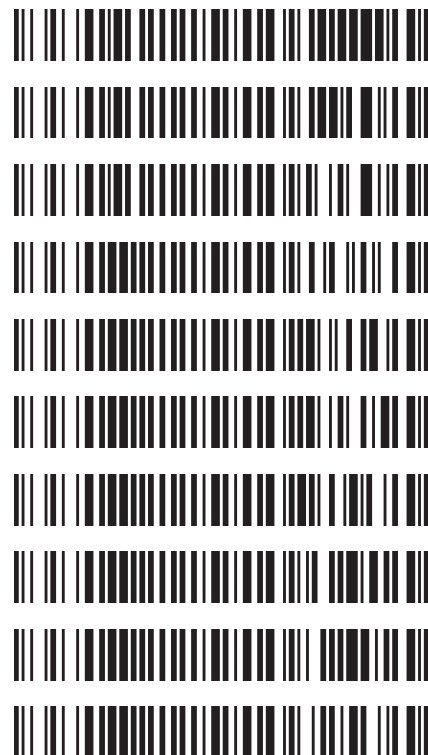


Worklist: 4513

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
M2020-3179	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-3180	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-3443	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2416	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2533	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2537	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2597	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2641	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2654	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2658	1	BCK	AM 27 Blood THC Quant by LC-QQQ



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

Extraction Date: 09/14/2020

Analyst: Celena Shrum

Plate lot#: IDP-108-2-200723

Plate Expiration: 01/23/2021

Mobile phase A: 0.1% Formic Acid in LCMS Water

Mobile phase B: 0.1% Formic acid in Acetonitrile

Blank Blood Lot: 445283-4

Column: UCT Selectra DA 100 x 2.1mm 3um

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. Using a calibrated pipette, add **1000µl blood (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** in wells of analytical plate.
- 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 6. Transfer **800µL of blood+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: 067104
- 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: 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^2 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 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 curve range: 3-100, Carboxy-THC curve range: 5-250, THC-OH not evaluated.

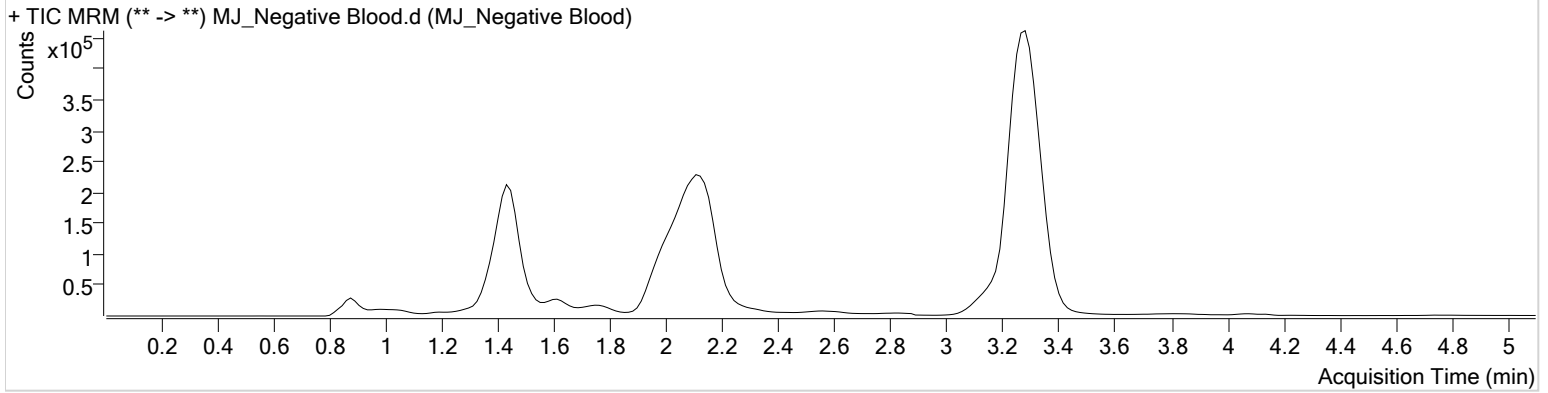
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Calibration Last Update 9/15/2020 3:04:56 PM

Instrument	Falco	Data File	MJ_Negative Blood.d
Type	Sample	Sample	MJ_Negative Blood
Acq. Method	AM 27 THC quant.m	Operator	Celena Shrum
Sample Position	P3-A2	Comment	
Injection Volume	10		
Acq. Date-Time	9/14/2020 4:17:57 PM		
Sample Info.			

