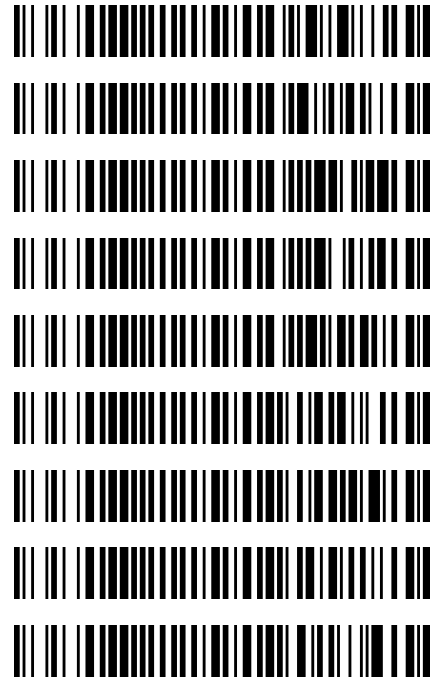


REVIEWED
By Sarah Pickle at 3:18 pm, Nov 05, 2020

TS 11/4/2020

Worklist: 4582

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
P2020-2798	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2812	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2839	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2840	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2848	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2876	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2877	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2880	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2924	2	BCK	AM 27 Blood THC Quant by LC-QQQ



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

Extraction Date: 11/03/2020
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: 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.

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 5-100 (evaluated qualitatively)*

TS

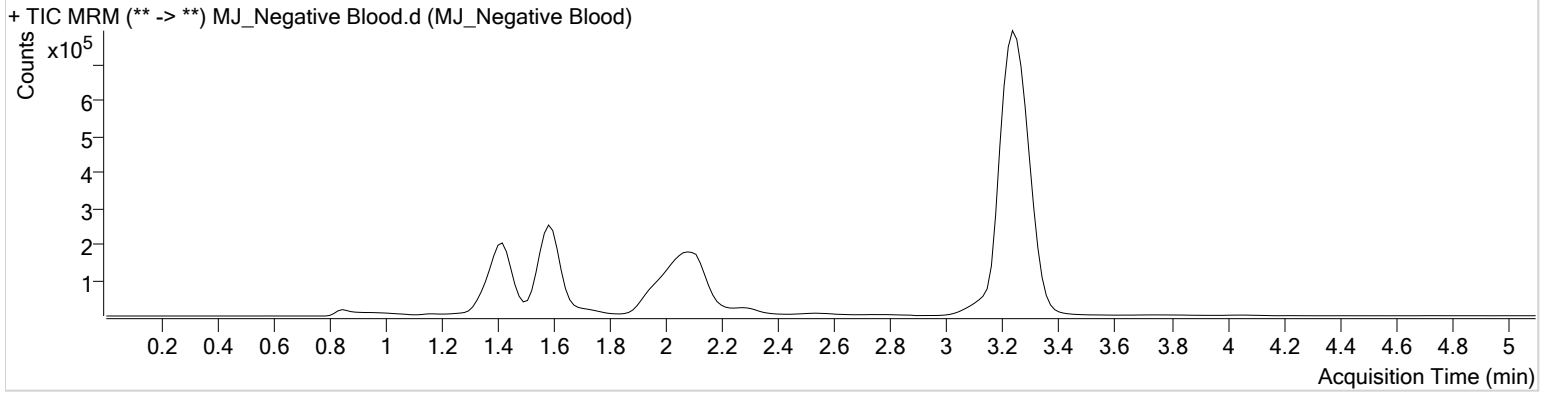


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/4/2020 7:52:17 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	P4-H5	Comment	
Injection Volume	10		
Acq. Date-Time	11/3/2020 1:53:30 PM		
Sample Info.			

