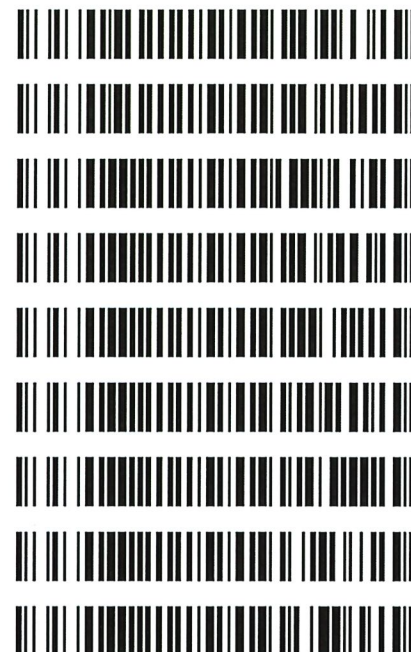


TS

REVIEWED
By Sarah Pickle at 10:11 am, Jul 24, 2020

Worklist: 4382

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
M2020-2515	3	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-2521	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1246	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1978	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1982	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2027	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2028	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2055	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2072	1	BCK	AM 27 Blood THC Quant by LC-QQQ



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

Extraction Date: 07/23/2020
Plate lot#: IDP-108-200303

Analyst: Tamara Salazar
Plate Expiration: 09/03/2020

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: Hemostat 445283-4
LCMS-QQQ ID: 069901

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.
- 3. Create worklist:

Analytic:

- 1. Remove standards, plate, controls, and samples from cold storage. Allow to reach room temperature.
- 2. Pipette **1000µL blood (calibrated pipette) Pipette ID: 42** in wells of analytical (standards) plate.
- 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 4. Pipette **500µL 0.1% formic acid in water for blood samples** 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)
- 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 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? Y / N
- 6. Enter QCs into control charting.
- 7. Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS: *Curves limited: THC-OH 3-100*

TS

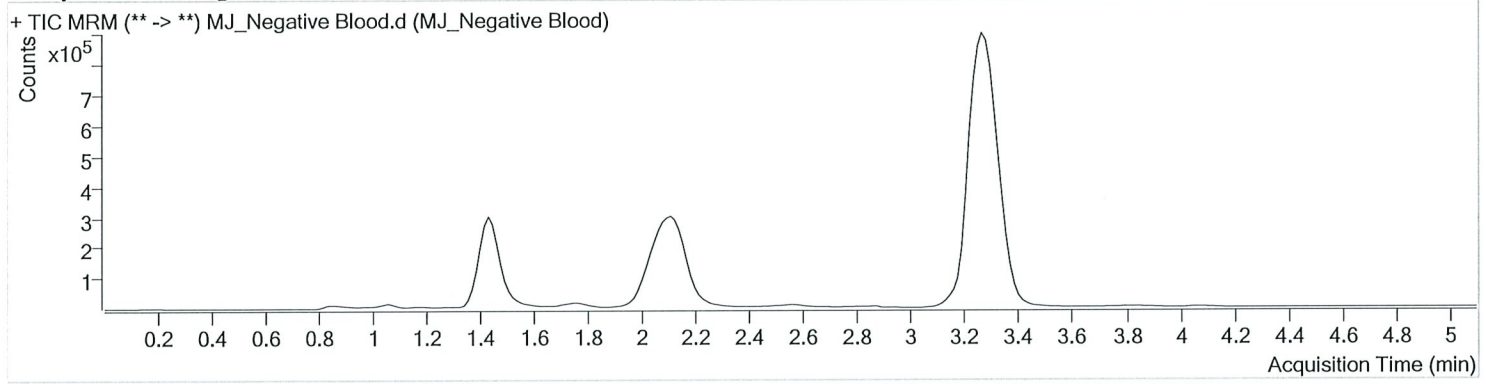


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 7/24/2020 9:16:43 AM

Instrument	Falco	Data File	MJ_Negative Blood.d
Type	Sample	Sample	MJ_Negative Blood
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P3-A2	Comment	
Injection Volume	10		
Acq. Date-Time	7/23/2020 4:58:32 PM		
Sample Info.			

