

REVIEWED

By Briany Wylie at 4:12 pm, Nov 18, 2020

TS

Worklist: 4603

<u>LAB_CASE</u>	<u>ITEM</u>	<u>ITEM_TYPE</u>	<u>DESCRIPTION</u>	
M2020-3425	2	BCK	AM 27 Blood THC Quant by LC-QQQ	
M2020-3939	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
M2020-4218	3	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3118	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3137	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3145	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3152	5	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3166	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3167	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3168	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3169	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3179	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3180	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3187	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-3344	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: 11/12/2020
Plate lot#: IDP-108-200723

Analyst: Tamara Salazar
Plate Expiration: 01/23/21

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: Lampire 20K20702
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.

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 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? 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 3-100, THC-OH 3-100*

Samples were extracted on 11/12/2020; however, a clog in the system caused the run to stop midway. The obstruction was cleared, and the run was continued on 11/13/2020.

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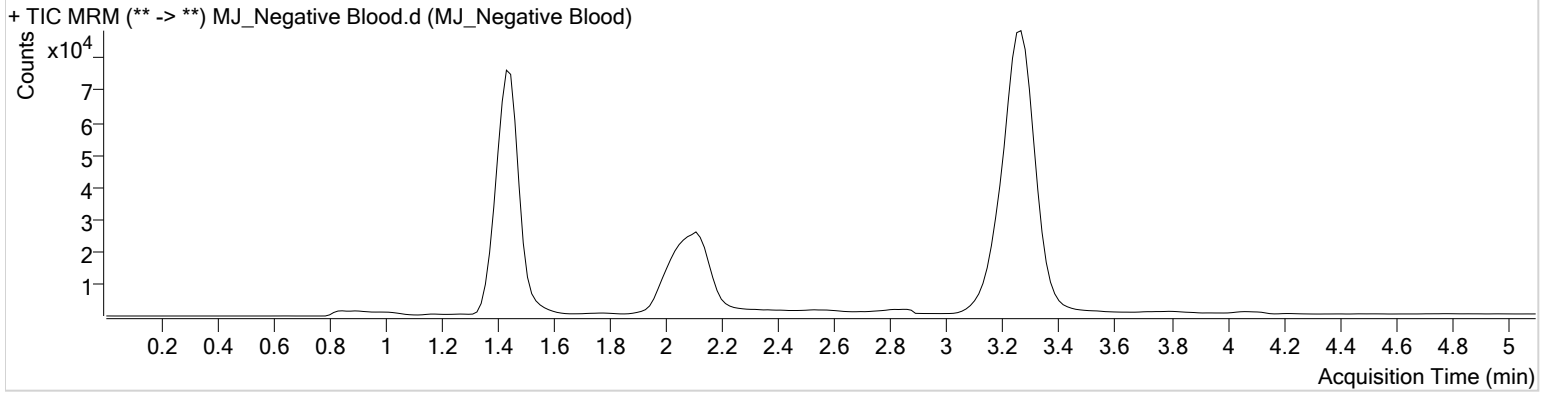


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/18/2020 11:22:46 AM

Instrument	Falco	Data File	MJ_Negative Blood.d
Type	Sample	Sample	MJ_Negative Blood
Acq. Method	AM 27 THCQ.m	Operator	Tamara Salazar
Sample Position	P4-H5	Comment	
Injection Volume	10		
Acq. Date-Time	11/12/2020 5:30:14 PM		
Sample Info.			

Sample Chromatogram



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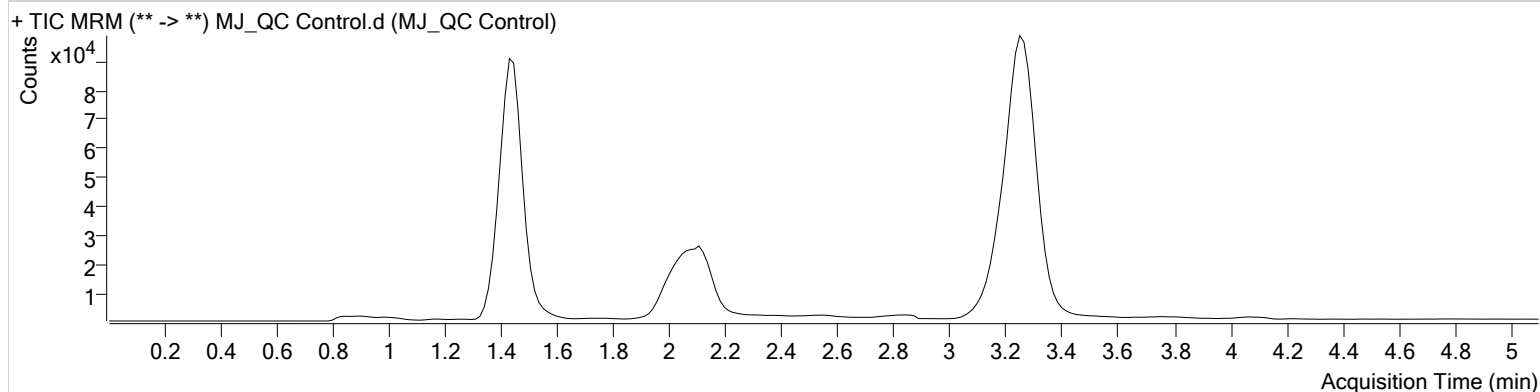
AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/18/2020 11:22:46 AM