Sample Chromatogram



TS

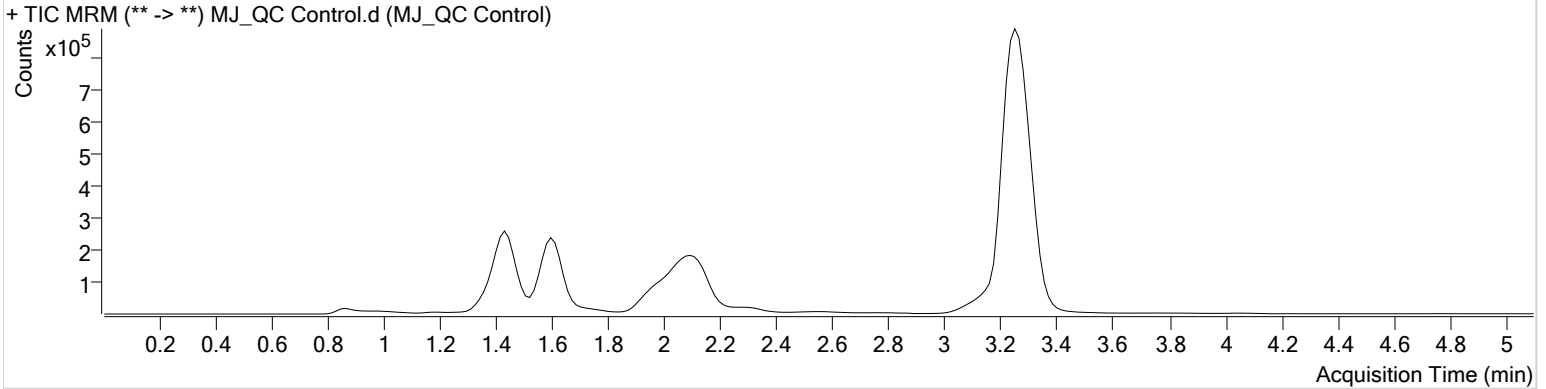


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/4/2020 7:52:17 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	P4-A6	Comment	
Injection Volume	10		
Acq. Date-Time	11/3/2020 1:38:18 PM		

Sample Chromatogram



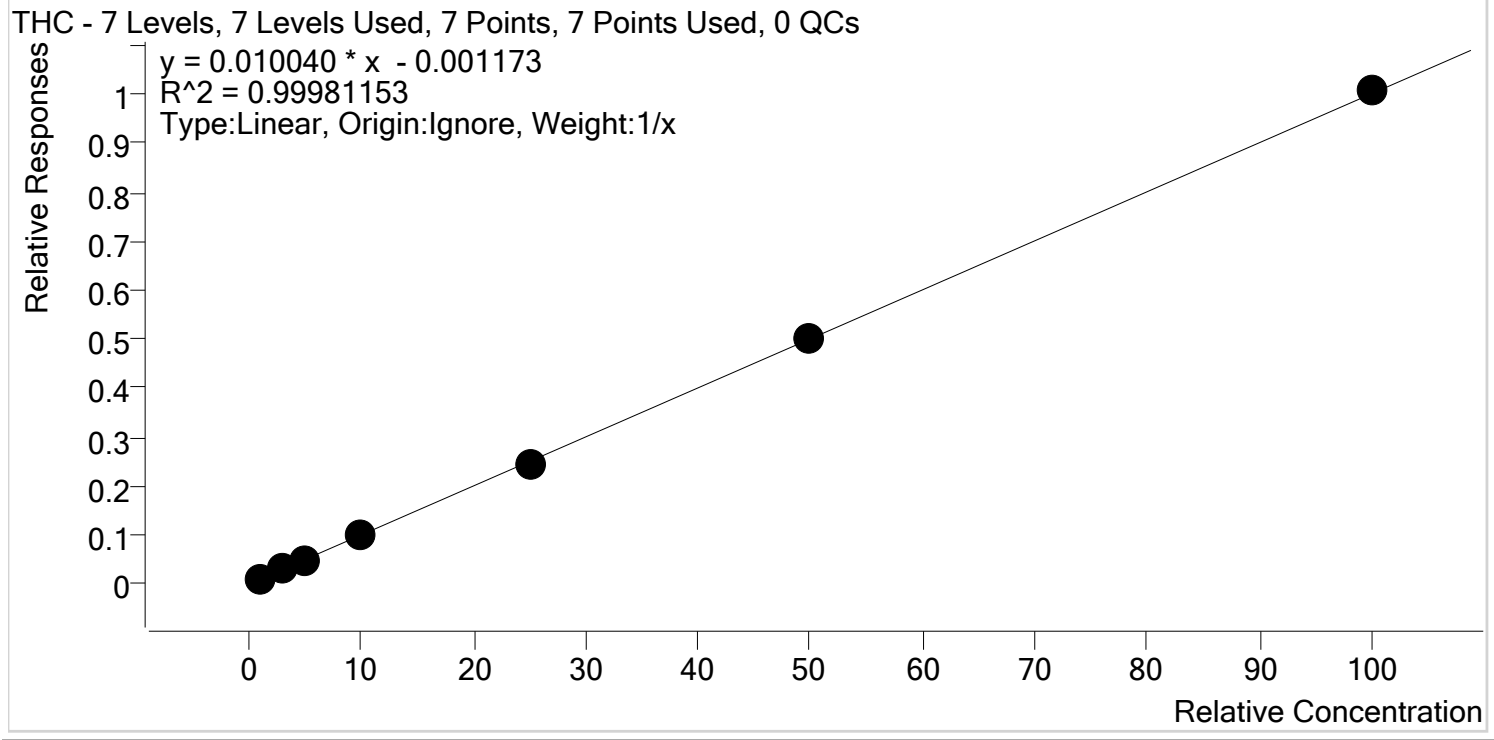
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	132846	∞	9.9	105.86	999653	3.2052 ng/ml
THC-COOH	1.459	112036	∞	61.6	∞	254525	15.0976 ng/ml
THC	3.270	273955	1373.97	27.2	216.70	6197257	4.5200 ng/ml