Sample Chromatogram



TS

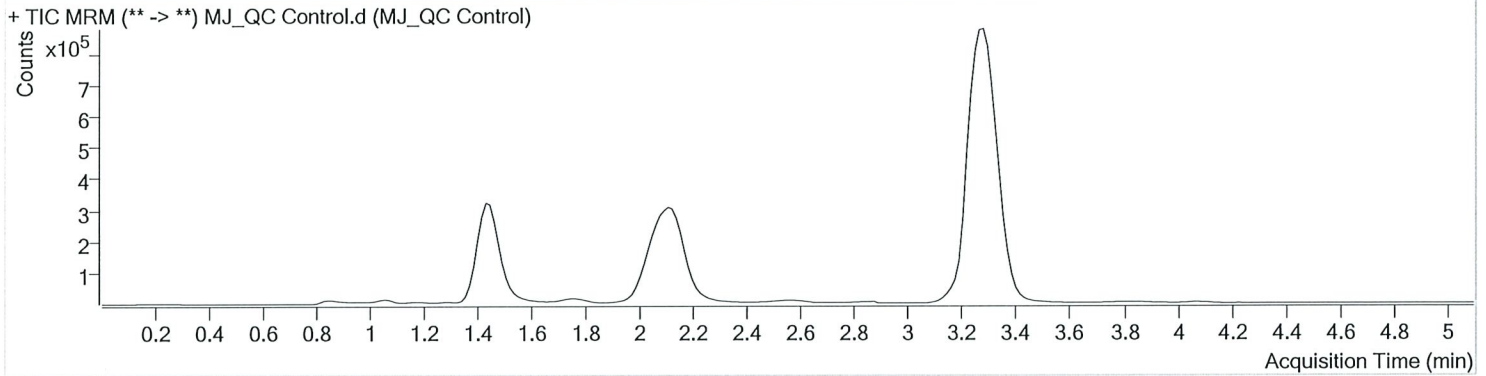


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 7/24/2020 9:16:43 AM

Instrument	Falco	Data File	MJ_QC Control.d
Type	Sample	Sample	MJ_QC Control
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P3-H1	Comment	
Injection Volume	10		
Acq. Date-Time	7/23/2020 4:43:20 PM		
Sample Info.			

Sample Chromatogram



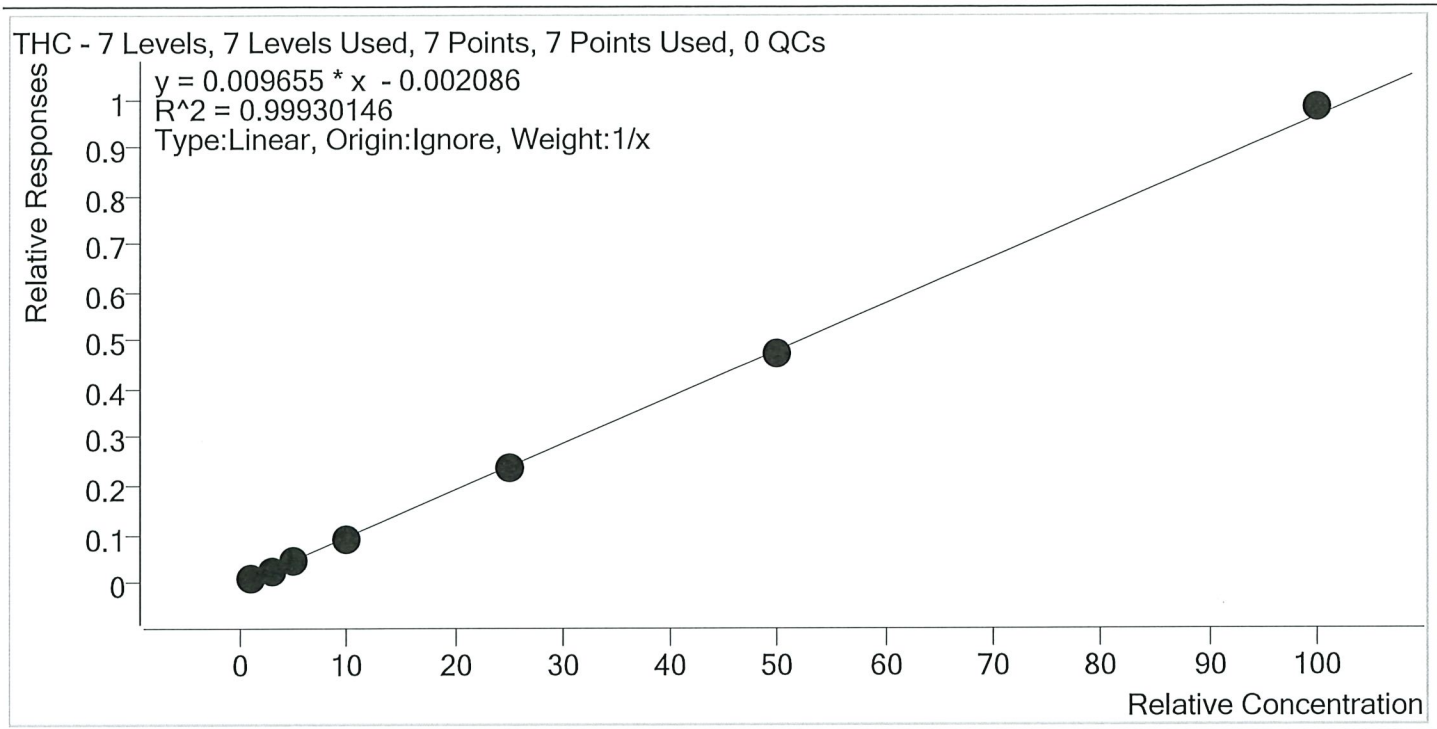
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	152344	∞	8.7	∞	1115782	4.6164 ng/ml
THC-COOH	1.474	122375	∞	57.9	∞	380190	14.4676 ng/ml
THC	3.285	256029	1325.75	28.1	226.12	6446278	4.3295 ng/ml