Instrument	Falco	Data File	MJ_QC Control.d
Type	Sample	Sample	MJ_QC Control
Acq. Method	AM 27 THCQ.m	Operator	Tamara Salazar
Sample Position	P4-A6	Comment	
Injection Volume	10		
Acq. Date-Time	11/12/2020 5:15:02 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	25624	246.69	15.5	51.60	324843	4.3805 ng/ml
THC-COOH	1.474	42510	∞	62.1	∞	95365	16.0984 ng/ml
THC	3.270	34511	197.83	29.6	∞	774552	4.7122 ng/ml

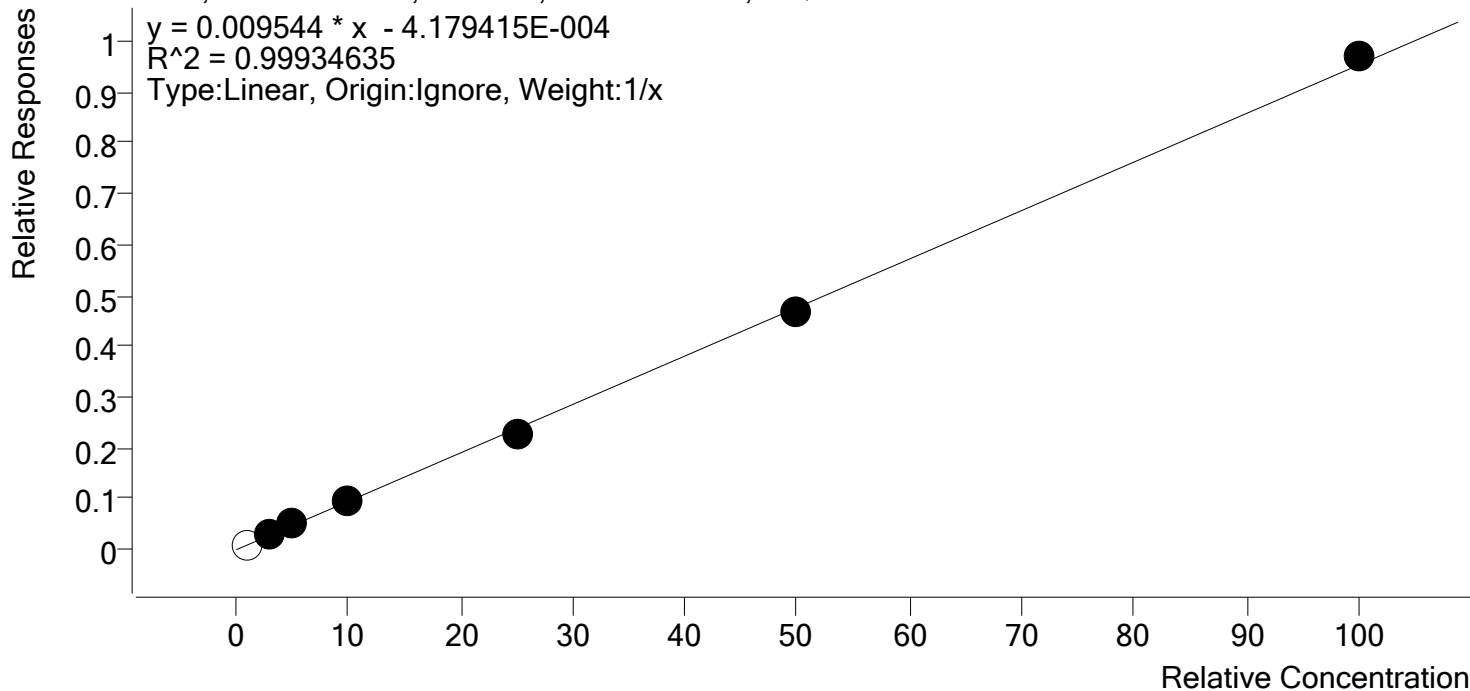
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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Last Cal. Update 11/18/2020 11:22 AM
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	1.1	108.4
MJ_Cal 2	2	✓	3.0	3.0	98.9
MJ_Cal 3	3	✓	5.0	5.3	105.7
MJ_Cal 4	4	✓	10.0	9.9	98.8
MJ_Cal 5	5	✓	25.0	24.2	97.0
MJ_Cal 6	6	✓	50.0	48.9	97.9
MJ_Cal 7	7	✓	100.0	101.7	101.7