TS



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Last Cal. Update 11/4/2020 7:52 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	108.9
MJ_Cal 2	2	✓	3.0	2.9	96.1
MJ_Cal 3	3	✓	5.0	4.9	98.5
MJ_Cal 4	4	✓	10.0	9.7	96.6
MJ_Cal 5	5	✓	25.0	24.7	99.0
MJ_Cal 6	6	✓	50.0	50.1	100.2
MJ_Cal 7	7	✓	100.0	100.6	100.6

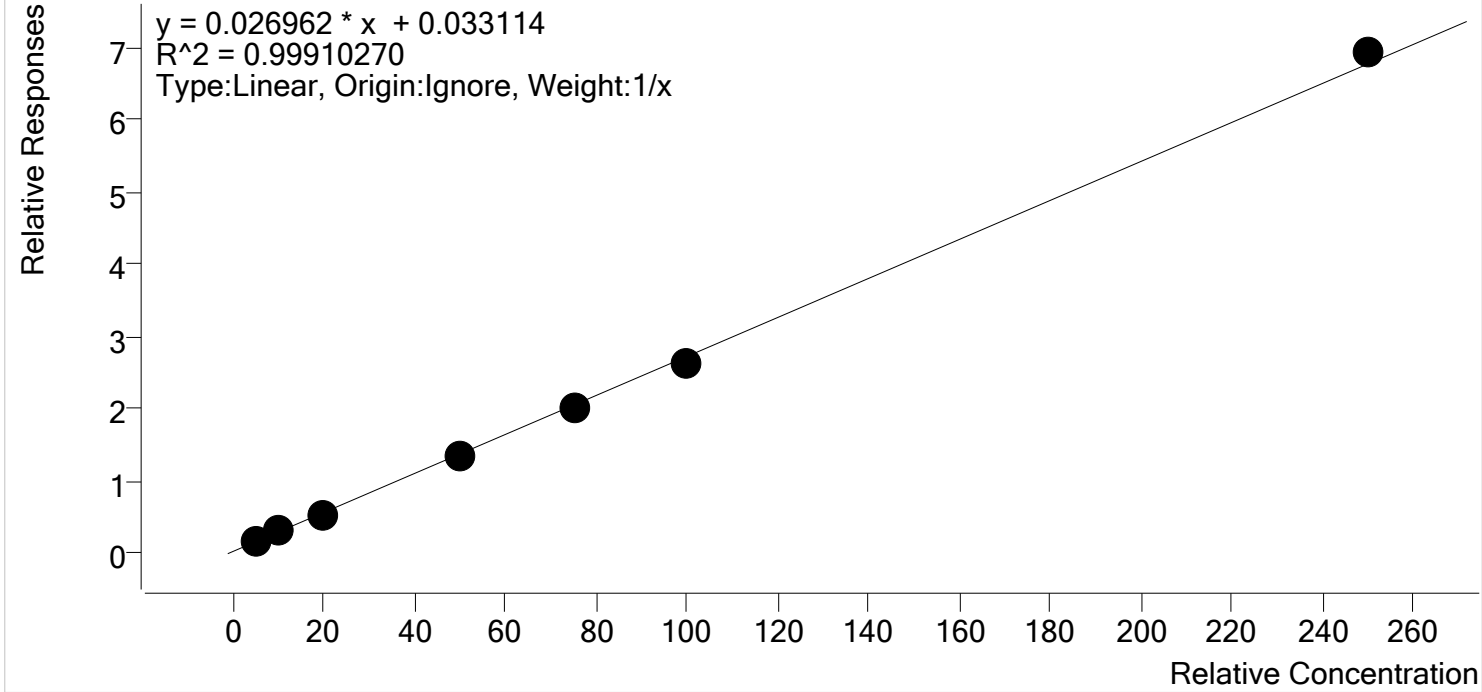
TS



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Last Cal. Update 11/4/2020 7:52 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