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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
 Last Cal. Update 7/24/2020 9:16 AM
 Analyst Name ISP\datastor
 Analyte THC Internal Standard THC-D3



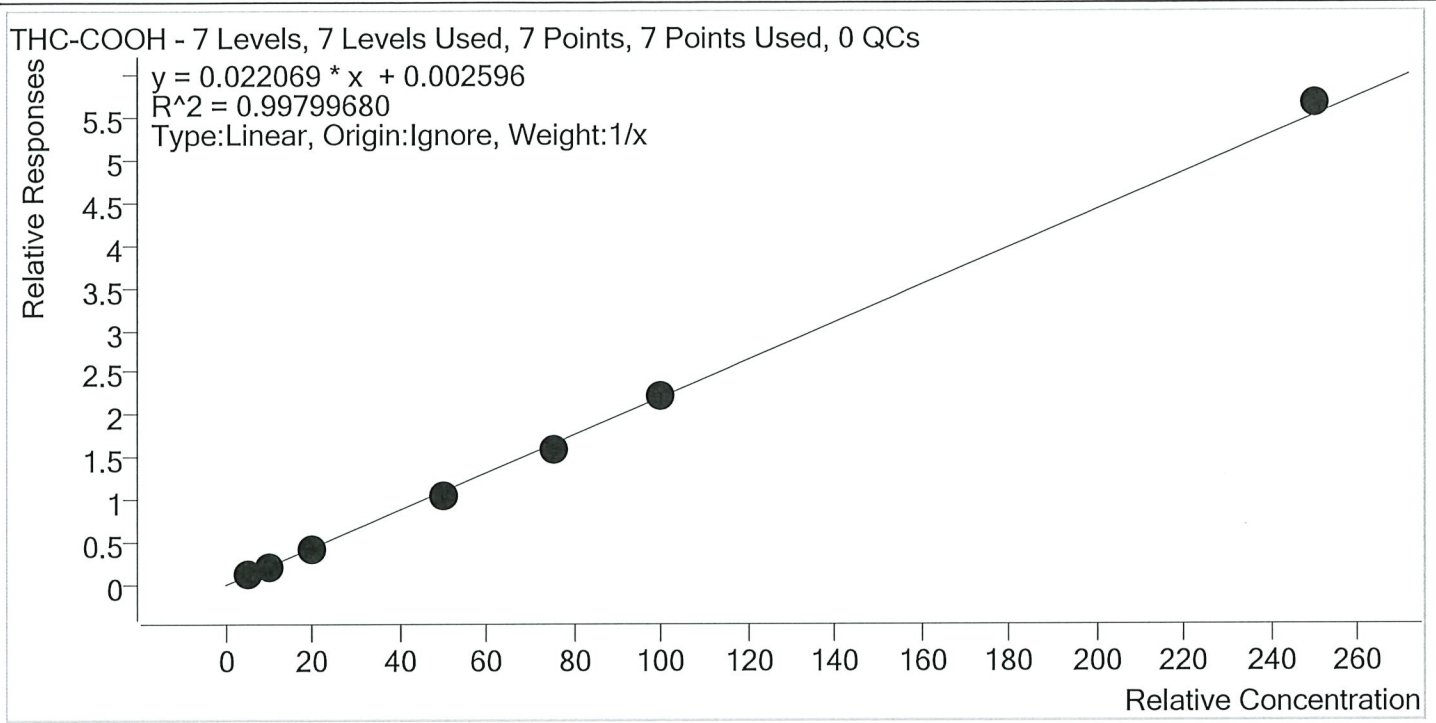
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ Cal 1	1	✓	1.0	1.1	113.9
MJ Cal 2	2	✓	3.0	2.9	96.7
MJ Cal 3	3	✓	5.0	4.8	96.3
MJ Cal 4	4	✓	10.0	9.5	94.9
MJ Cal 5	5	✓	25.0	24.4	97.6
MJ Cal 6	6	✓	50.0	49.3	98.7
MJ Cal 7	7	✓	100.0	101.9	101.9