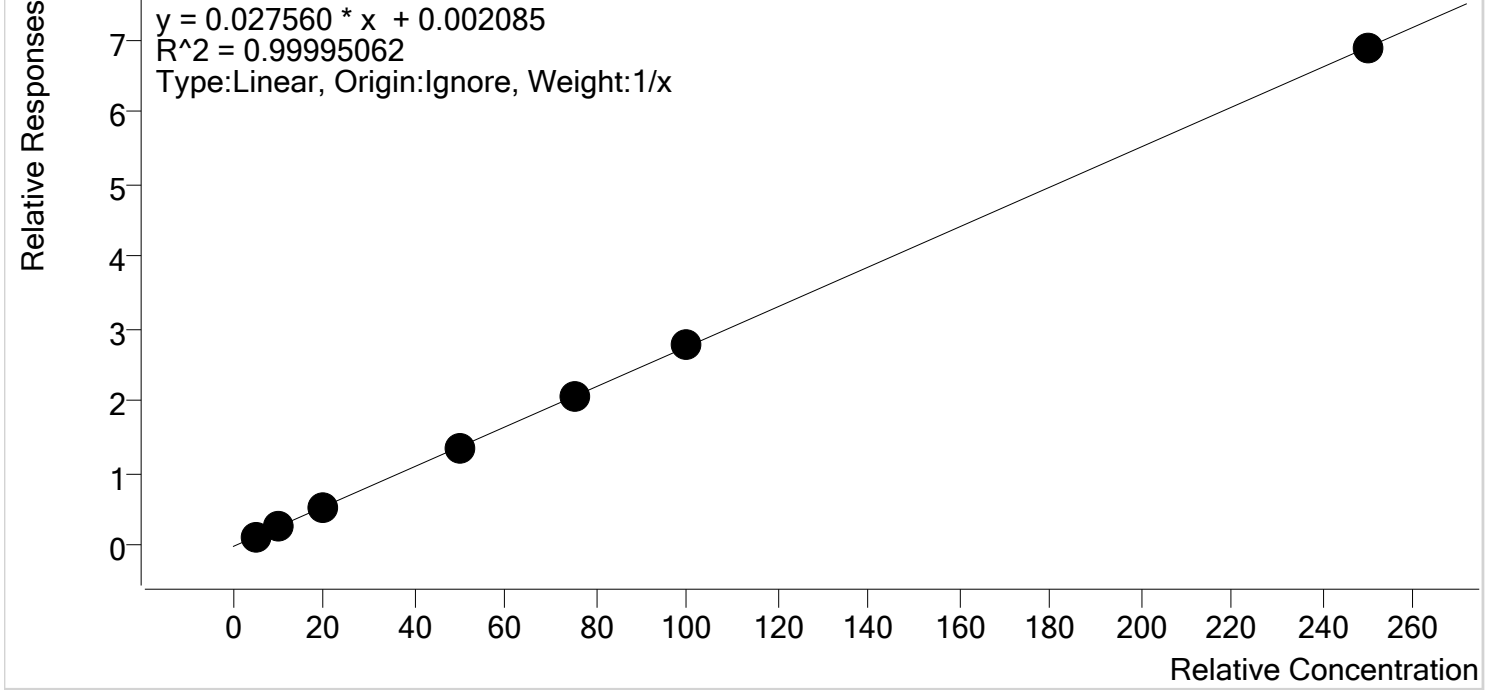


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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Last Cal. Update 11/18/2020 11:22 AM
Analyst Name ISP\datastor
Analyte THC-COOH **Internal Standard** THC-COOH-D9

