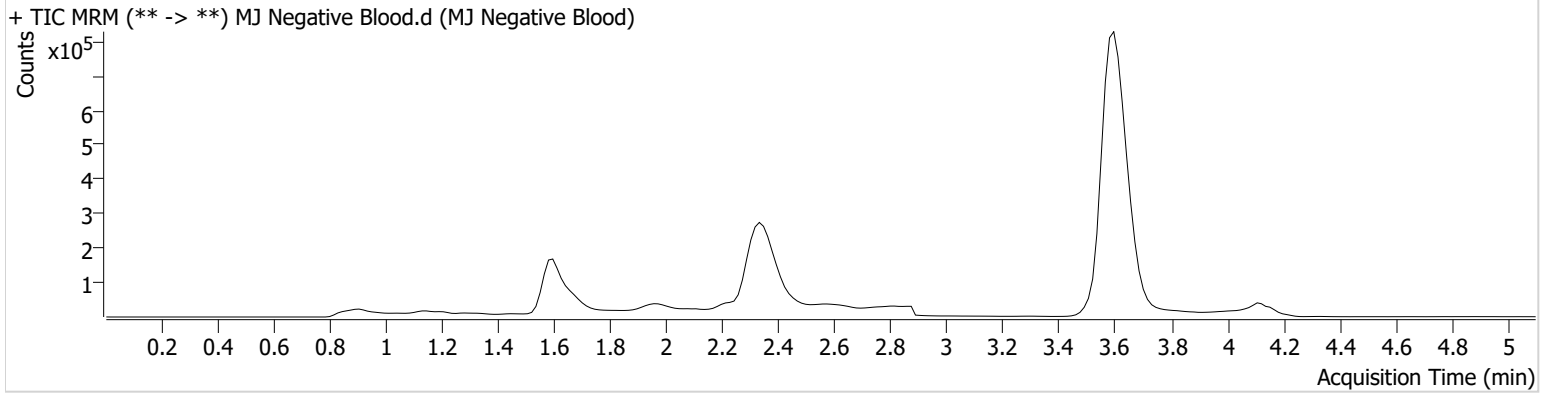


Batch results D:\MassHunter\Data\2022\AM 27-28\2-2-22 AM 27 28 CS\QuantResults\AM 27 THC and Carboxy-THC Only.batch.bin

Calibration Last Update 2/4/2022 12:06:51 PM

Instrument	Falco (069901)	Data File	MJ Negative Blood.d
Type	Sample	Sample	MJ Negative Blood
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-B2	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 4:43:48 PM		
Sample Info.			

Sample Chromatogram



AM #27 Cannabinoid Quant. Results

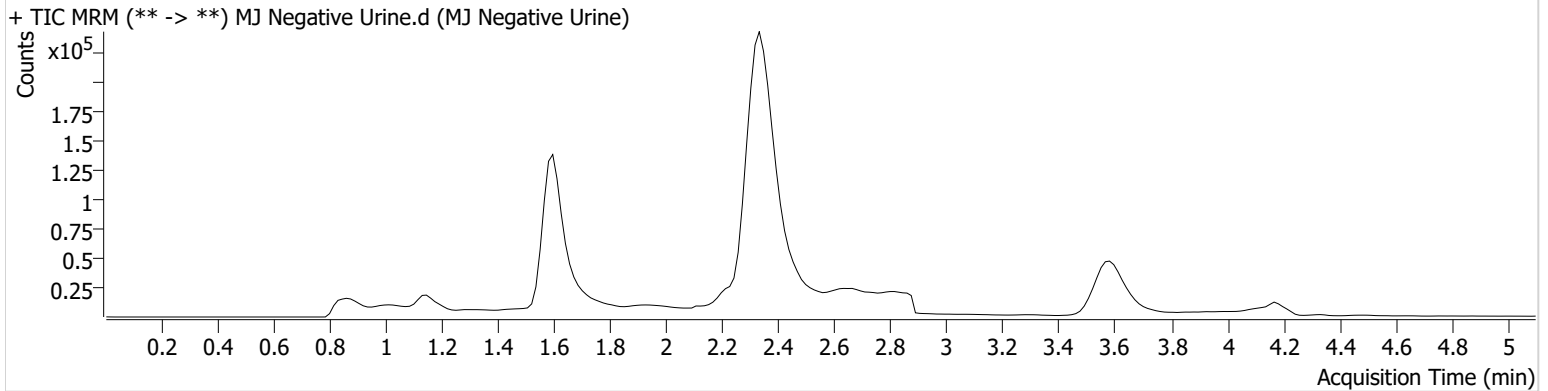


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Calibration Last Update 2/4/2022 12:06:51 PM

Instrument	Falco (069901)	Data File	MJ Negative Urine.d
Type	Sample	Sample	MJ Negative Urine
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-E3	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 7:31:20 PM		
Sample Info.			