TS



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
 Last Cal. Update 7/24/2020 9:16 AM
 Analyst Name ISP\datastor
 Analyte THC-COOH Internal Standard THC-COOH-D9



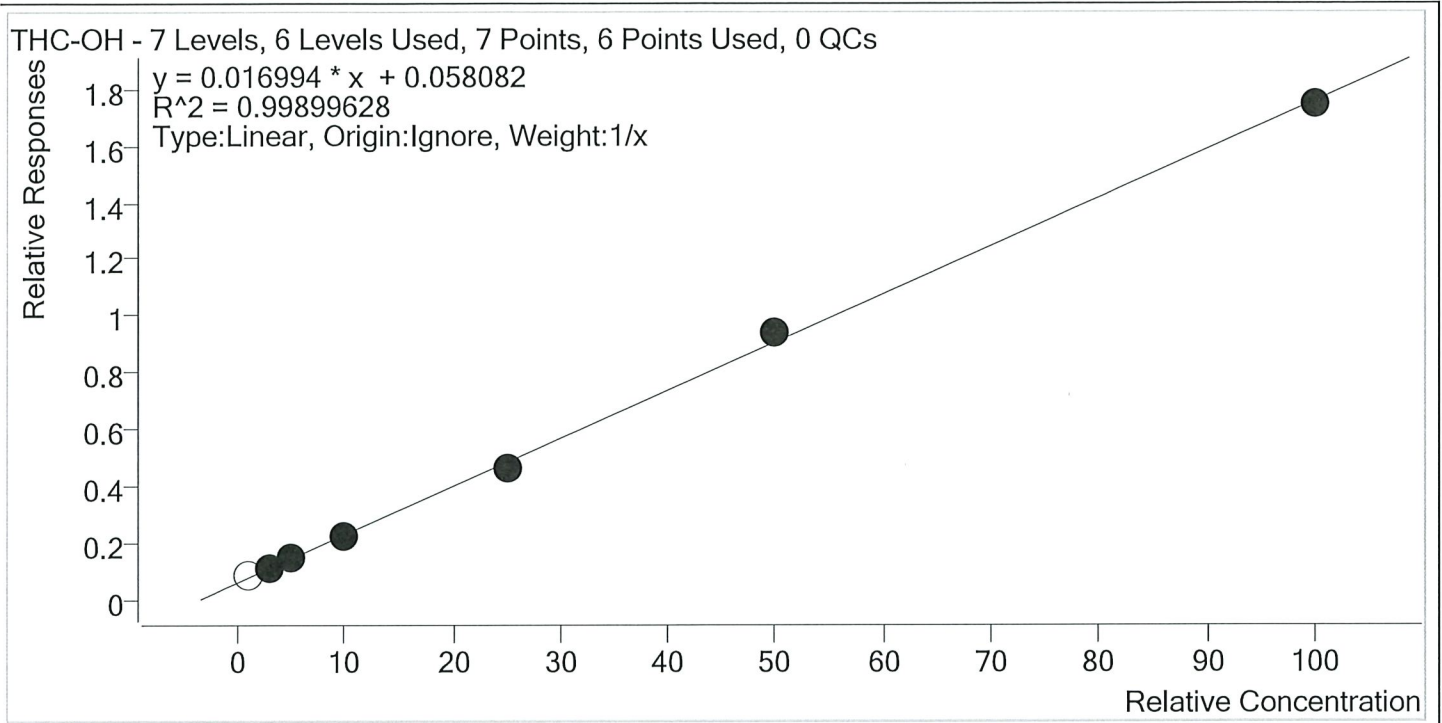
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ Cal 1	1	✓	5.0	5.9	119.0
MJ Cal 2	2	✓	10.0	9.3	93.4
MJ Cal 3	3	✓	20.0	19.0	94.8
MJ Cal 4	4	✓	50.0	47.0	94.0
MJ Cal 5	5	✓	75.0	71.7	95.6
MJ Cal 6	6	✓	100.0	100.7	100.7
MJ Cal 7	7	✓	250.0	256.4	102.6