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



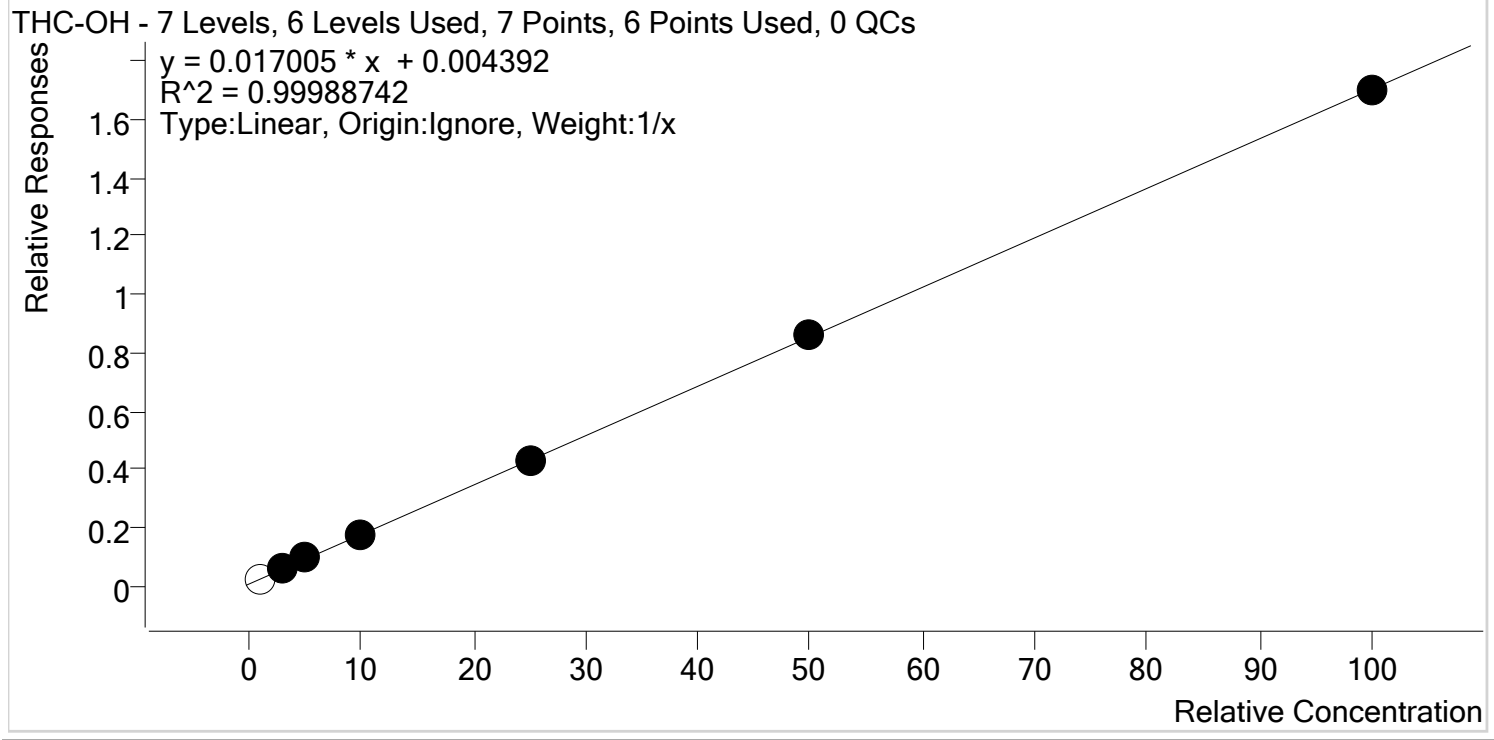
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	✓	5.0	4.9	97.2
MJ_Cal 2	2	✓	10.0	10.2	102.0
MJ_Cal 3	3	✓	20.0	20.1	100.5
MJ_Cal 4	4	✓	50.0	49.7	99.4
MJ_Cal 5	5	✓	75.0	75.7	100.9
MJ_Cal 6	6	✓	100.0	100.2	100.2
MJ_Cal 7	7	✓	250.0	249.2	99.7

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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Last Cal. Update 11/18/2020 11:22 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	x	1.0	1.1	109.3
MJ_Cal 2	2	✓	3.0	2.9	95.4
MJ_Cal 3	3	✓	5.0	5.2	103.9
MJ_Cal 4	4	✓	10.0	10.0	100.2
MJ_Cal 5	5	✓	25.0	25.2	100.7
MJ_Cal 6	6	✓	50.0	50.0	100.1
MJ_Cal 7	7	✓	100.0	99.7	99.7