Sample Chromatogram



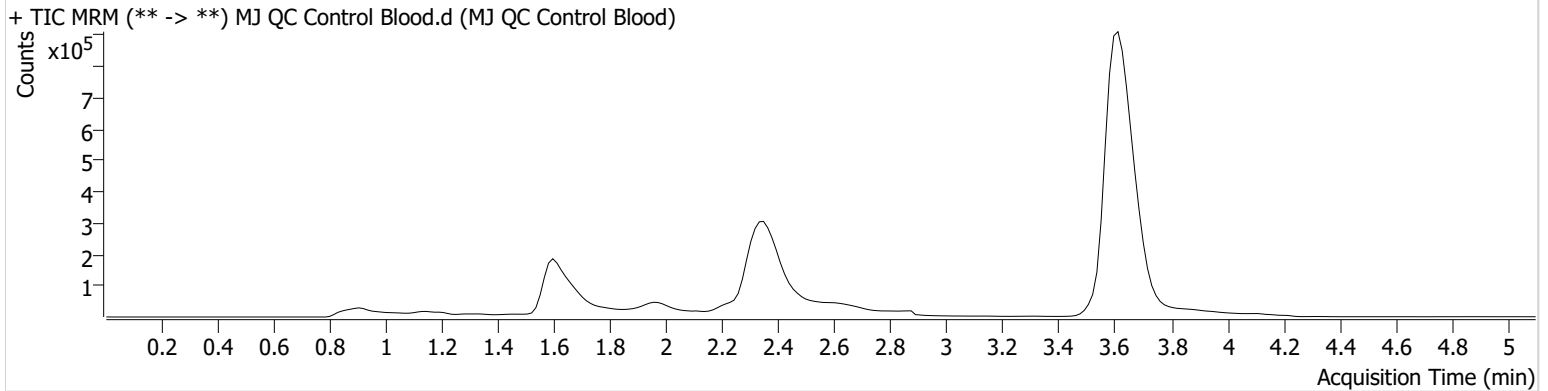
AM #27 Cannabinoid Quant. Results



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Calibration Last Update 2/4/2022 12:06:51 PM

Instrument	Falco (069901)	Data File	MJ QC Control Blood.d
Type	QC	Sample	MJ QC Control Blood
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-H1	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 4:28:34 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.640	48488	∞	77.6	∞	172411	13.7125 ng/ml
THC	3.631	265930	∞	26.8	∞	6095102	4.6876 ng/ml

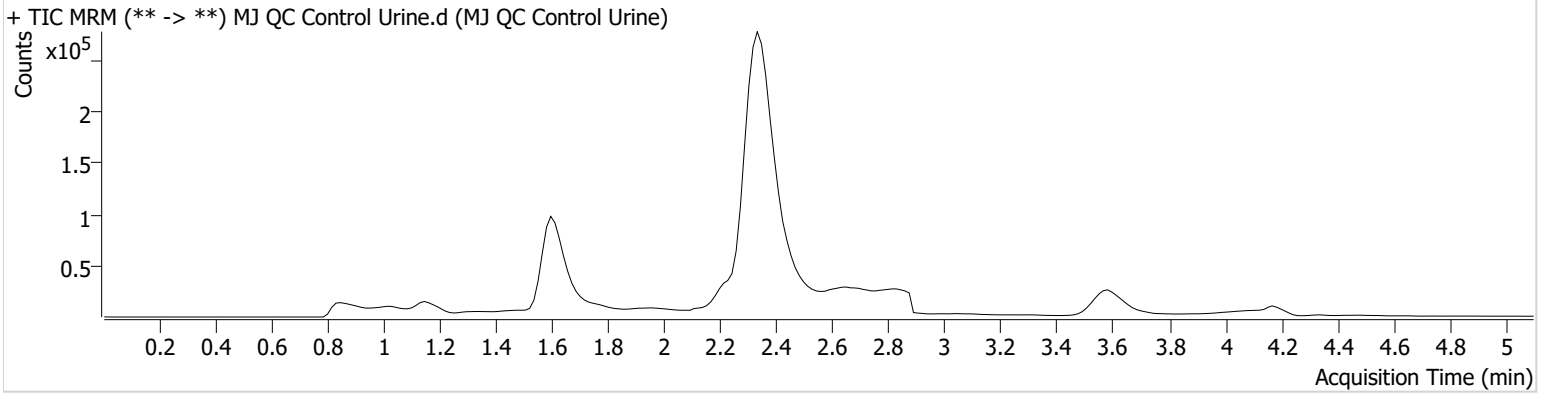
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2022\AM 27-28\2-2-22 AM 27 28 CS\QuantResults\AM 27 THC and Carboxy-THC Only.batch.bin
Calibration Last Update 2/4/2022 12:06:51 PM