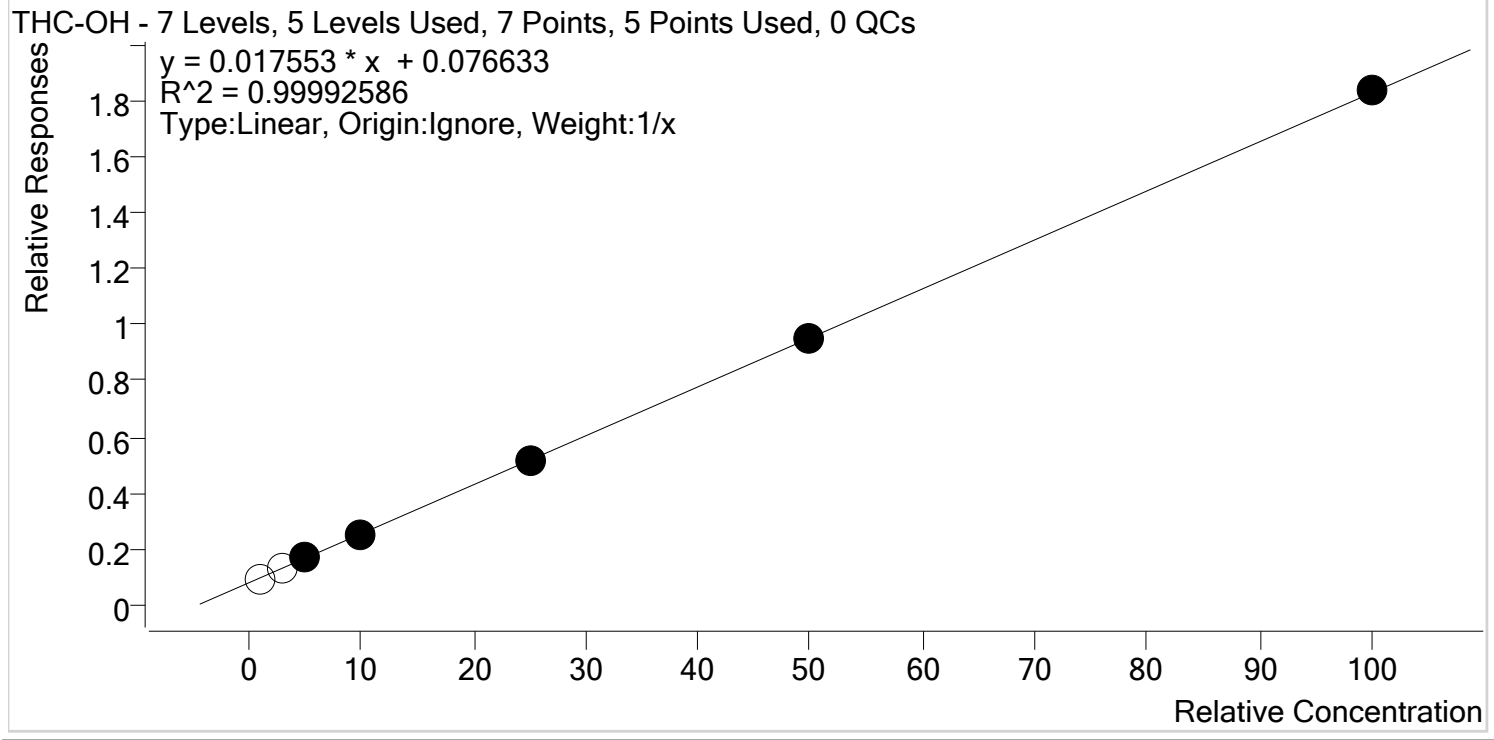
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	✓	5.0	5.4	108.7
MJ_Cal 2	2	✓	10.0	9.9	99.1
MJ_Cal 3	3	✓	20.0	19.2	96.2
MJ_Cal 4	4	✓	50.0	49.5	99.0
MJ_Cal 5	5	✓	75.0	73.5	98.0
MJ_Cal 6	6	✓	100.0	96.6	96.6
MJ_Cal 7	7	✓	250.0	255.8	102.3