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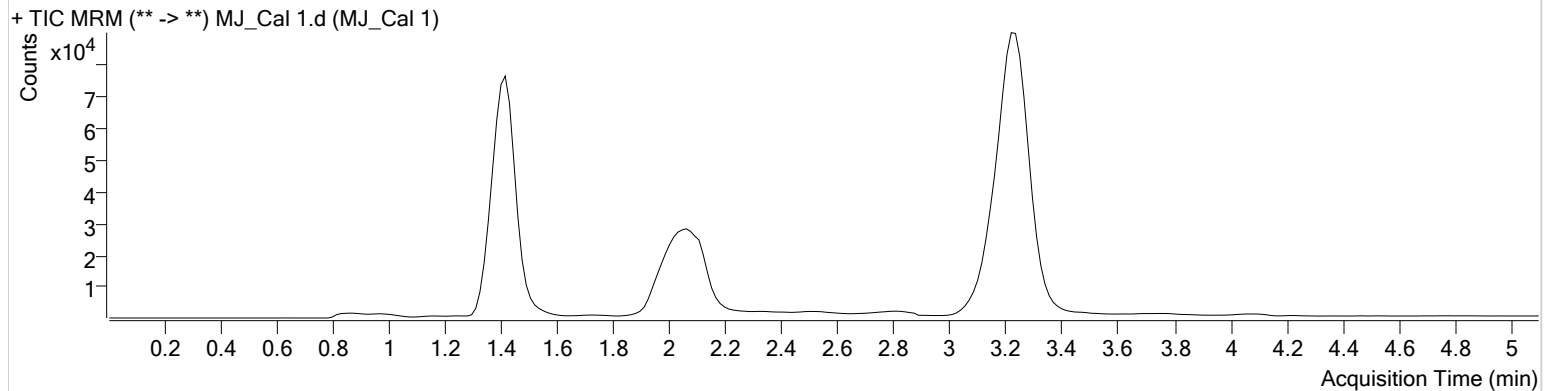


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/18/2020 11:22:46 AM

Instrument Falco **Data File** MJ_Cal 1.d
Type Cal **Sample** MJ_Cal 1
Acq. Method AM 27 THCQ.m **Operator** Tamara Salazar
Sample Position P4-H6 **Comment**
Injection Volume 10
Acq. Date-Time 11/12/2020 4:14:03 PM
Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.423	7544	7.06 Low	13.8	14.44	328423	1.0925 ng/ml Low
THC-COOH	1.444	12709	11.50	65.0	∞	93387	4.8622 ng/ml Low
THC	3.239	7478	6.82 Low	35.6 High	8.16 Low	753530	1.0835 ng/ml Low

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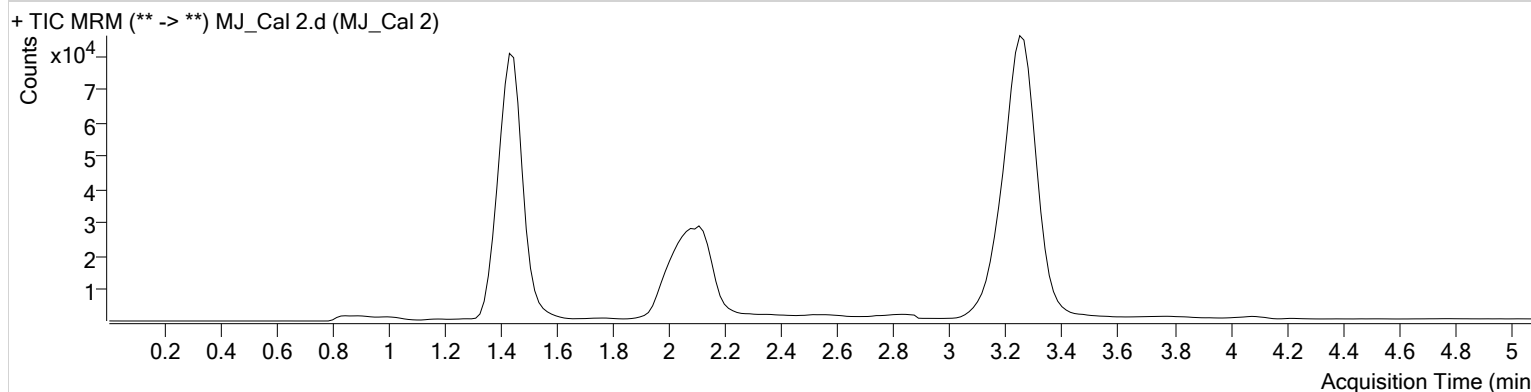


