

REVIEWED

By Tamara Salazar at 8:29 am, Aug 11, 2020

8/7/2020

Worklist: 4420

<u>LAB_CASE</u>	<u>ITEM</u>	<u>ITEM_TYPE</u>	<u>DESCRIPTION</u>	
M2020-2568	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
M2020-2806	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-2066	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-2073	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-2074	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-2075	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-2091	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-2093	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-2103	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-2108	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2020-2150	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: 08/06/20

Analyst: Sarah Pickle

Plate lot#: IDP-108-2-200303

Plate Expiration: 09/03/20

Mobile phase A: 0.1% Formic Acid in LCMS Water
MTBE

Mobile phase B: 0.1% Formic acid in Acetonitrile
Hexane

Blank Blood Lot: Hemostat 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. Pipette **1000 µL blood (calibrated pipette)** in wells of analytical (standards) plate. **Pipette ID: #27**
- 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 4. Pipette **500 µL 0.1% formic acid in LCMS water** in wells of analytical plate.
- 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 6. Transfer **800 µL 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 85-95 PSI- Selector to the right) Manifold ID: 067104
- 8. Wait 5 minutes.
- 9. Add **2.25 mL MTBE. (Add in 3 increments of 750 µL)**
- 10. Wait 5 minutes.
- 11. Apply positive pressure for approx. 15 seconds. *(12-15 PSI- Selector to the left).*
- 12. Add **2.25 mL Hexane. (Add in 3 increments of 750 µL)**
- 13. Wait 5 minutes.
- 14. Apply positive pressure for approx. 15 seconds. *(12-15 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 **100 µL 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: ~~10ng/mL~~ ^{5 ng/mL 8/11/20 P} (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: *Hands of the analyst Sophie Jackson. Curves limited: THC-OH 3-100*

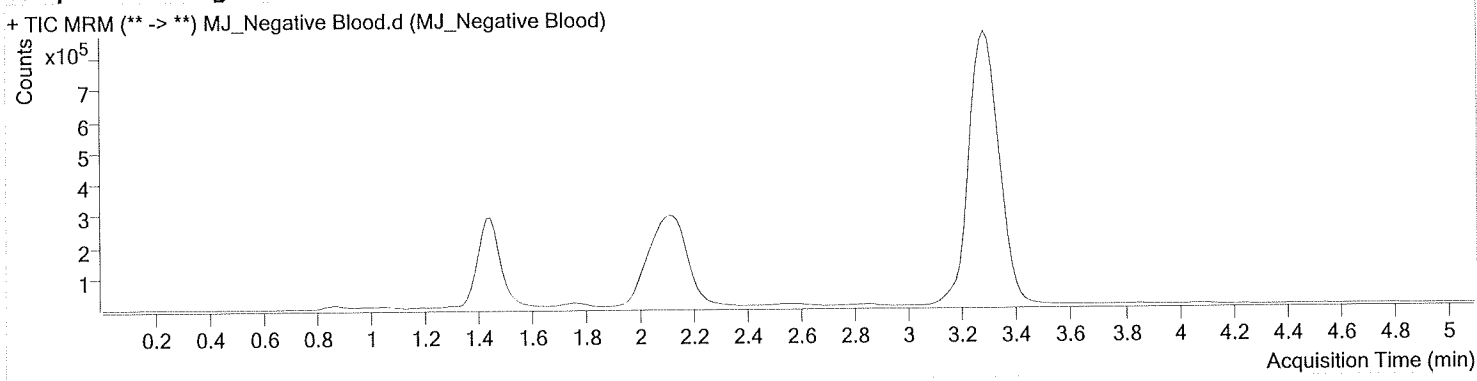


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
Calibration Last Update 8/7/2020 9:36:29 AM

Instrument	Falco	Data File	MJ_Negative Blood.d
Type	Sample	Sample	MJ_Negative Blood
Acq. Method	AM 27 THC quant.m	Operator	Sarah Pickle
Sample Position	P3-H5	Comment	
Injection Volume	10		
Acq. Date-Time	8/6/2020 2:00:45 PM		
Sample Info.			