TS



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Last Cal. Update 11/4/2020 7:52 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	0.8	83.8
MJ_Cal 2	2	x	3.0	2.8	94.2
MJ_Cal 3	3	✓	5.0	5.1	101.8
MJ_Cal 4	4	✓	10.0	9.9	99.4
MJ_Cal 5	5	✓	25.0	24.6	98.6
MJ_Cal 6	6	✓	50.0	49.9	99.8
MJ_Cal 7	7	✓	100.0	100.4	100.4

TS

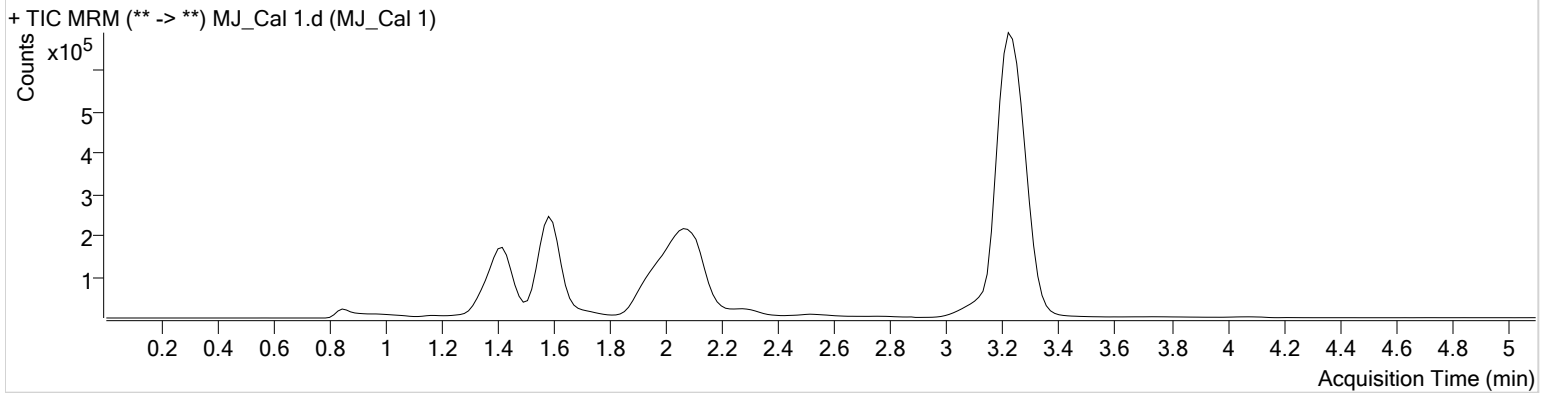


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/4/2020 7:52:17 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 P4-H6 **Comment**
Injection Volume 10
Acq. Date-Time 11/3/2020 12:37:04 PM
Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.468	74199	∞	5.6 Low	13.20	812285	0.8382 ng/ml Low
THC-COOH	1.429	36024	∞	55.5	∞	200499	5.4356 ng/ml
THC	3.239	49243	254.38	29.8	∞	5043542	1.0894 ng/ml Low

TS



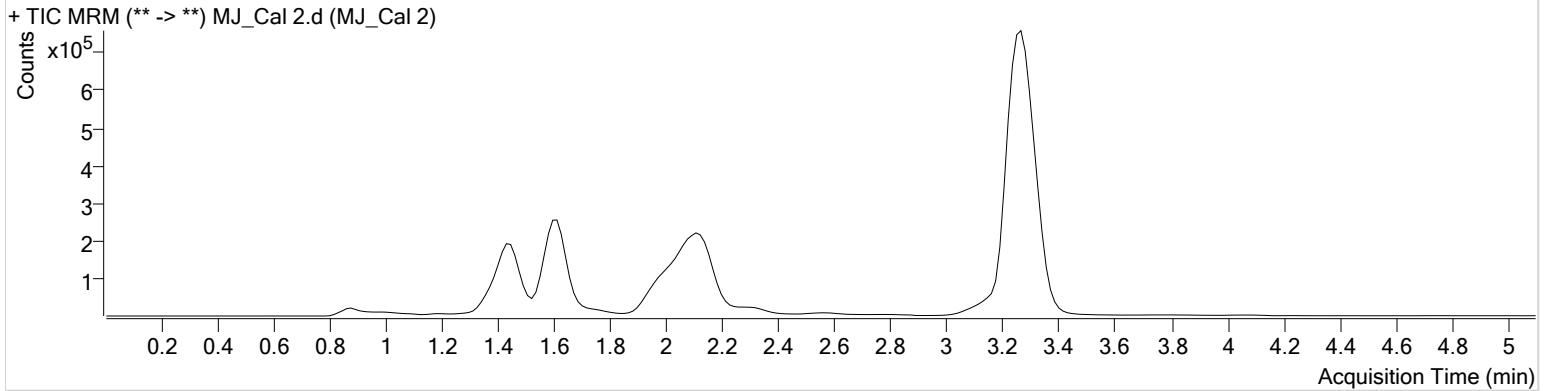
AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/4/2020 7:52:17 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	P4-G6	Comment	
Injection Volume	10		
Acq. Date-Time	11/3/2020 12:44:49 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.483 High	104832	∞	7.5 Low	∞	830439	2.8260 ng/ml Low
THC-COOH	1.474	60634	∞	59.7	∞	201898	9.9104 ng/ml
THC	3.270	148818	135.72	28.6	85.91	5358673	2.8830 ng/ml Low