Instrument	Falco (069901)	Data File	MJ QC Control Urine.d
Type	QC	Sample	MJ QC Control Urine
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-A2	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 7:38:56 PM		
Sample Info.			

Sample Chromatogram



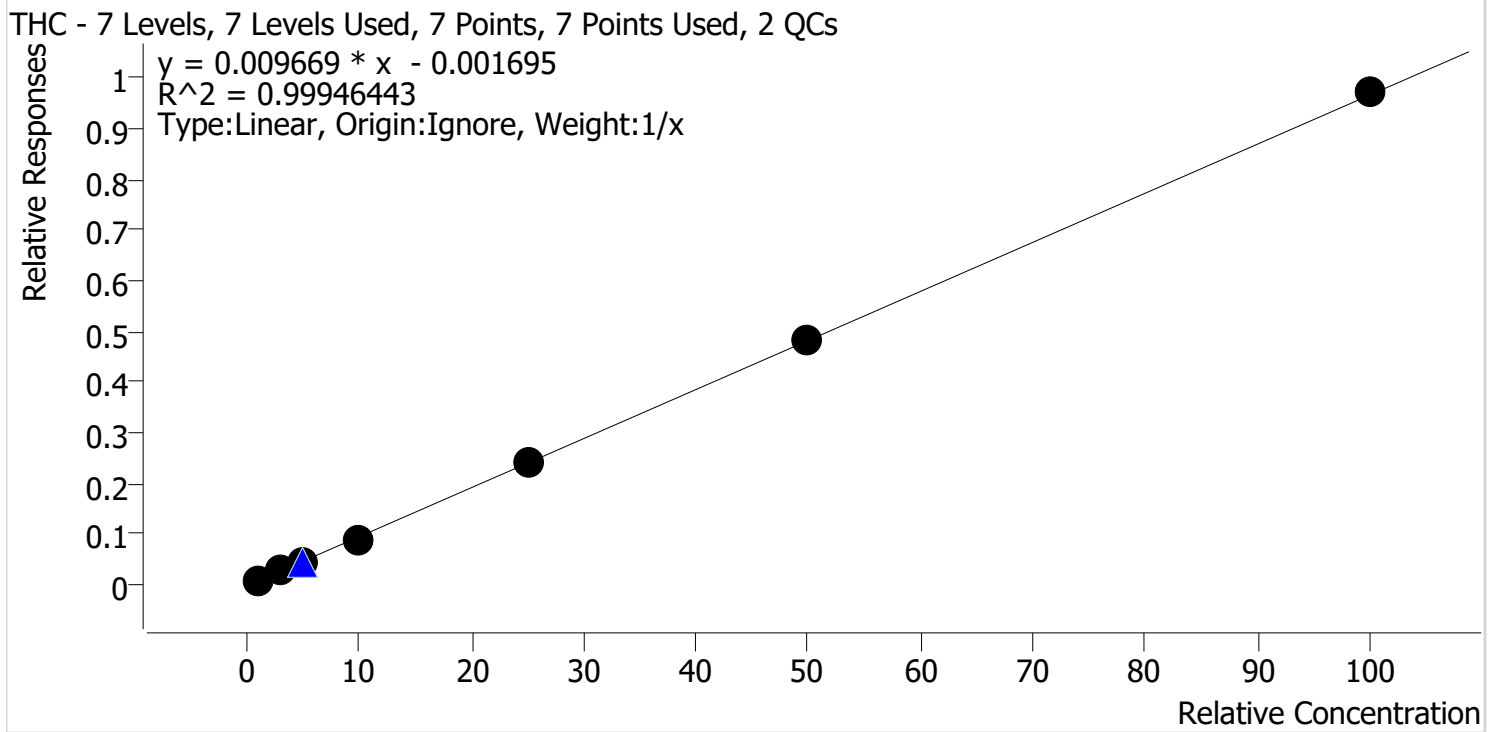
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.640	38482	∞	66.5	∞	113766	16.0749 ng/ml
THC	3.601	7632	∞	79.1 High*	∞	175348	4.6764 ng/ml

*Okay as only carboxy-THC is being evaluated for the urines for this run.