Sample Chromatogram





AM #27 Cannabinoid Quant. Results

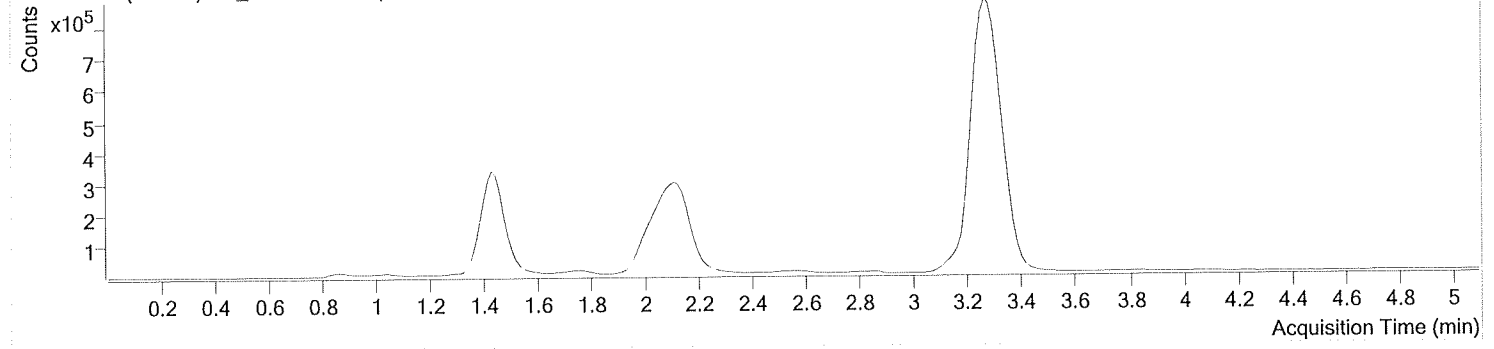
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Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
Calibration Last Update 8/7/2020 9:36:29 AM

Instrument	Falco	Data File	MJ_QC Control.d
Type	Sample	Sample	MJ_QC Control
Acq. Method	AM 27 THC quant.m	Operator	Sarah Pickle
Sample Position	P3-A6	Comment	
Injection Volume	10		
Acq. Date-Time	8/6/2020 1:45:32 PM		

Sample Chromatogram

+ TIC MRM (** -> **) MJ_QC Control.d (MJ_QC Control)



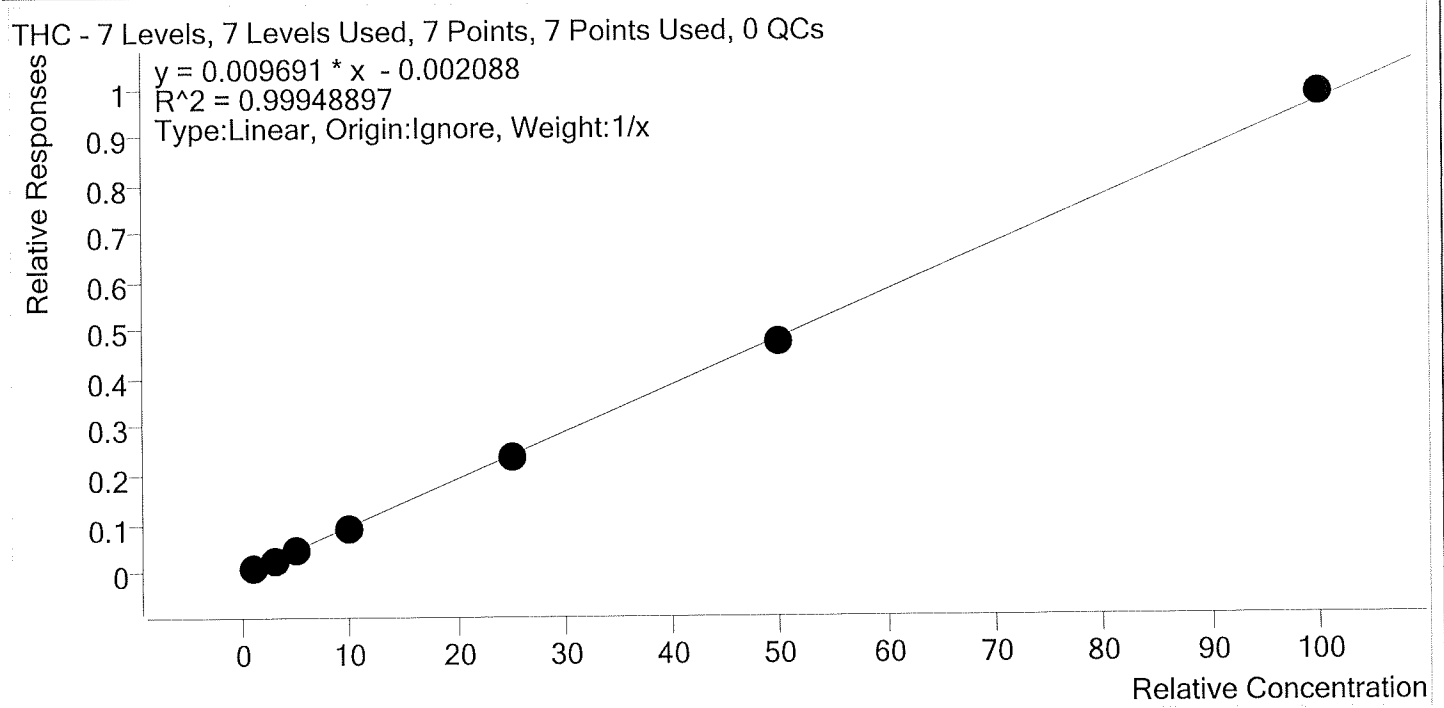
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	182827	∞	8.8	∞	1256957	5.0729 ng/ml
THC-COOH	1.459	150709	∞	55.6	∞	422993	15.5388 ng/ml
THC	3.285	272383	∞	28.0	1265.00	6854563	4.3161 ng/ml