TS



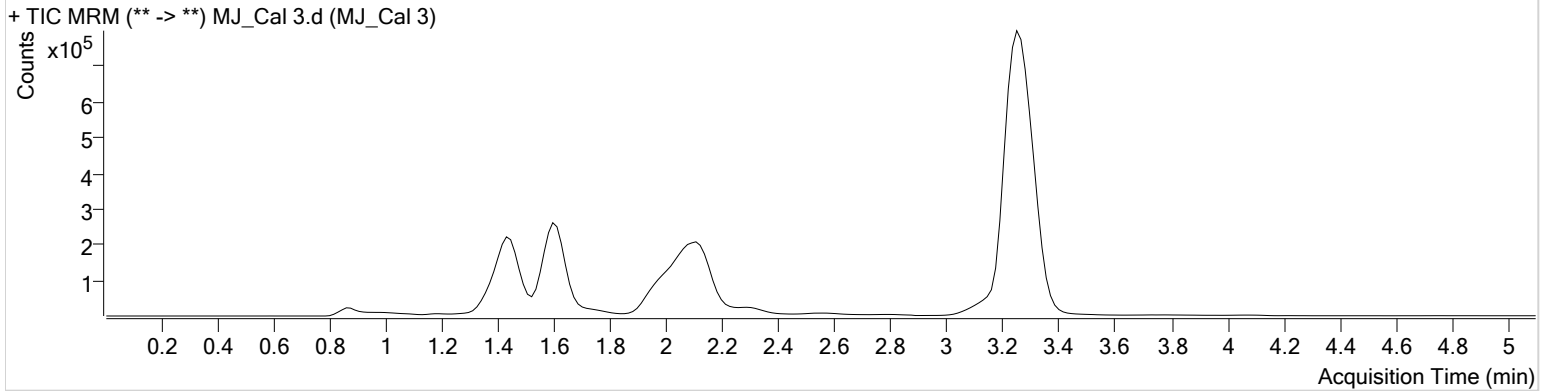
AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/4/2020 7:52:17 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	P4-F6	Comment	
Injection Volume	10		
Acq. Date-Time	11/3/2020 12:52:25 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	142818	∞	9.0	76.28	860443	5.0903 ng/ml
THC-COOH	1.459	114594	∞	61.4	∞	207563	19.2483 ng/ml
THC	3.270	267255	1654.10	25.7	135.31	5533740	4.9273 ng/ml