Sample Chromatogram



AM #27 Cannabinoid Quant. Results

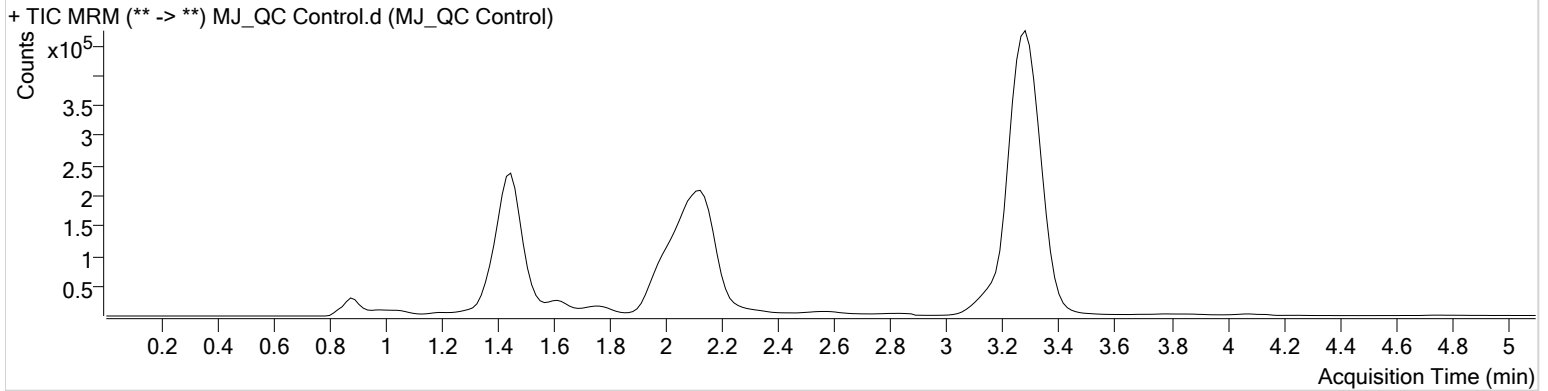


Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Calibration Last Update 9/15/2020 3:04:56 PM

Instrument	Falco	Data File	MJ_QC Control.d
Type	Sample	Sample	MJ_QC Control
Acq. Method	AM 27 THC quant.m	Operator	Celena Shrum
Sample Position	P3-H1	Comment	
Injection Volume	10		
Acq. Date-Time	9/14/2020 4:02:46 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.474	107560	∞	57.0	184.60	283387	15.7688 ng/ml
THC-OH	1.468	158227	∞	7.9	∞	918527	4.6696 ng/ml
THC	3.300	151924	∞	27.6	35.44	3766670	4.6803 ng/ml

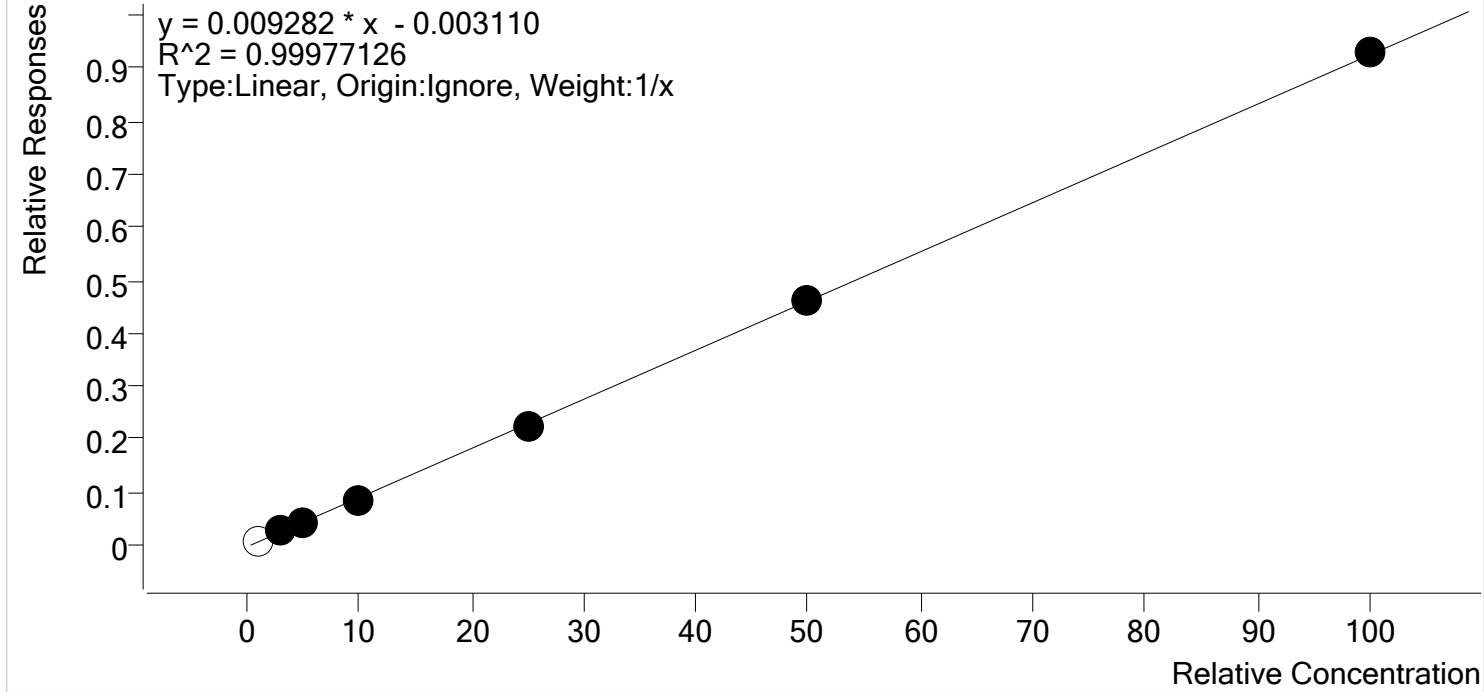
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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Last Cal. Update 9/15/2020 3:04 PM
Analyst Name ISP\datastor
Analyte THC **Internal Standard** THC-D3