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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
 Last Cal. Update 8/7/2020 9:36 AM
 Analyst Name ISP\datastor
 Analyte THC Internal Standard THC-D3



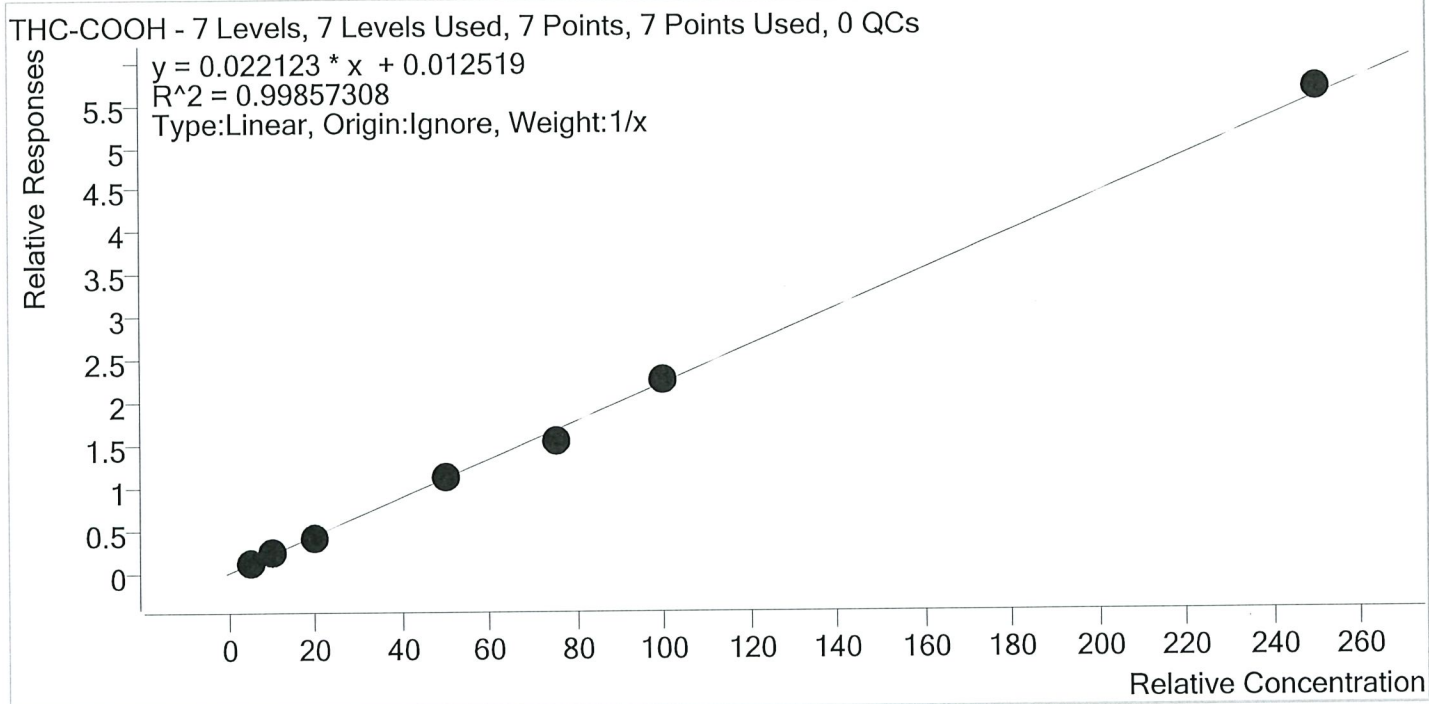
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ Cal 1	1	✓	1.0	1.1	112.7
MJ Cal 2	2	✓	3.0	2.9	96.4
MJ Cal 3	3	✓	5.0	4.8	95.4
MJ Cal 4	4	✓	10.0	9.7	97.2
MJ Cal 5	5	✓	25.0	24.4	97.7
MJ Cal 6	6	✓	50.0	49.4	98.9
MJ Cal 7	7	✓	100.0	101.6	101.6