TS

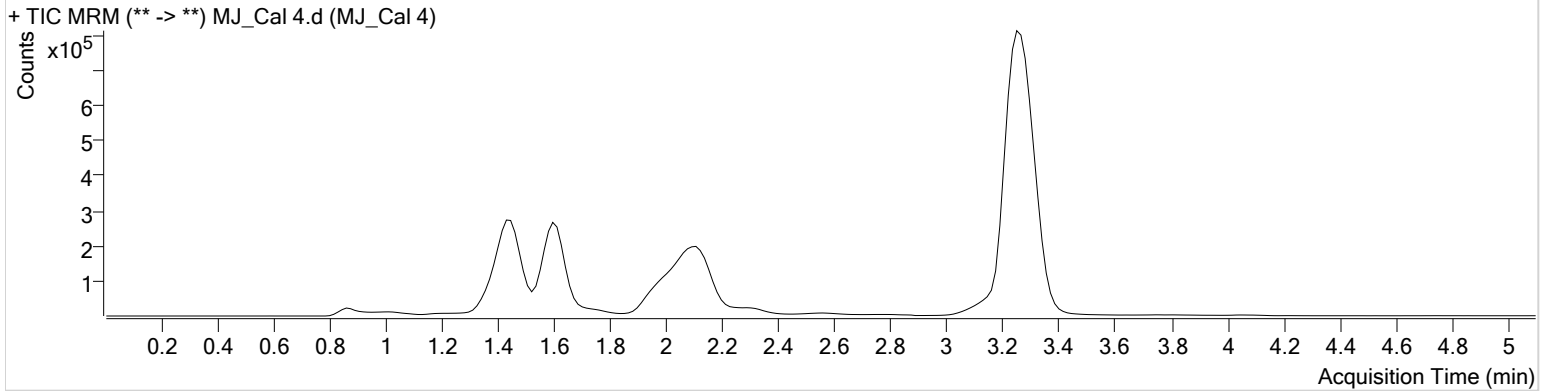


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/4/2020 7:52:17 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	P4-E6	Comment	
Injection Volume	10		
Acq. Date-Time	11/3/2020 1:00:01 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	216118	∞	10.5	∞	860720	9.9390 ng/ml
THC-COOH	1.459	281876	∞	57.6	∞	206158	49.4831 ng/ml
THC	3.270	519452	7013.35	26.4	913.73	5419682	9.6636 ng/ml

TS

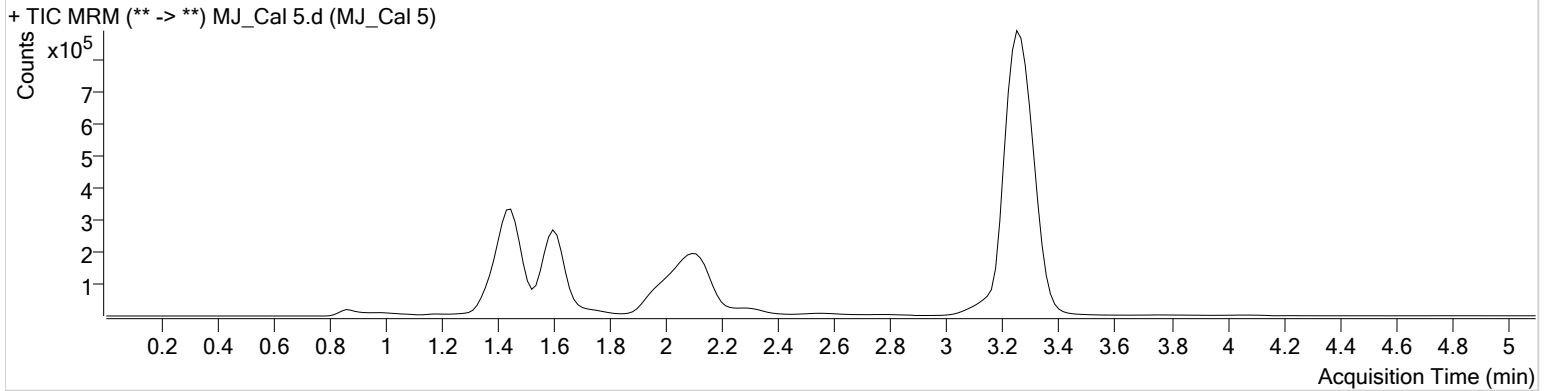


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/4/2020 7:52:17 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	P4-D6	Comment	
Injection Volume	10		
Acq. Date-Time	11/3/2020 1:07:37 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	425805	∞	12.1	567.62	836239	24.6434 ng/ml
THC-COOH	1.459	397961	∞	58.5	∞	197439	73.5290 ng/ml
THC	3.270	1246554	5833.83	25.8	454.83	5041381	24.7457 ng/ml