THC - 7 Levels, 6 Levels Used, 7 Points, 6 Points Used, 0 QCs

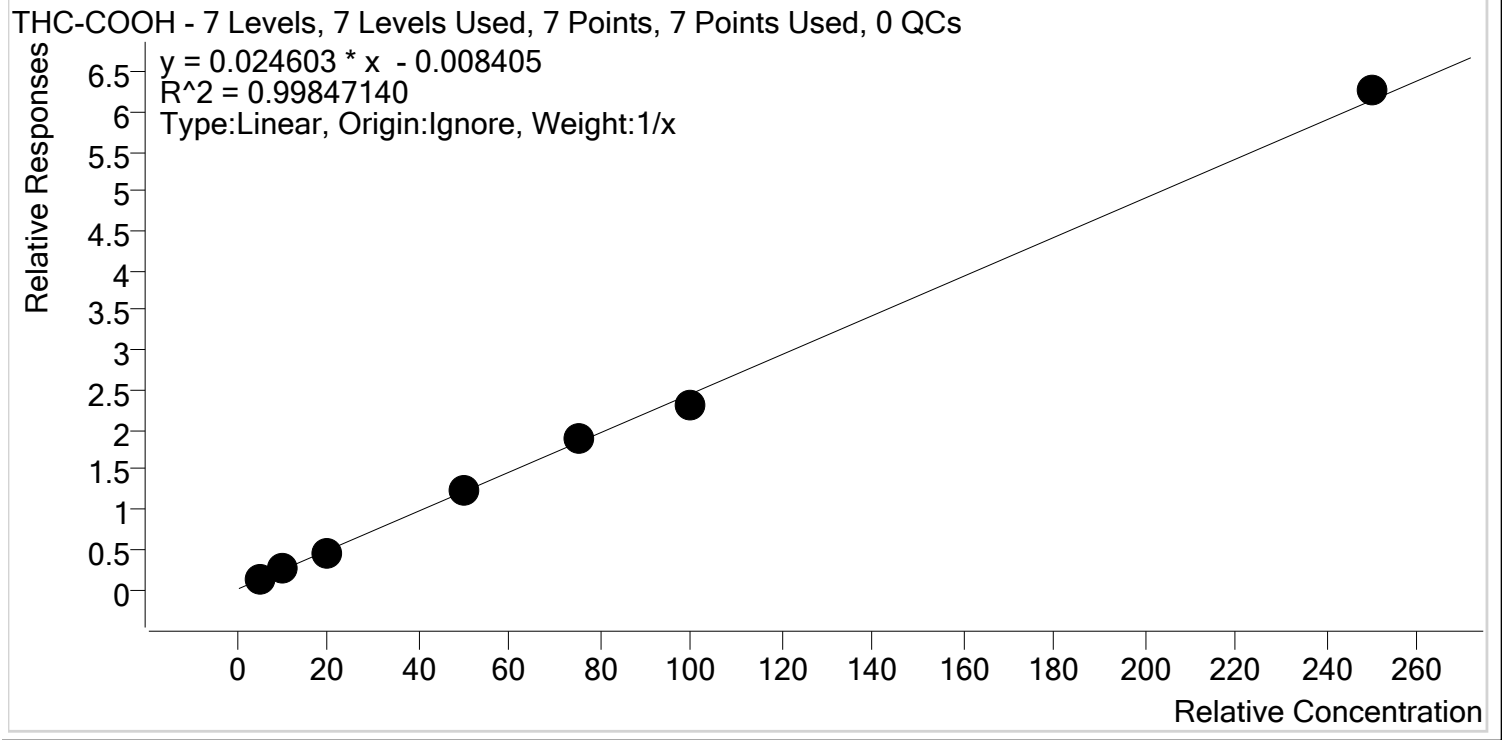


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	x	1.0	1.3	125.3
MJ_Cal 2	2	✓	3.0	3.1	103.6
MJ_Cal 3	3	✓	5.0	5.1	101.4
MJ_Cal 4	4	✓	10.0	9.6	95.6
MJ_Cal 5	5	✓	25.0	24.6	98.6
MJ_Cal 6	6	✓	50.0	50.2	100.3
MJ_Cal 7	7	✓	100.0	100.5	100.5