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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
 Last Cal. Update 8/7/2020 9:36 AM
 Analyst Name ISP\datastor
 Analyte THC-COOH Internal Standard THC-COOH-D9



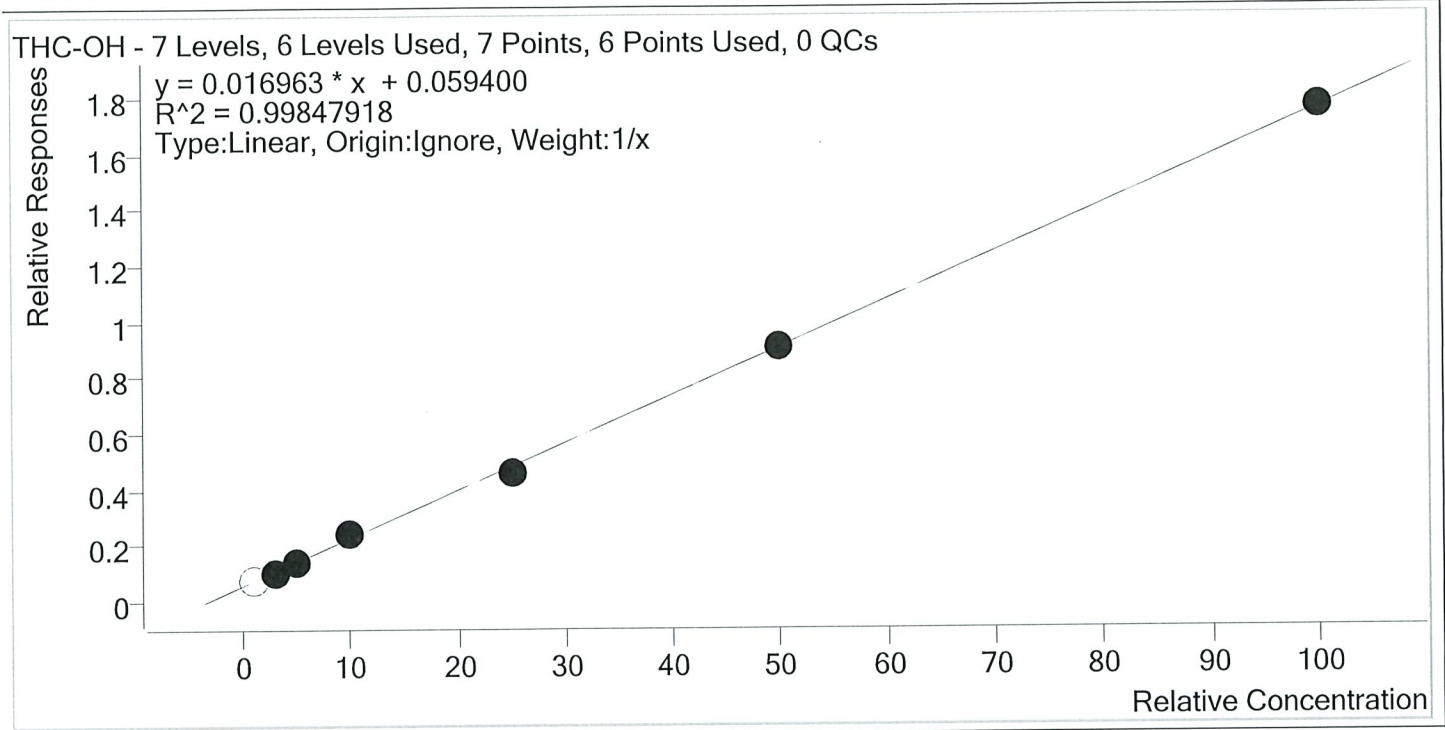
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	✓	5.0	5.5	109.0
MJ_Cal 2	2	✓	10.0	10.2	102.5
MJ_Cal 3	3	✓	20.0	18.4	92.1
MJ_Cal 4	4	✓	50.0	49.8	99.5
MJ_Cal 5	5	✓	75.0	70.4	93.8
MJ_Cal 6	6	✓	100.0	101.2	101.2
MJ_Cal 7	7	✓	250.0	254.5	101.8