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/18/2020 11:22:46 AM

Instrument	Falco	Data File	MJ_Cal 2.d
Type	Cal	Sample	MJ_Cal 2
Acq. Method	AM 27 THCQ.m	Operator	Tamara Salazar
Sample Position	P4-G6	Comment	
Injection Volume	10		
Acq. Date-Time	11/12/2020 4:21:47 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	17409	203.55	14.9	17.26	328074	2.8623 ng/ml Low
THC-COOH	1.474	26086	232.27	62.5	340.25	92102	10.2013 ng/ml
THC	3.270	19551	68.10	30.4	11.60	700860	2.9667 ng/ml Low

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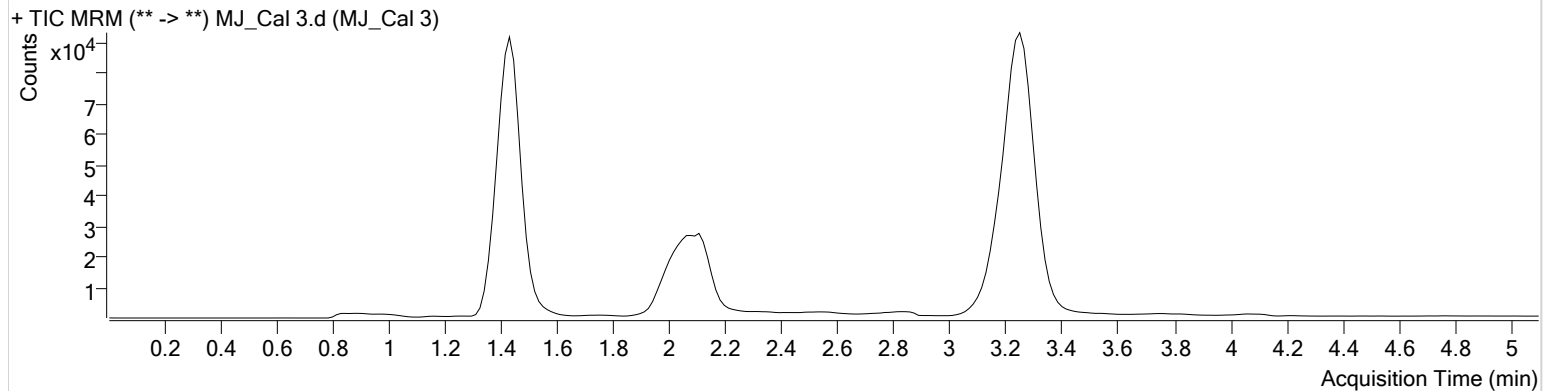


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/18/2020 11:22:46 AM

Instrument	Falco	Data File	MJ_Cal 3.d
Type	Cal	Sample	MJ_Cal 3
Acq. Method	AM 27 THCQ.m	Operator	Tamara Salazar
Sample Position	P4-F6	Comment	
Injection Volume	10		
Acq. Date-Time	11/12/2020 4:29:23 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	30371	∞	13.7	60.58	327540	5.1946 ng/ml
THC-COOH	1.459	51546	∞	63.4	∞	92691	20.1022 ng/ml
THC	3.270	36380	282.27	27.4	23.93	726986	5.2871 ng/ml

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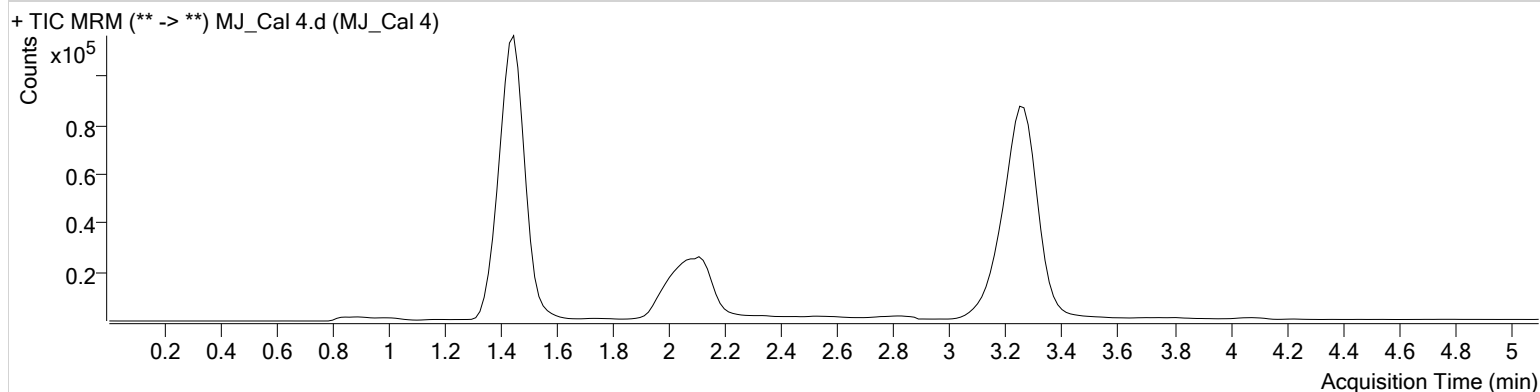