TS



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
Last Cal. Update 7/24/2020 9:16 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	1.7	168.5
MJ Cal 2	2	✓	3.0	3.0	101.4
MJ Cal 3	3	✓	5.0	5.2	103.1
MJ Cal 4	4	✓	10.0	9.7	96.6
MJ Cal 5	5	✓	25.0	23.9	95.7
MJ Cal 6	6	✓	50.0	51.9	103.9
MJ Cal 7	7	✓	100.0	99.3	99.3

TS

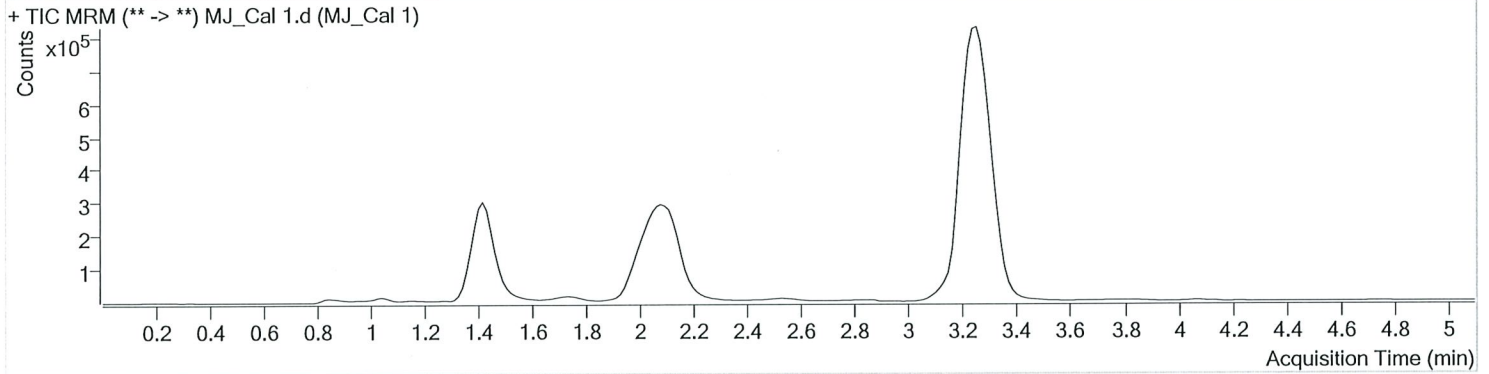


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 7/24/2020 9:16:43 AM

Instrument	Falco	Data File	MJ_Cal 1.d
Type	Cal	Sample	MJ_Cal 1
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P3-A1	Comment	
Injection Volume	10		
Acq. Date-Time	7/23/2020 3:42:29 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.483	105169	∞	4.8 Low	14.58	1212680	1.6854 ng/ml Low
THC-COOH	1.444	55895	∞	49.0	∞	417502	5.9488 ng/ml
THC	3.254	59068	1424.10	25.7	40.42	6625112	1.1394 ng/ml Low