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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
 Last Cal. Update 8/7/2020 9:36 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	1.2	122.2
MJ Cal 2	2	✓	3.0	2.8	92.3
MJ Cal 3	3	✓	5.0	5.0	99.5
MJ Cal 4	4	✓	10.0	11.3	112.5
MJ Cal 5	5	✓	25.0	24.0	96.1
MJ Cal 6	6	✓	50.0	49.7	99.4
MJ Cal 7	7	✓	100.0	100.3	100.3

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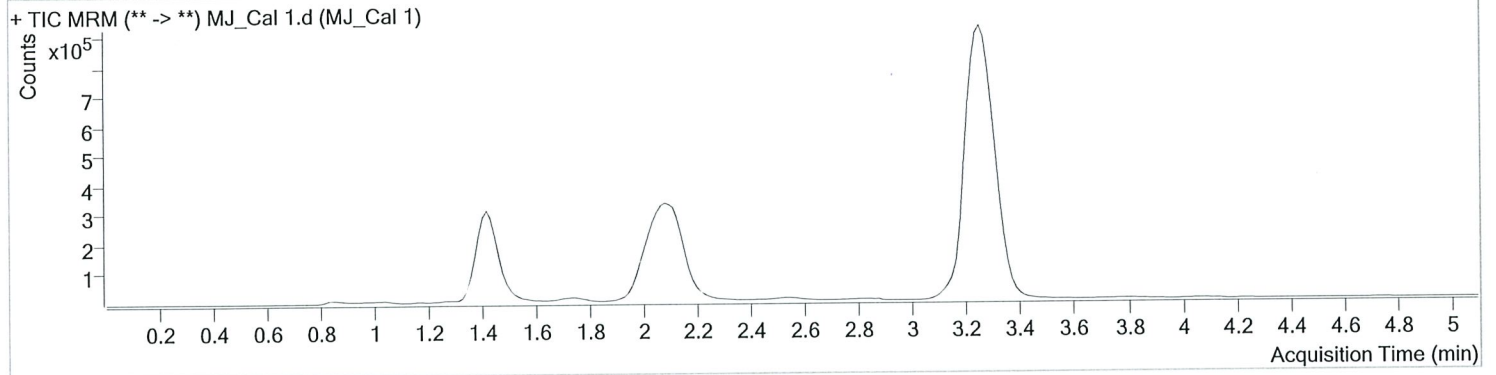


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
Calibration Last Update 8/7/2020 9:36:29 AM

Instrument	Falco	Data File	MJ_Cal 1.d
Type	Cal	Sample	MJ_Cal 1
Acq. Method	AM 27 THC quant.m	Operator	Sarah Pickle
Sample Position	P3-H6	Comment	
Injection Volume	10		
Acq. Date-Time	8/6/2020 12:44:44 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.483	98243	∞	5.2 Low	19.30	1226111	1.2218 ng/ml Low
THC-COOH	1.444	55399	∞	46.2	∞	416176	5.4510 ng/ml
THC	3.270	63486	550.75	27.2	149.42	7187065	1.1270 ng/ml Low

SJ P

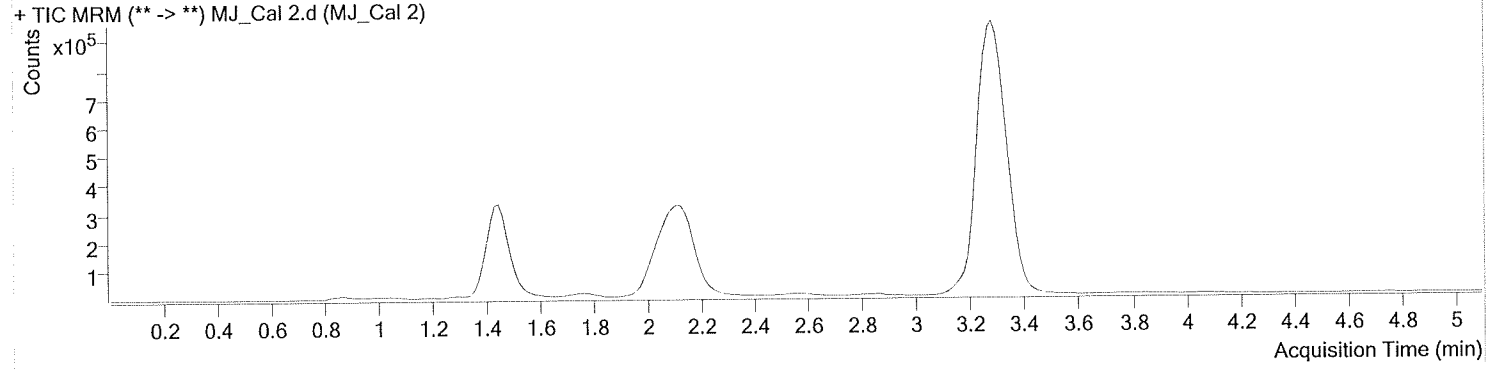


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
Calibration Last Update 8/7/2020 9:36:29 AM