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/18/2020 11:22:46 AM

Instrument	Falco	Data File	MJ_Cal 4.d
Type	Cal	Sample	MJ_Cal 4
Acq. Method	AM 27 THCQ.m	Operator	Tamara Salazar
Sample Position	P4-E6	Comment	
Injection Volume	10		
Acq. Date-Time	11/12/2020 4:37:00 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	58601	∞	13.6	186.37	335196	10.0228 ng/ml
THC-COOH	1.459	130287	∞	63.6	∞	94953	49.7107 ng/ml
THC	3.270	64176	408.13	27.4	59.91	683273	9.8850 ng/ml

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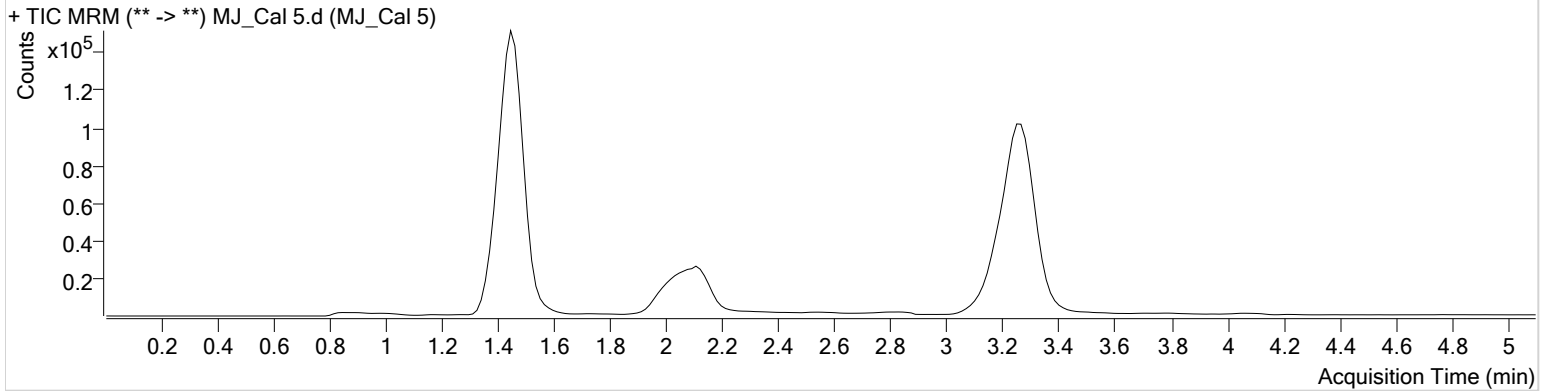


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/18/2020 11:22:46 AM

Instrument	Falco	Data File	MJ_Cal 5.d
Type	Cal	Sample	MJ_Cal 5
Acq. Method	AM 27 THCQ.m	Operator	Tamara Salazar
Sample Position	P4-D6	Comment	
Injection Volume	10		
Acq. Date-Time	11/12/2020 4:44:36 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	142615	∞	13.9	223.53	329809	25.1709 ng/ml
THC-COOH	1.474	194163	1451.26	64.0	∞	92984	75.6909 ng/ml
THC	3.270	160592	1088.26	29.7	∞	695323	24.2434 ng/ml