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Last Cal. Update 9/15/2020 3:04 PM
Analyst Name ISP\datastor
Analyte THC-COOH **Internal Standard** THC-COOH-D9

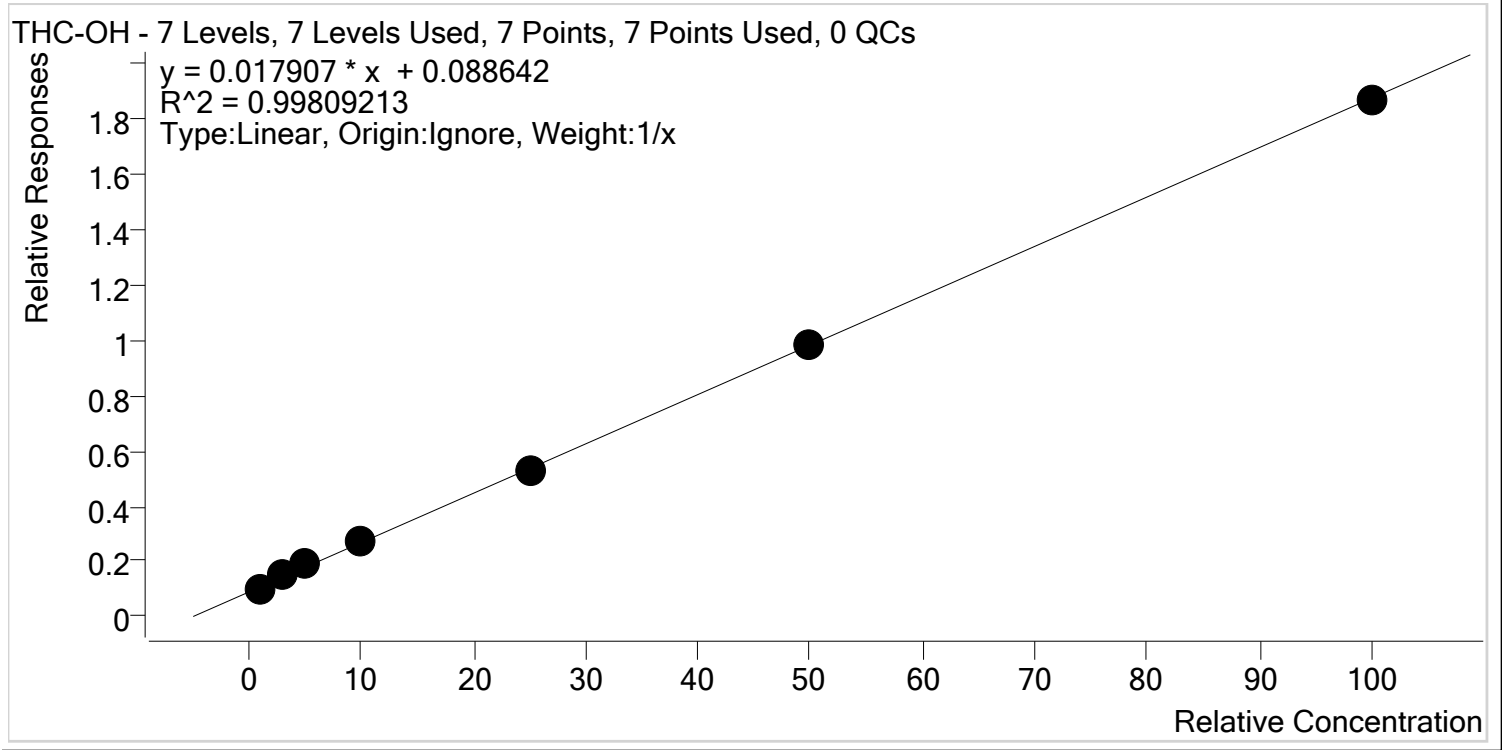


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	✓	5.0	5.4	108.0
MJ_Cal 2	2	✓	10.0	9.9	98.9
MJ_Cal 3	3	✓	20.0	18.9	94.3
MJ_Cal 4	4	✓	50.0	50.4	100.8
MJ_Cal 5	5	✓	75.0	76.5	102.1
MJ_Cal 6	6	✓	100.0	94.0	94.0
MJ_Cal 7	7	✓	250.0	254.9	102.0