Instrument	Falco	Data File	MJ_Cal 2.d
Type	Cal	Sample	MJ_Cal 2
Acq. Method	AM 27 THC quant.m	Operator	Sarah Pickle
Sample Position	P3-G6	Comment	
Injection Volume	10		
Acq. Date-Time	8/6/2020 12:52:28 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.483	131943	∞	8.8	∞	1240645	2.7677 ng/ml Low
THC-COOH	1.474	101107	∞	49.9	∞	422635	10.2475 ng/ml
THC	3.300	188240	1395.24	28.6	∞	7254462	2.8932 ng/ml Low

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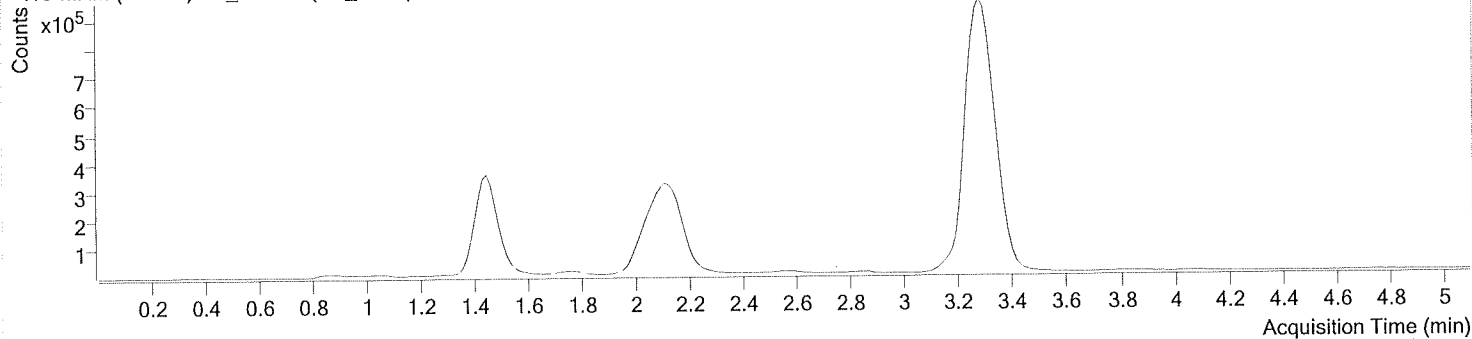
AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
Calibration Last Update 8/7/2020 9:36:29 AM

Instrument	Falco	Data File	MJ_Cal 3.d
Type	Cal	Sample	MJ_Cal 3
Acq. Method	AM 27 THC quant.m	Operator	Sarah Pickle
Sample Position	P3-F6	Comment	
Injection Volume	10		
Acq. Date-Time	8/6/2020 1:00:03 PM		

Sample Chromatogram

+ TIC MRM (** -> **) MJ_Cal 3.d (MJ_Cal 3)



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	178379	∞	8.8	∞	1240873	4.9727 ng/ml
THC-COOH	1.474	175245	∞	61.8	∞	417053	18.4275 ng/ml
THC	3.300	318582	1122.93	27.8	∞	7214494	4.7724 ng/ml

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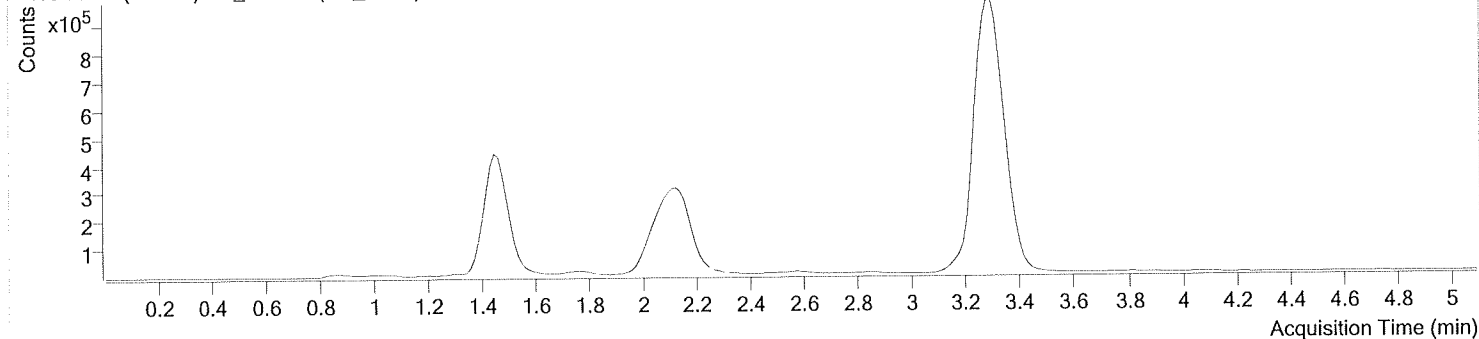
AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
Calibration Last Update 8/7/2020 9:36:29 AM