TS

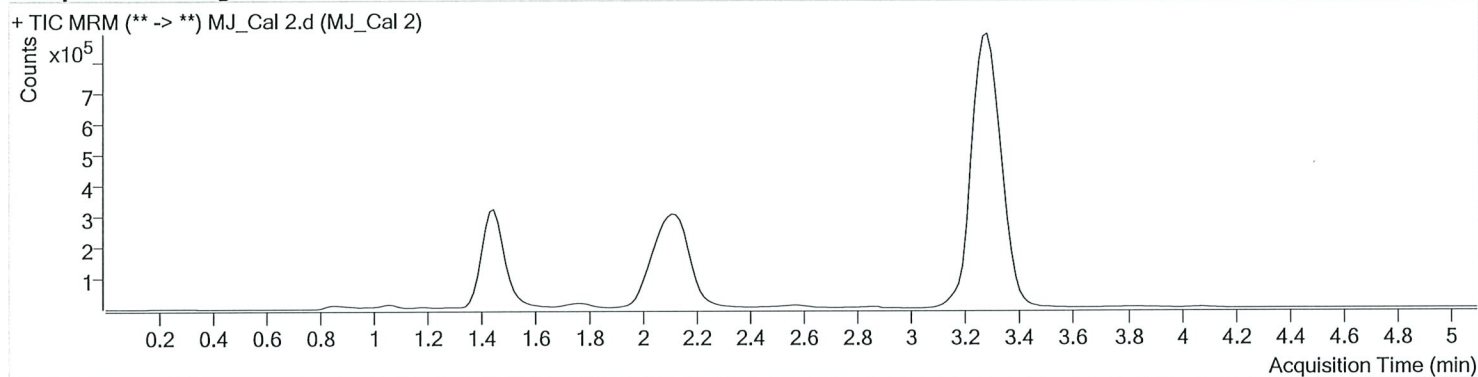


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 7/24/2020 9:16:43 AM

Instrument	Falco	Data File	MJ_Cal 2.d
Type	Cal	Sample	MJ_Cal 2
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P3-B1	Comment	
Injection Volume	10		
Acq. Date-Time	7/23/2020 3:50:13 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.498	132082	∞	8.8	51.56	1202885	3.0435 ng/ml
THC-COOH	1.474	85009	∞	61.6	∞	407191	9.3423 ng/ml
THC	3.285	173496	393.14	29.1	∞	6692336	2.9010 ng/ml Low

TS

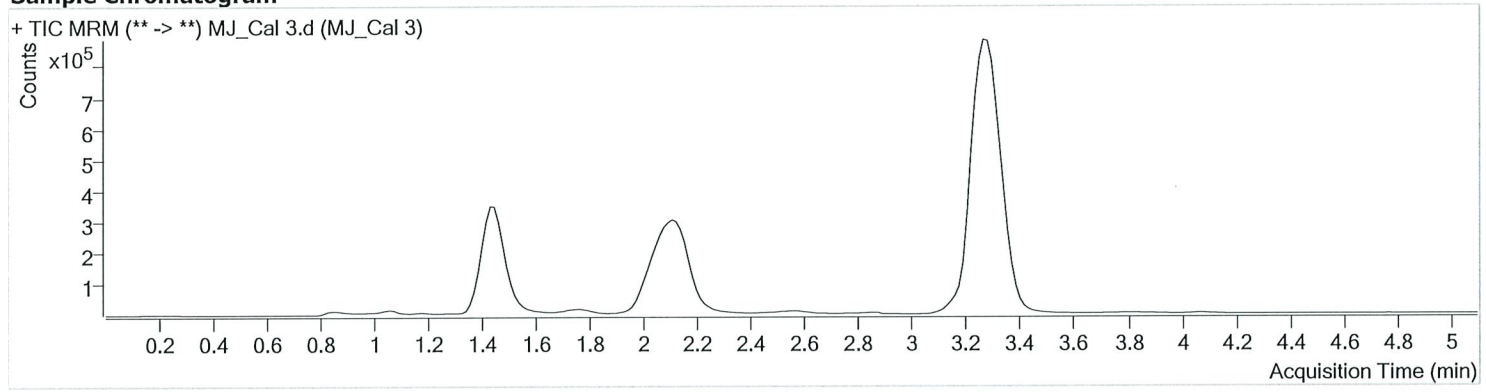


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 7/24/2020 9:16:43 AM

Instrument	Falco	Data File	MJ_Cal 3.d
Type	Cal	Sample	MJ_Cal 3
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P3-C1	Comment	
Injection Volume	10		
Acq. Date-Time	7/23/2020 3:57:49 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	175577	∞	9.0	175.21	1205508	5.1525 ng/ml
THC-COOH	1.474	172036	475.34	59.7	4731.53	408539	18.9637 ng/ml
THC	3.285	291342	427.31	27.2	∞	6564225	4.8128 ng/ml