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Last Cal. Update 9/15/2020 3:04 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	0.7	67.7
MJ_Cal 2	2	✓	3.0	3.6	118.9
MJ_Cal 3	3	✓	5.0	5.7	113.0
MJ_Cal 4	4	✓	10.0	10.3	103.0
MJ_Cal 5	5	✓	25.0	24.6	98.5
MJ_Cal 6	6	✓	50.0	49.7	99.4
MJ_Cal 7	7	✓	100.0	99.5	99.5

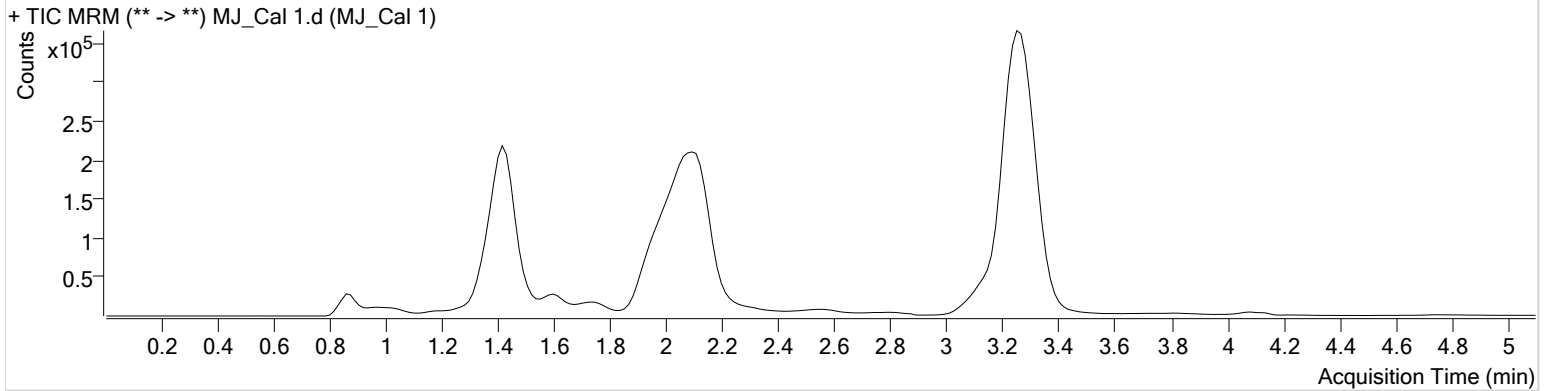


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Calibration Last Update 9/15/2020 3:04:56 PM

Instrument Falco **Data File** MJ_Cal 1.d
Type Cal **Sample** MJ_Cal 1
Acq. Method AM 27 THC quant.m **Operator** Celena Shrum
Sample Position P3-A1 **Comment**
Injection Volume 10
Acq. Date-Time 9/14/2020 3:01:53 PM
Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.444	34811	∞	54.2	65.16	279753	5.3993 ng/ml
THC-OH	1.483	99560	∞	5.4 Low	∞	988100	0.6767 ng/ml Low
THC	3.270	27127	∞	33.2 High	∞	3185097	1.2526 ng/ml Low

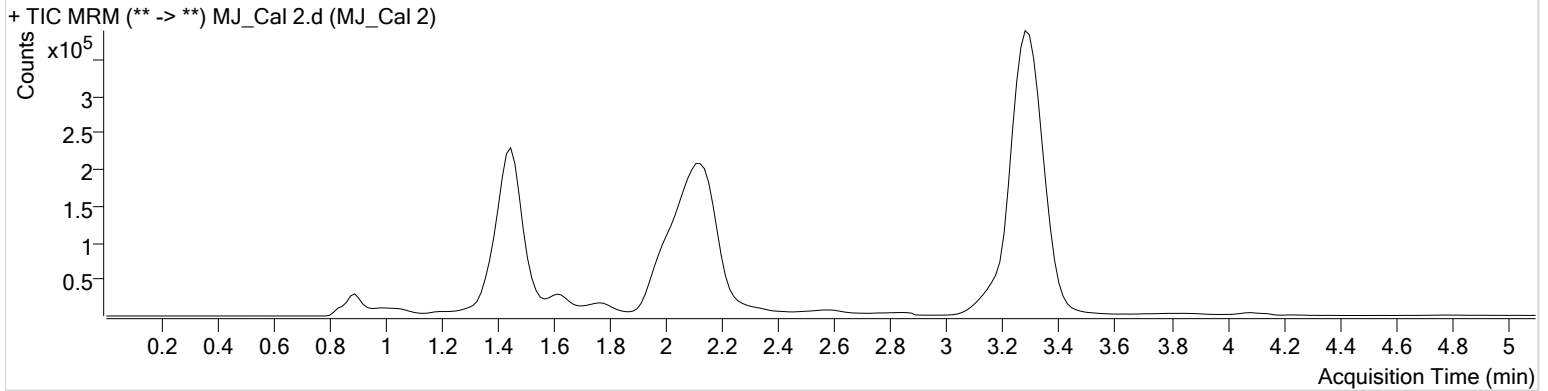
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Calibration Last Update 9/15/2020 3:04:56 PM