Instrument	Falco	Data File	MJ_Cal 4.d
Type	Cal	Sample	MJ_Cal 4
Acq. Method	AM 27 THC quant.m	Operator	Sarah Pickle
Sample Position	P3-E6	Comment	
Injection Volume	10		
Acq. Date-Time	8/6/2020 1:07:37 PM		

Sample Chromatogram

+ TIC MRM (** -> **) MJ_Cal 4.d (MJ_Cal 4)



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	305631	∞	9.8	333.16	1221181	11.2523 ng/ml
THC-COOH	1.474	456336	∞	57.6	∞	409884	49.7576 ng/ml
THC	3.300	646176	7176.54	26.3	353.09	7013444	9.7231 ng/ml

SJ P



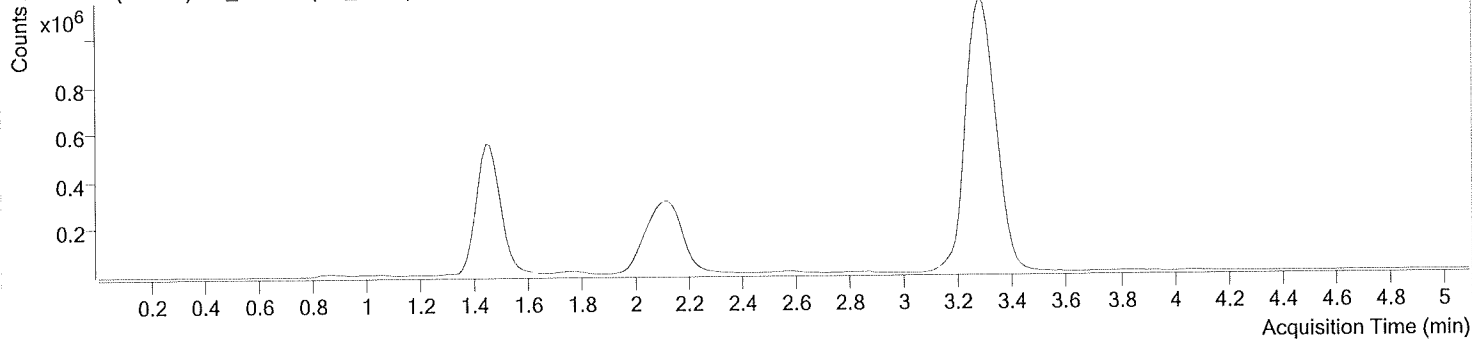
AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
Calibration Last Update 8/7/2020 9:36:29 AM

Instrument	Falco	Data File	MJ_Cal 5.d
Type	Cal	Sample	MJ_Cal 5
Acq. Method	AM 27 THC quant.m	Operator	Sarah Pickle
Sample Position	P3-D6	Comment	
Injection Volume	10		
Acq. Date-Time	8/6/2020 1:15:11 PM		

Sample Chromatogram

+ TIC MRM (** -> **) MJ_Cal 5.d (MJ_Cal 5)



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	566769	∞	12.2	∞	1213969	24.0210 ng/ml
THC-COOH	1.474	633201	∞	61.1	∞	403406	70.3831 ng/ml
THC	3.300	1642714	3829.43	26.1	∞	7003020	24.4218 ng/ml