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2022\AM 27-28\2-2-22 AM 27 28 CS\QuantResults\AM 27 THC and Carboxy-THC Only.batch.bin
Last Cal. Update 2/4/2022 12:06 PM
Analyst Name ISP\datastor
Analyte THC **Internal Standard** THC-D3

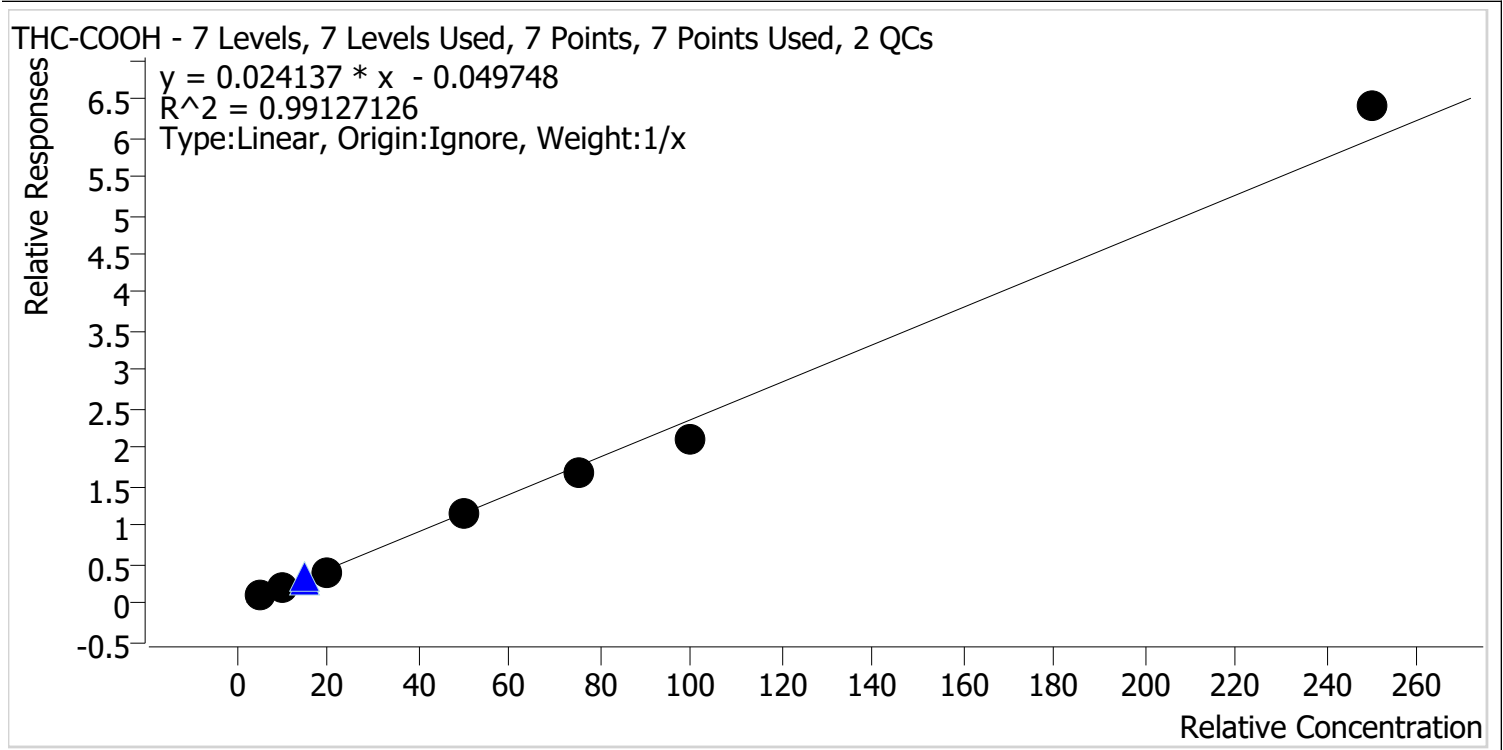


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ Cal 1	1	✓	1.0	1.1	110.9
MJ Cal 2	2	✓	3.0	2.9	96.1
MJ Cal 3	3	✓	5.0	4.9	98.9
MJ Cal 4	4	✓	10.0	9.2	92.1
MJ Cal 5	5	✓	25.0	25.4	101.6
MJ Cal 6	6	✓	50.0	49.9	99.8
MJ Cal 7	7	✓	100.0	100.5	100.5