15

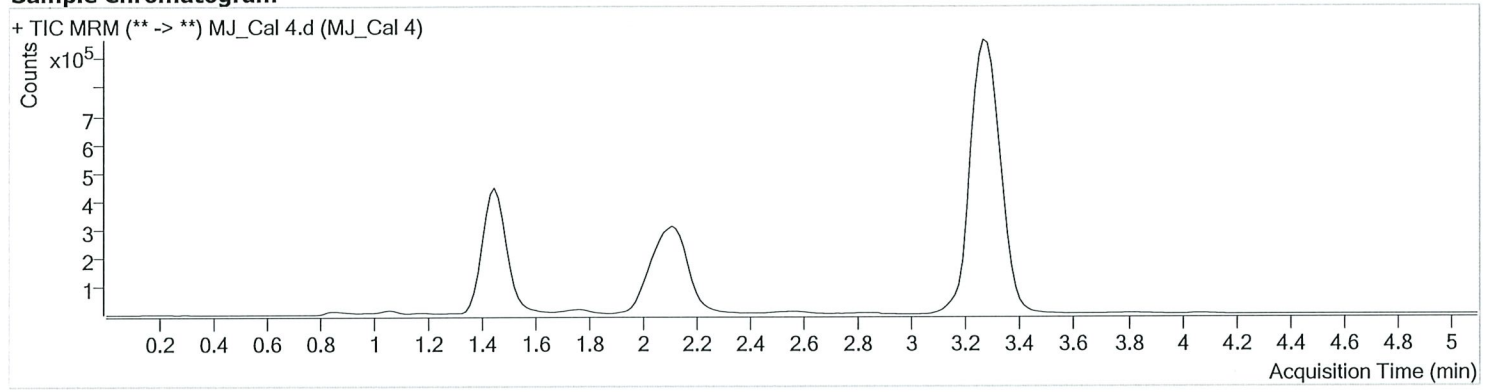


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 7/24/2020 9:16:43 AM

Instrument	Falco	Data File	MJ_Cal 4.d
Type	Cal	Sample	MJ_Cal 4
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P3-D1	Comment	
Injection Volume	10		
Acq. Date-Time	7/23/2020 4:05:24 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	271873	∞	10.0	∞	1222884	9.6643 ng/ml
THC-COOH	1.474	432223	∞	60.0	∞	415874	46.9767 ng/ml
THC	3.285	608977	1738.43	25.4	565.62	6799298	9.4922 ng/ml

TS

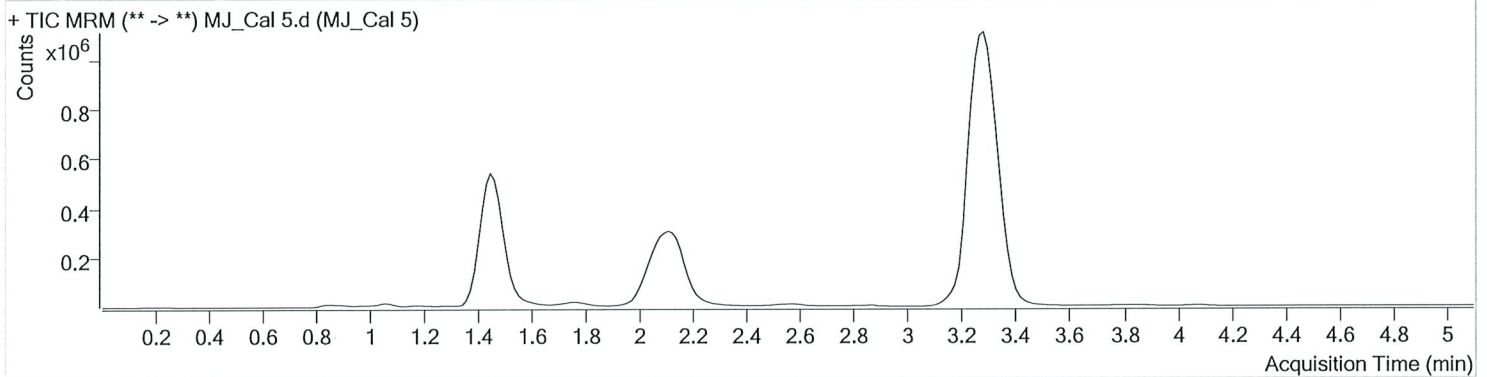


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 7/24/2020 9:16:43 AM

Instrument	Falco	Data File	MJ_Cal 5.d
Type	Cal	Sample	MJ_Cal 5
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P3-E1	Comment	
Injection Volume	10		
Acq. Date-Time	7/23/2020 4:12:59 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	527422	∞	12.1	687.66	1134919	23.9279 ng/ml
THC-COOH	1.474	603936	∞	61.2	∞	381235	71.6652 ng/ml
THC	3.285	1542422	3893.53	25.9	1231.71	6607438	24.3929 ng/ml