SJ B



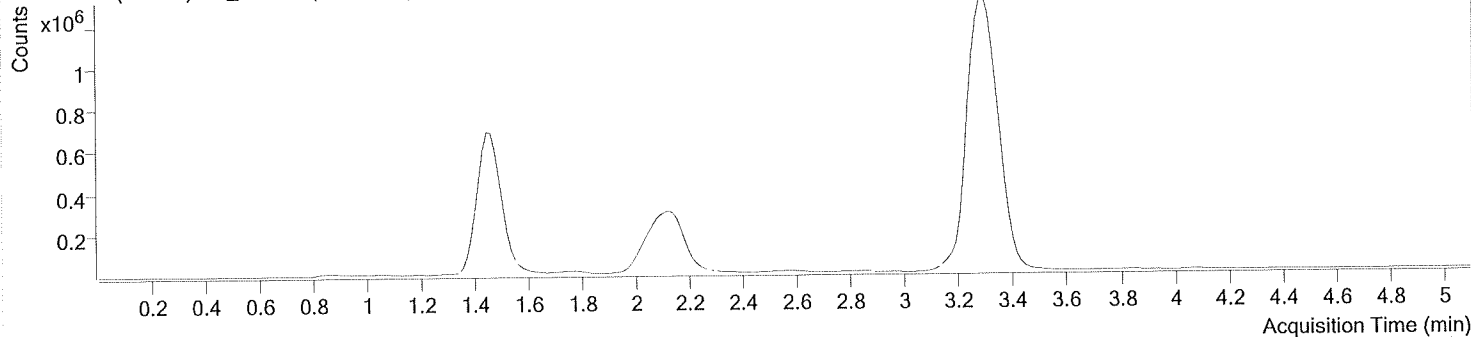
AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
Calibration Last Update 8/7/2020 9:36:29 AM

Instrument	Falco	Data File	MJ_Cal 6.d
Type	Cal	Sample	MJ_Cal 6
Acq. Method	AM 27 THC quant.m	Operator	Sarah Pickle
Sample Position	P3-C6	Comment	
Injection Volume	10		
Acq. Date-Time	8/6/2020 1:22:46 PM		

Sample Chromatogram

+ TIC MRM (** -> **) MJ_Cal 6.d (MJ_Cal 6)



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	1070947	∞	12.7	∞	1186801	49.6949 ng/ml
THC-COOH	1.474	866738	∞	57.2	∞	385005	101.1920 ng/ml
THC	3.300	3178991	15991.00	26.2	∞	6665732	49.4301 ng/ml

SJ
D

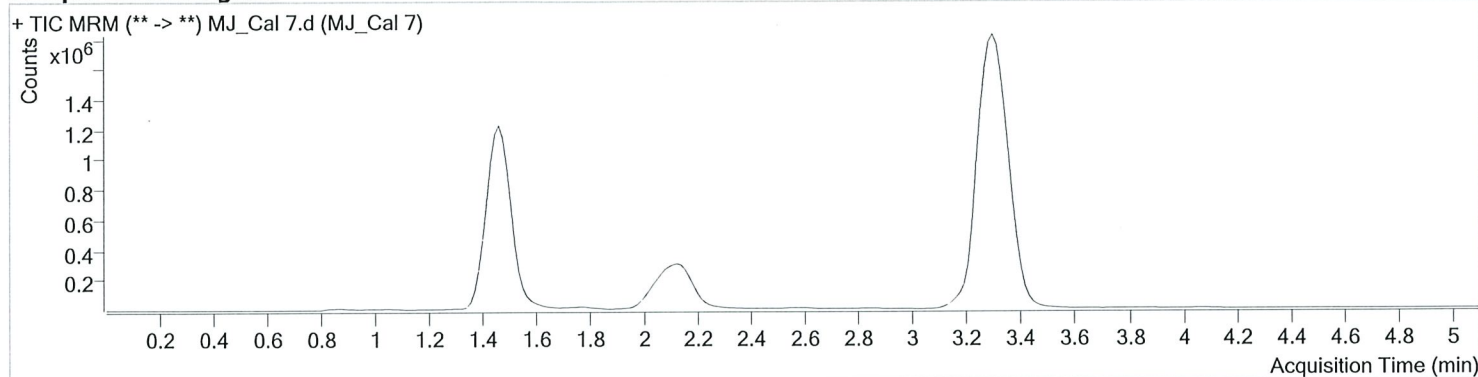


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\080620 AM 27 28 SJ SP\QuantResults\AM 27 SJ SP.batch.bin
Calibration Last Update 8/7/2020 9:36:29 AM

Instrument	Falco	Data File	MJ_Cal 7.d
Type	Cal	Sample	MJ_Cal 7
Acq. Method	AM 27 THC quant.m	Operator	Sarah Pickle
Sample Position	P3-B6	Comment	
Injection Volume	10		
Acq. Date-Time	8/6/2020 1:30:21 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	2087849	∞	12.8	∞	1185836	100.2914 ng/ml
THC-COOH	1.474	2092023	∞	58.4	∞	370673	254.5414 ng/ml
THC	3.300	6499033	3119.23	26.3	∞	6612894	101.6324 ng/ml