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2022\AM 27-28\2-2-22 AM 27 28 CS\QuantResults\AM 27 THC and Carboxy-THC Only.batch.bin
Last Cal. Update 2/4/2022 12:06 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	6.5	129.6
MJ Cal 2	2	✓	10.0	9.4	93.5
MJ Cal 3	3	✓	20.0	17.6	88.0
MJ Cal 4	4	✓	50.0	49.2	98.4
MJ Cal 5	5	✓	75.0	70.7	94.3
MJ Cal 6	6	✓	100.0	89.2	89.2
MJ Cal 7	7	✓	250.0	267.4	107.0

AM #27 Cannabinoid Quant. Results

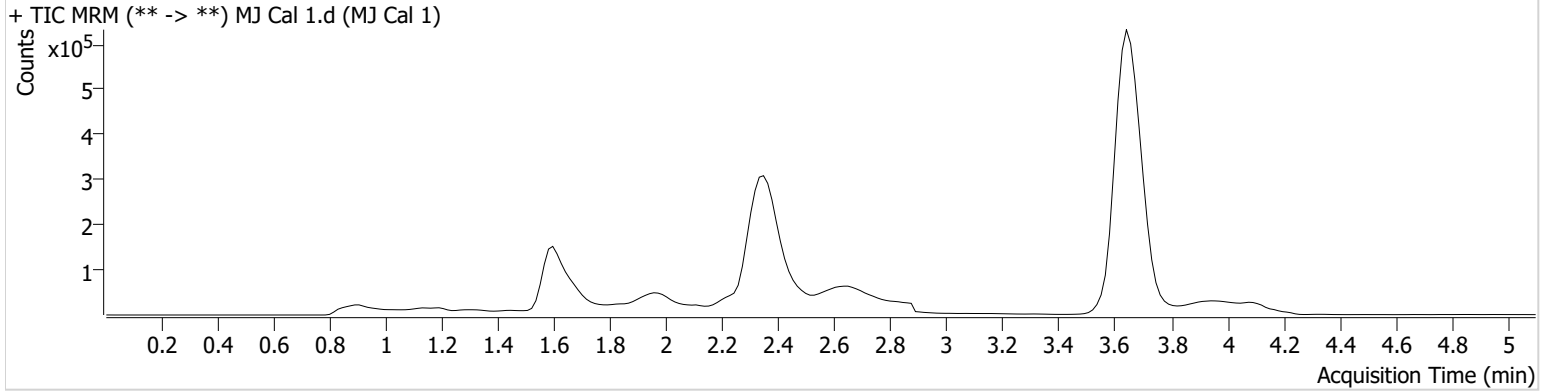


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Calibration Last Update 2/4/2022 12:06:51 PM

Instrument	Falco (069901)	Data File	MJ Cal 1.d
Type	Cal	Sample	MJ Cal 1
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-A1	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 3:35:12 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.625	16095	∞	55.8	∞	150894	6.4801 ng/ml
THC	3.661	34881	410.14	31.5	∞	3864151	1.1089 ng/ml

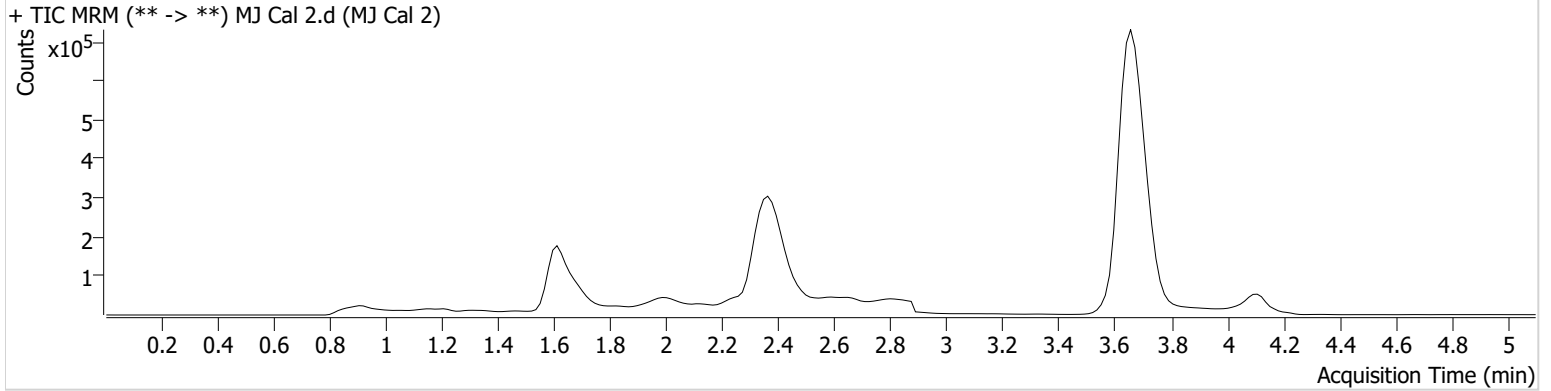
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2022\AM 27-28\2-2-22 AM 27 28 CS\QuantResults\AM 27 THC and Carboxy-THC Only.batch.bin
Calibration Last Update 2/4/2022 12:06:51 PM