Instrument	Falco	Data File	MJ_Cal 2.d
Type	Cal	Sample	MJ_Cal 2
Acq. Method	AM 27 THC quant.m	Operator	Celena Shrum
Sample Position	P3-B1	Comment	
Injection Volume	10		
Acq. Date-Time	9/14/2020 3:09:38 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.474	68301	∞	55.5	∞	290630	9.8938 ng/ml
THC-OH	1.498	144459	∞	6.6 Low	∞	947023	3.5682 ng/ml
THC	3.300	82004	∞	31.8	∞	3184228	3.1095 ng/ml

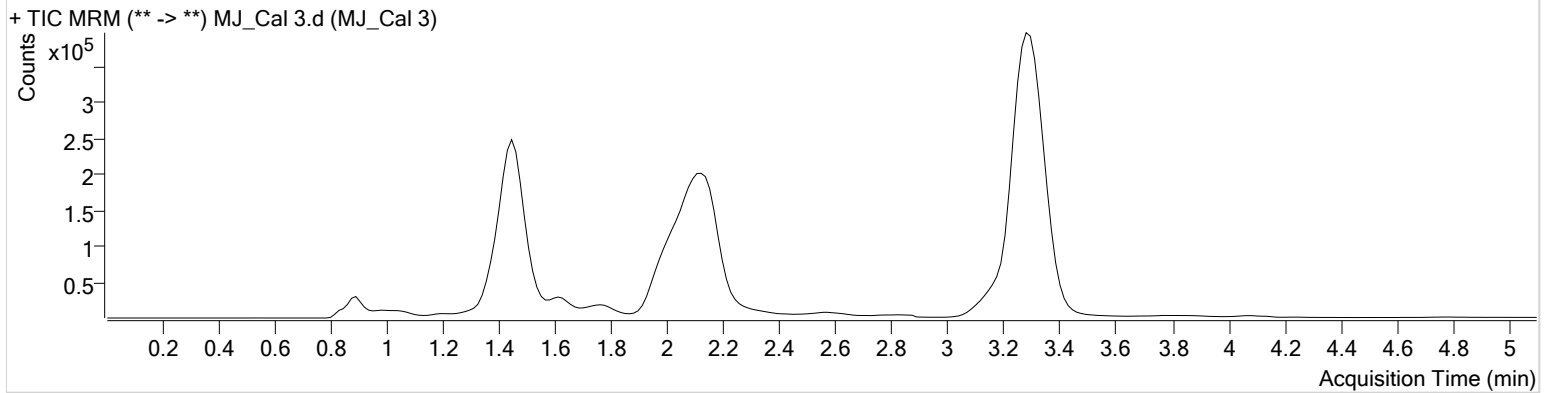
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Calibration Last Update 9/15/2020 3:04:56 PM

Instrument	Falco	Data File	MJ_Cal 3.d
Type	Cal	Sample	MJ_Cal 3
Acq. Method	AM 27 THC quant.m	Operator	Celena Shrum
Sample Position	P3-C1	Comment	
Injection Volume	10		
Acq. Date-Time	9/14/2020 3:17:12 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.474	136295	∞	59.1	∞	299164	18.8593 ng/ml
THC-OH	1.468	177482	∞	7.8	∞	934893	5.6513 ng/ml
THC	3.300	140503	∞	28.6	101.39	3195599	5.0718 ng/ml