TS

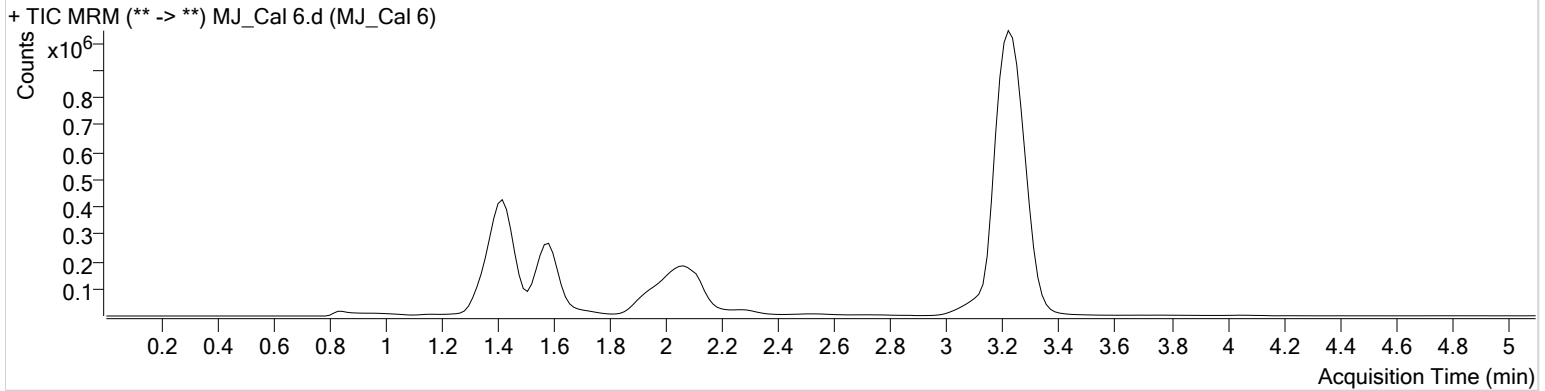


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/4/2020 7:52:17 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	P4-C6	Comment	
Injection Volume	10		
Acq. Date-Time	11/3/2020 1:15:23 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.408	800087	∞	12.4	441.98	839926	49.9032 ng/ml
THC-COOH	1.429	525654	∞	60.5	∞	199220	96.6332 ng/ml
THC	3.224	2460530	2170.01	26.0	∞	4902611	50.1069 ng/ml

TS

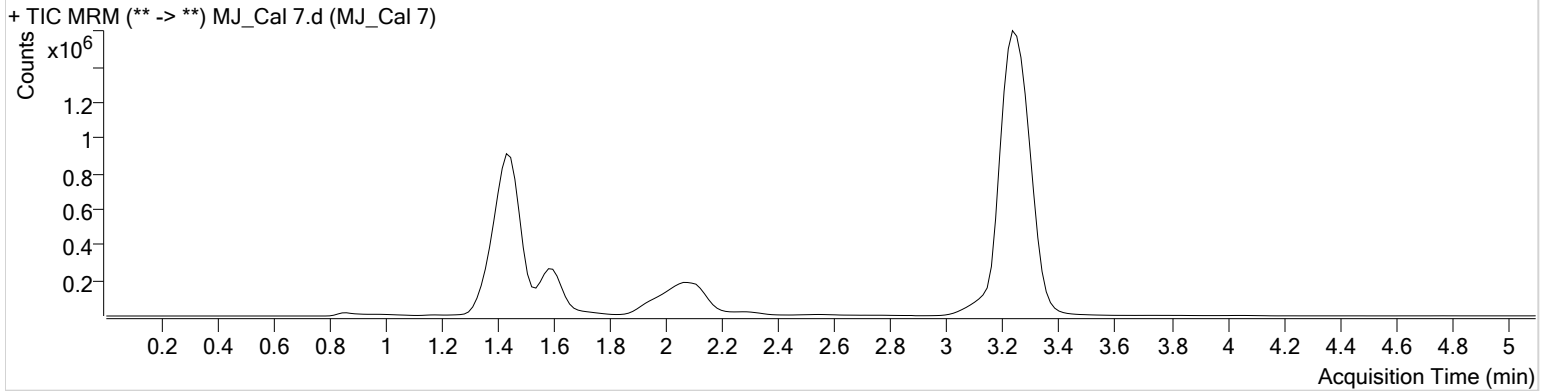


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\110320 AM 27 28 TS\QuantResults\AM 27.batch.bin
Calibration Last Update 11/4/2020 7:52:17 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	P4-B6	Comment	
Injection Volume	10		
Acq. Date-Time	11/3/2020 1:23:03 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.423	1782611	∞	12.9	3364.58	969153	100.4242 ng/ml
THC-COOH	1.459	1609733	∞	61.7	∞	232319	255.7604 ng/ml
THC	3.239	5440696	6138.72	26.3	3601.34	5394017	100.5841 ng/ml