Instrument	Falco (069901)	Data File	MJ Cal 2.d
Type	Cal	Sample	MJ Cal 2
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-B1	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 3:42:58 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.655	25771	∞	75.6	∞	146444	9.3518 ng/ml
THC	3.661	119048	1011.11	27.4	149.13	4546049	2.8836 ng/ml

AM #27 Cannabinoid Quant. Results

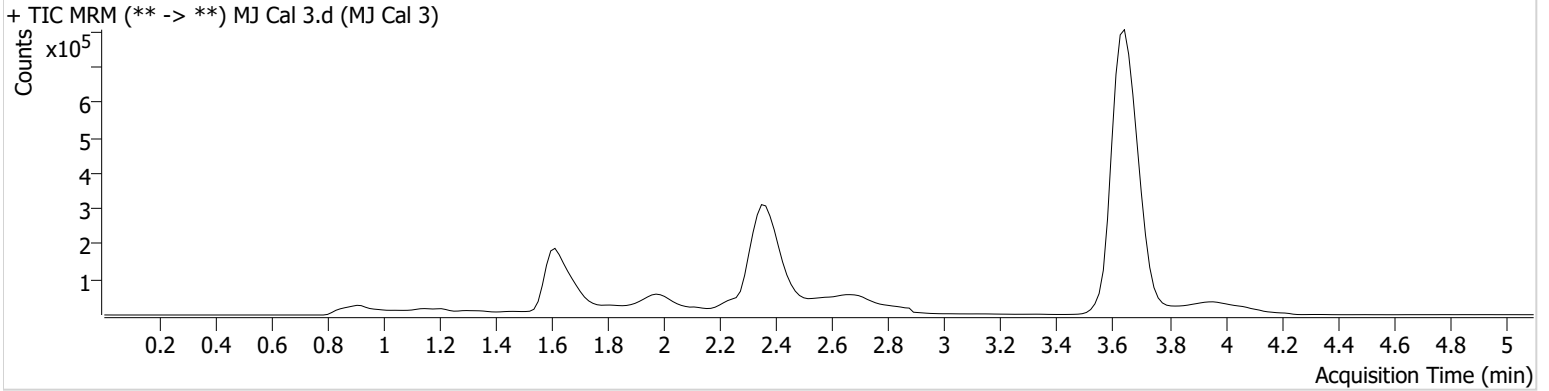


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Calibration Last Update 2/4/2022 12:06:51 PM

Instrument	Falco (069901)	Data File	MJ Cal 3.d
Type	Cal	Sample	MJ Cal 3
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-C1	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 3:50:34 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.640	56127	∞	76.6	∞	149596	17.6051 ng/ml
THC	3.661	218293	∞	26.4	∞	4733942	4.9443 ng/ml