AM #27 Cannabinoid Quant. Results

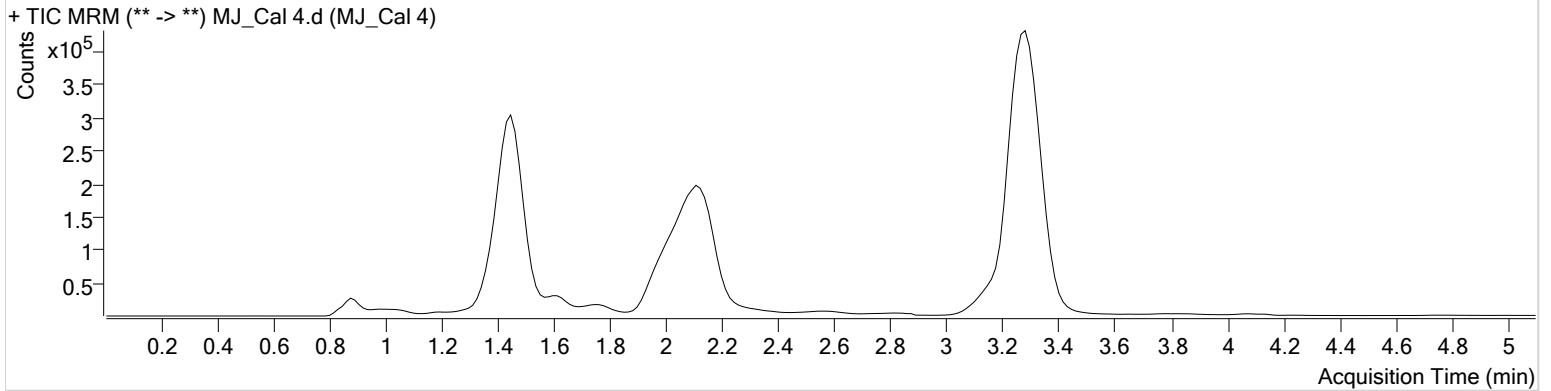


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Calibration Last Update 9/15/2020 3:04:56 PM

Instrument	Falco	Data File	MJ_Cal 4.d
Type	Cal	Sample	MJ_Cal 4
Acq. Method	AM 27 THC quant.m	Operator	Celena Shrum
Sample Position	P3-D1	Comment	
Injection Volume	10		
Acq. Date-Time	9/14/2020 3:24:48 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.474	331851	∞	60.7	∞	269563	50.3796 ng/ml
THC-OH	1.453	246201	∞	9.6	∞	901744	10.2967 ng/ml
THC	3.285	283359	∞	26.8	∞	3310729	9.5555 ng/ml

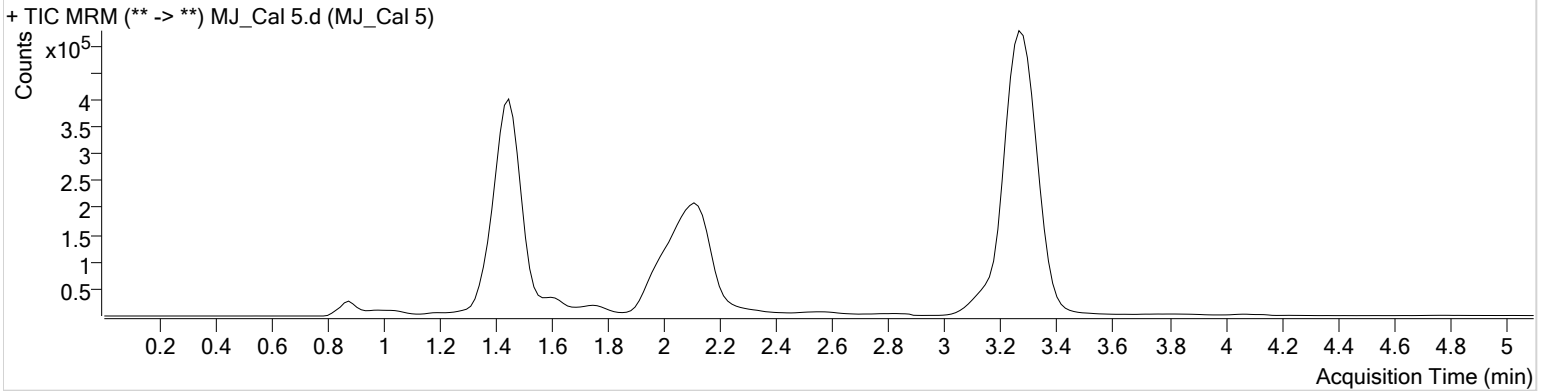
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Calibration Last Update 9/15/2020 3:04:56 PM

Instrument	Falco	Data File	MJ_Cal 5.d
Type	Cal	Sample	MJ_Cal 5
Acq. Method	AM 27 THC quant.m	Operator	Celena Shrum
Sample Position	P3-E1	Comment	
Injection Volume	10		
Acq. Date-Time	9/14/2020 3:32:25 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.474	514512	∞	61.2	2101.25	274440	76.5435 ng/ml
THC-OH	1.438 Low	491811	∞	11.3	∞	928505	24.6290 ng/ml
THC	3.285	782781	∞	25.6	∞	3469020	24.6444 ng/ml