TS

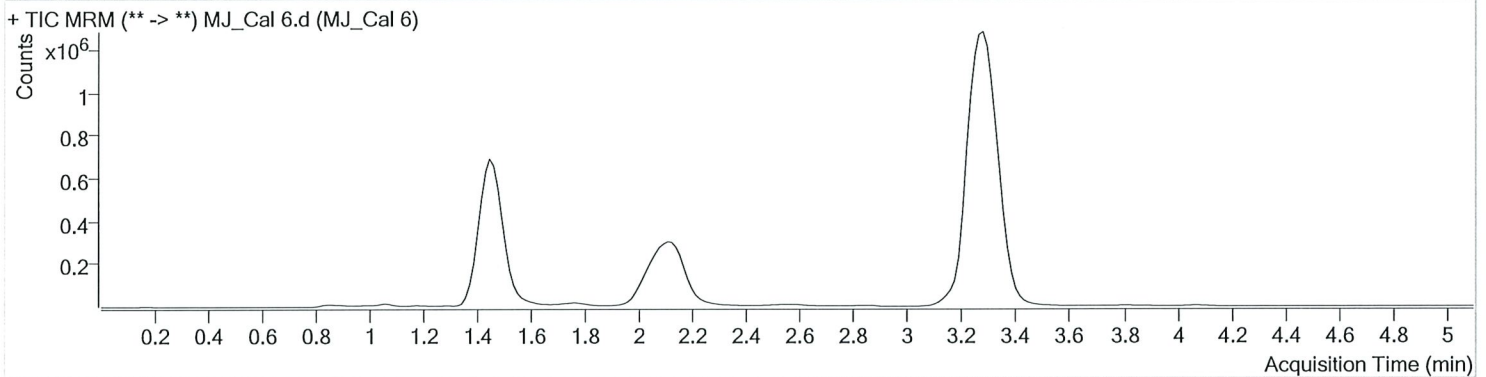


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 7/24/2020 9:16:43 AM

Instrument	Falco	Data File	MJ_Cal 6.d
Type	Cal	Sample	MJ_Cal 6
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P3-F1	Comment	
Injection Volume	10		
Acq. Date-Time	7/23/2020 4:20:33 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	1087468	∞	12.1	∞	1156073	51.9333 ng/ml
THC-COOH	1.474	856479	∞	57.6	∞	384861	100.7230 ng/ml
THC	3.285	3048459	8157.76	26.0	836.05	6426322	49.3461 ng/ml

TS

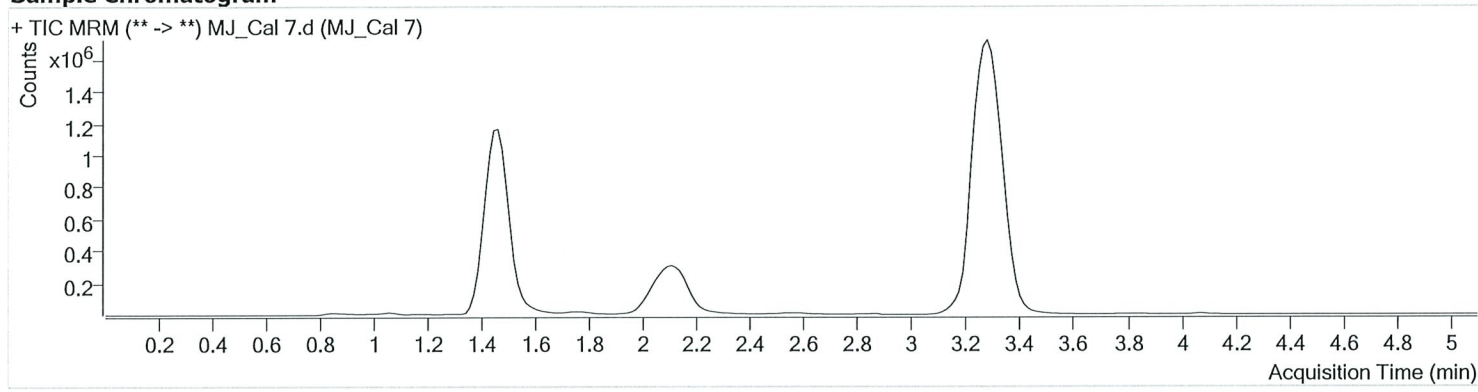


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\072320 AM 27 AM 28 P1 and P2 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 7/24/2020 9:16:43 AM

Instrument	Falco	Data File	MJ_Cal 7.d
Type	Cal	Sample	MJ_Cal 7
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P3-G1	Comment	
Injection Volume	10		
Acq. Date-Time	7/23/2020 4:28:08 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	1920504	∞	12.5	1284.98	1100414	99.2784 ng/ml
THC-COOH	1.474	1983833	∞	58.6	∞	350464	256.3803 ng/ml
THC	3.285	5944750	21748.02	26.3	∞	6054017	101.9157 ng/ml