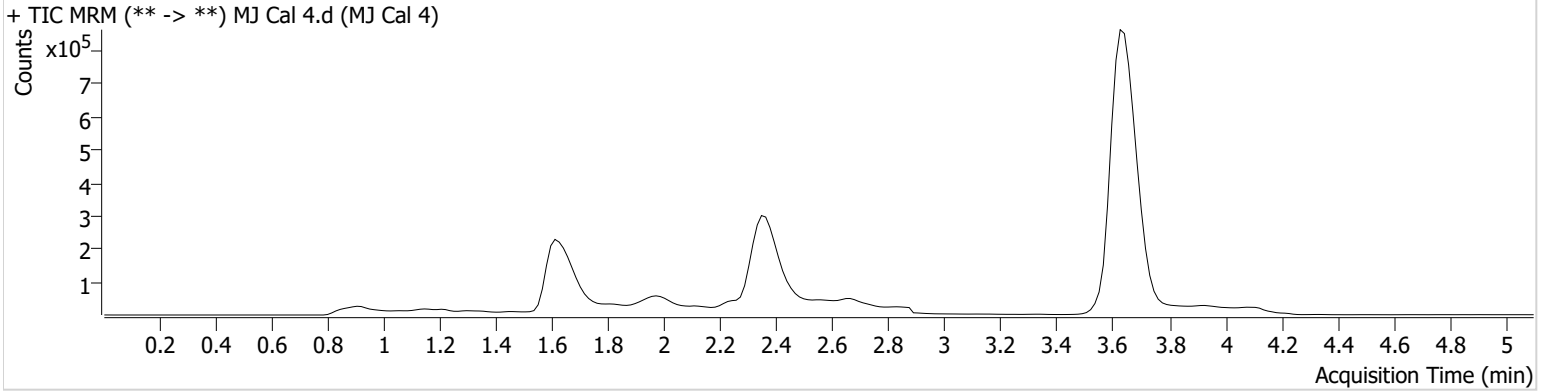
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2022\AM 27-28\2-2-22 AM 27 28 CS\QuantResults\AM 27 THC and Carboxy-THC Only.batch.bin
Calibration Last Update 2/4/2022 12:06:51 PM

Instrument	Falco (069901)	Data File	MJ Cal 4.d
Type	Cal	Sample	MJ Cal 4
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-D1	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 3:58:09 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.655	176014	∞	66.7	∞	154754	49.1828 ng/ml
THC	3.646	452090	∞	25.3	∞	5173229	9.2133 ng/ml

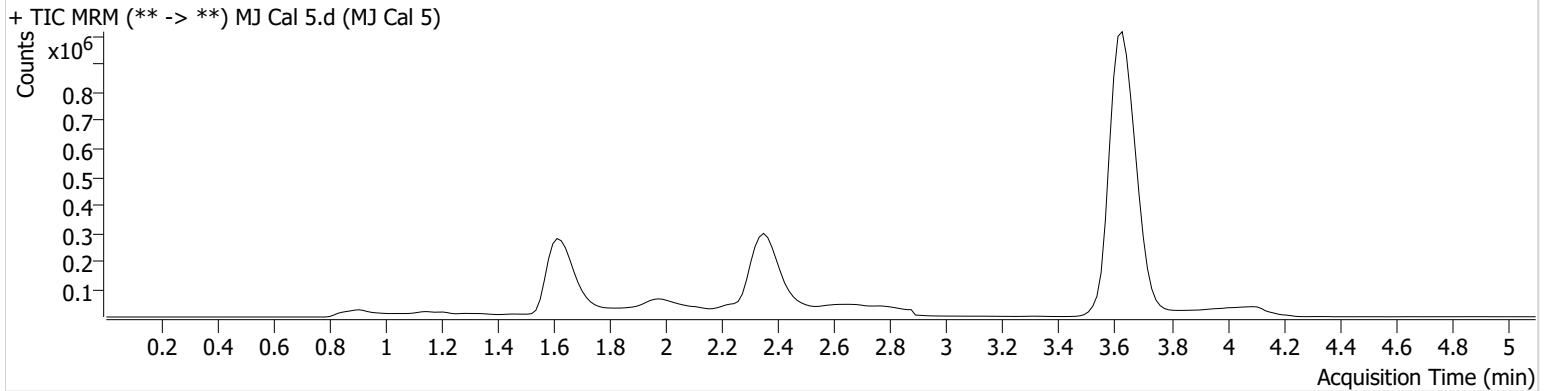
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2022\AM 27-28\2-2-22 AM 27 28 CS\QuantResults\AM 27 THC and Carboxy-THC Only.batch.bin
Calibration Last Update 2/4/2022 12:06:51 PM

Instrument	Falco (069901)	Data File	MJ Cal 5.d
Type	Cal	Sample	MJ Cal 5
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-E1	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 4:05:45 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.640	278052	∞	64.8	∞	167771	70.7241 ng/ml
THC	3.631	1216003	∞	25.3	∞	4984360	25.4065 ng/ml