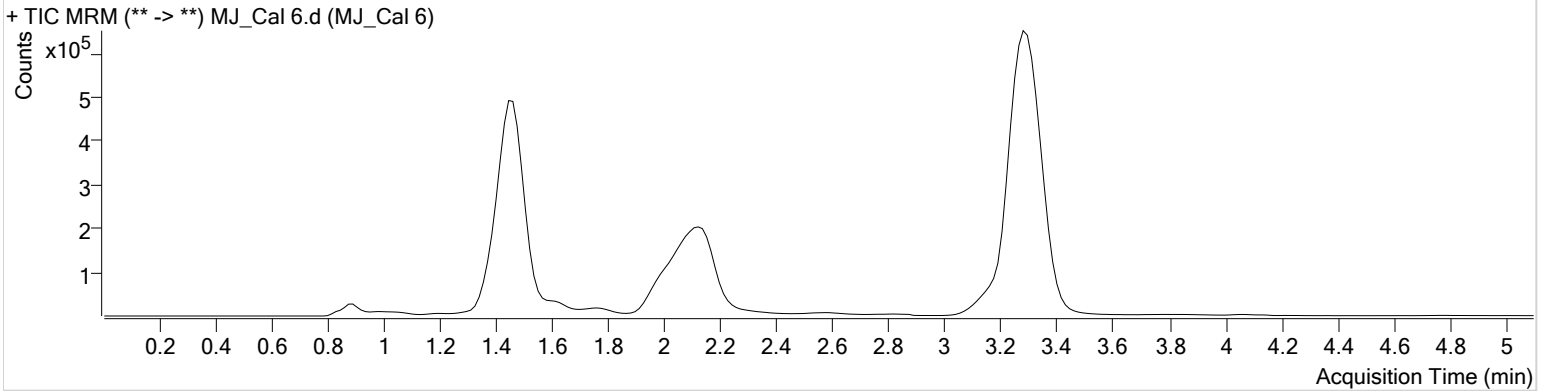
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Calibration Last Update 9/15/2020 3:04:56 PM

Instrument	Falco	Data File	MJ_Cal 6.d
Type	Cal	Sample	MJ_Cal 6
Acq. Method	AM 27 THC quant.m	Operator	Celena Shrum
Sample Position	P3-F1	Comment	
Injection Volume	10		
Acq. Date-Time	9/14/2020 3:39:59 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.474	652463	∞	60.1	∞	283199	93.9859 ng/ml
THC-OH	1.453	876126	∞	12.4 High	∞	895154	49.7061 ng/ml
THC	3.300	1608643	∞	25.6	∞	3477921	50.1637 ng/ml

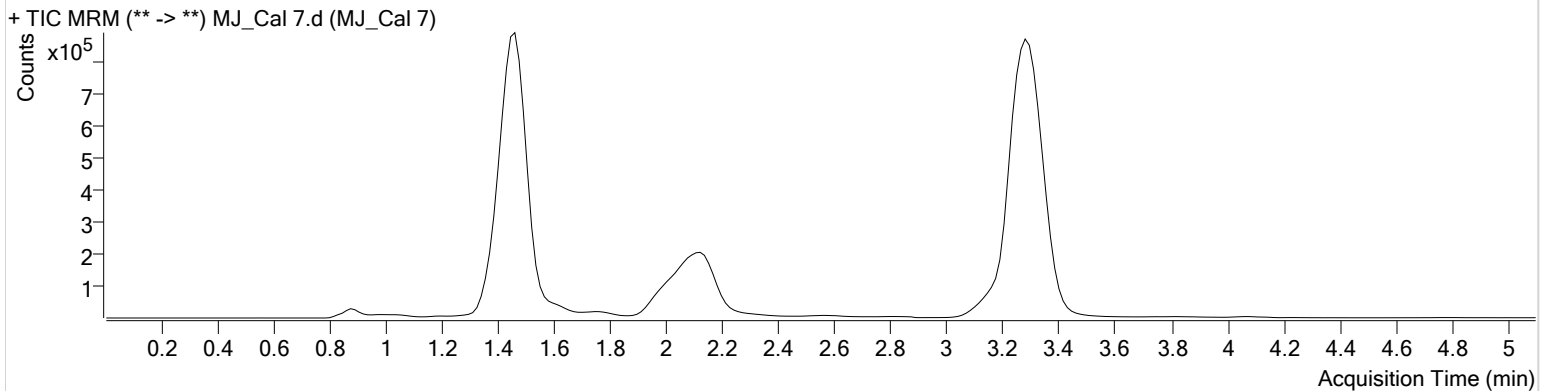
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2020\AM 27-28\AM 27-28 091420 CS\QuantResults\THCQ.batch.bin
Calibration Last Update 9/15/2020 3:04:56 PM

Instrument	Falco	Data File	MJ_Cal 7.d
Type	Cal	Sample	MJ_Cal 7
Acq. Method	AM 27 THC quant.m	Operator	Celena Shrum
Sample Position	P3-G1	Comment	
Injection Volume	10		
Acq. Date-Time	9/14/2020 3:47:34 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.474	1612512	12046.23	61.1	∞	257434	254.9386 ng/ml
THC-OH	1.438 Low	1655350	∞	13.2 High	∞	885253	99.4721 ng/ml
THC	3.285	3171861	∞	25.9	∞	3412969	100.4551 ng/ml