TS

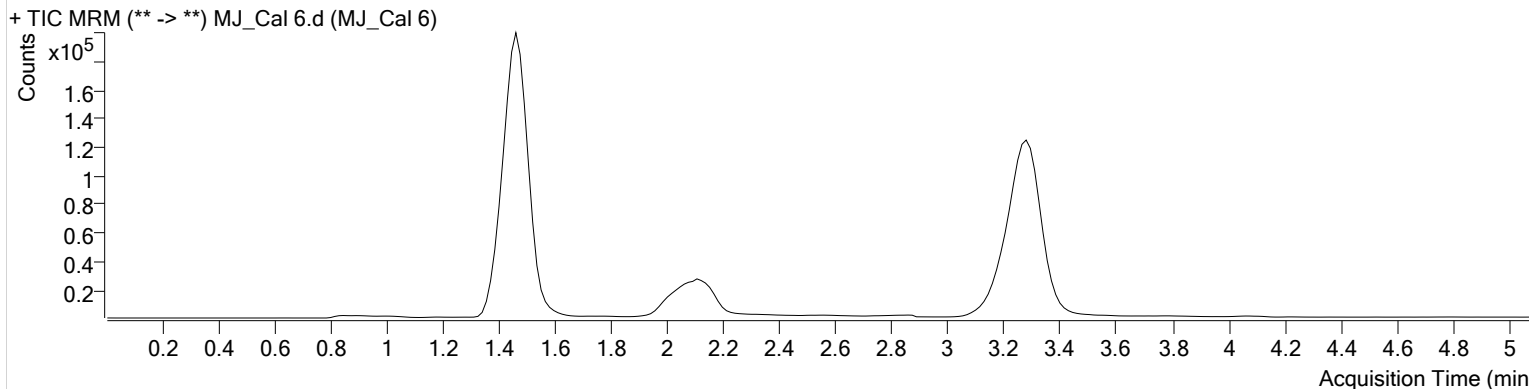


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/18/2020 11:22:46 AM

Instrument	Falco	Data File	MJ_Cal 6.d
Type	Cal	Sample	MJ_Cal 6
Acq. Method	AM 27 THCQ.m	Operator	Tamara Salazar
Sample Position	P4-C6	Comment	
Injection Volume	10		
Acq. Date-Time	11/12/2020 4:52:12 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	293014	∞	13.7	1627.35	342603	50.0370 ng/ml
THC-COOH	1.489	261243	∞	63.9	∞	94536	100.1930 ng/ml
THC	3.285	317752	1807.93	28.3	∞	681094	48.9261 ng/ml

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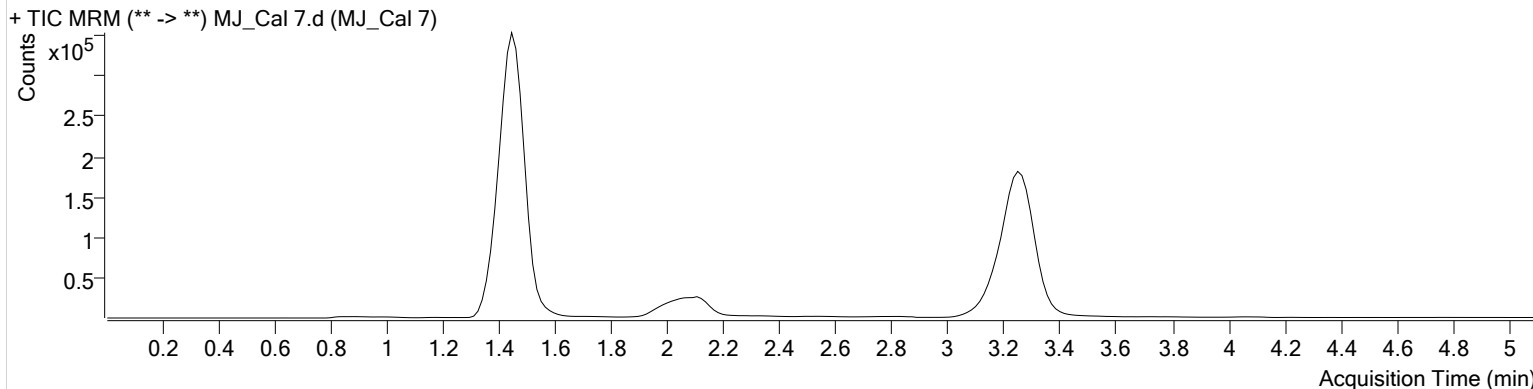


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111220 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/18/2020 11:22:46 AM

Instrument	Falco	Data File	MJ_Cal 7.d
Type	Cal	Sample	MJ_Cal 7
Acq. Method	AM 27 THCQ.m	Operator	Tamara Salazar
Sample Position	P4-B6	Comment	
Injection Volume	10		
Acq. Date-Time	11/12/2020 4:59:49 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	573665	∞	14.0	1680.36	337456	99.7124 ng/ml
THC-COOH	1.459	620212	∞	64.8	∞	90263	249.2397 ng/ml
THC	3.270	688270	9559.82	26.3	1406.86	709465	101.6917 ng/ml