AM #27 Cannabinoid Quant. Results

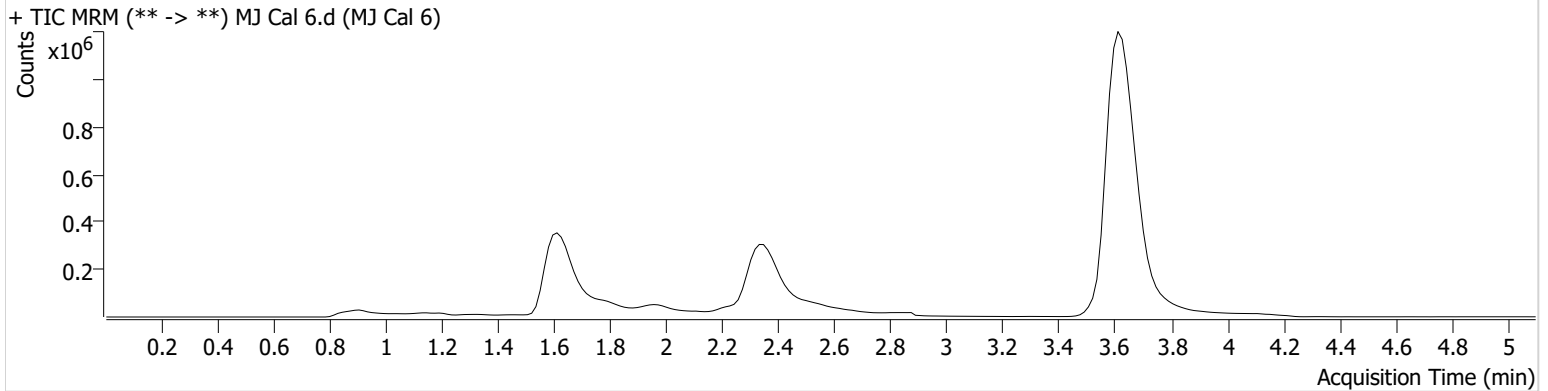


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Instrument	Falco (069901)	Data File	MJ Cal 6.d
Type	Cal	Sample	MJ Cal 6
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-F1	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 4:13:22 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.640	408670	∞	63.7	∞	194273	89.2123 ng/ml
THC	3.631	2643088	∞	25.2	∞	5496657	49.9061 ng/ml

AM #27 Cannabinoid Quant. Results

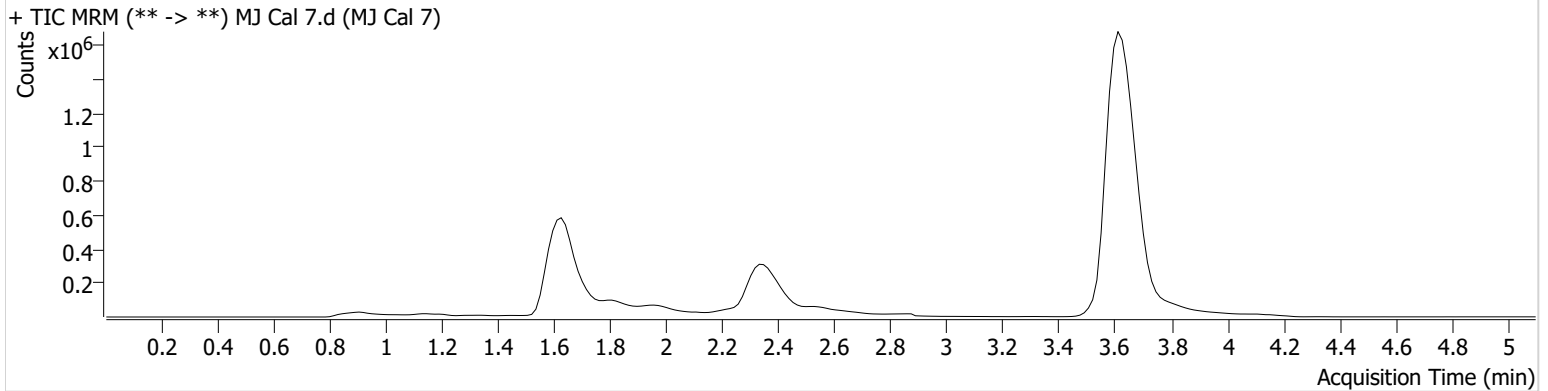


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Calibration Last Update 2/4/2022 12:06:51 PM

Instrument	Falco (069901)	Data File	MJ Cal 7.d
Type	Cal	Sample	MJ Cal 7
Acq. Method	AM 27 THCQ.m	Operator	Celena Shrum
Sample Position	P5-G1	Comment	
Injection Volume	10		
Acq. Date-Time	2/2/2022 4:20:58 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.640	981610	∞	62.6	∞	153243	267.4437 ng/ml
THC	3.631	5375854	∞	25.3	∞	5539754	100.5373 ng/ml