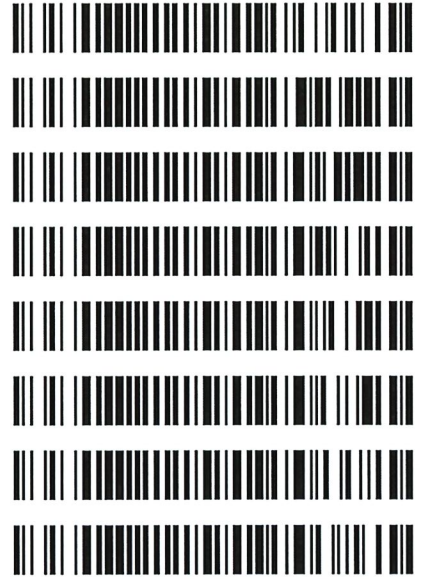


REVIEWED
By Sarah Pickle at 3:29 pm, Jul 01, 2020

B 6/26/2020

Worklist: 4323

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
P2020-1500	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1626	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1668	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1670	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1671	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1673	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1674	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1677	1	BCK	AM 27 Blood THC Quant by LC-QQQ



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

15

Extraction Date: 06/29/2020
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 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 (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*

AM #27 Cannabinoid Quant. Results

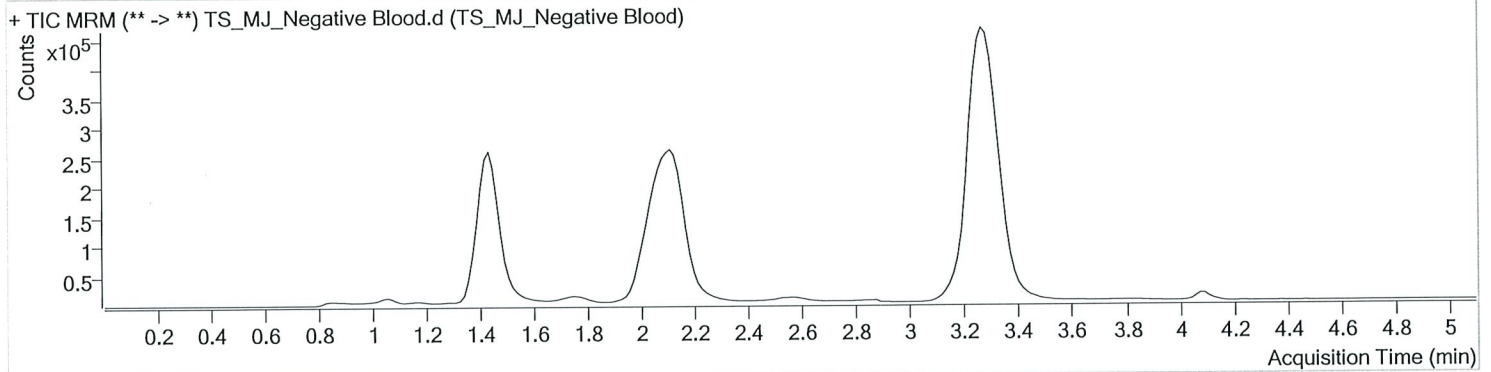
13



Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
Calibration Last Update 7/1/2020 1:27:56 PM

Instrument	Falco	Data File	TS_MJ_Negative Blood.d
Type	Sample	Sample	TS_MJ_Negative Blood
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P4-H5	Comment	
Injection Volume	10		
Acq. Date-Time	6/30/2020 1:33:54 PM		
Sample Info.			

Sample Chromatogram



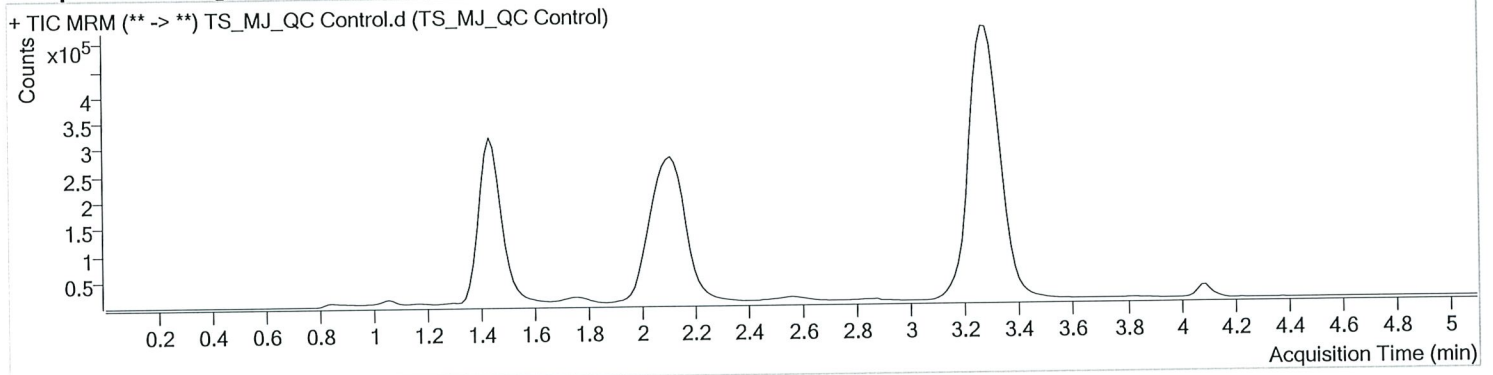


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
Calibration Last Update 7/1/2020 1:27:56 PM

Instrument	Falco	Data File	TS_MJ_QC Control.d
Type	Sample	Sample	TS_MJ_QC Control
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P4-A6	Comment	
Injection Volume	10		
Acq. Date-Time	6/30/2020 1:18:40 PM		

Sample Chromatogram



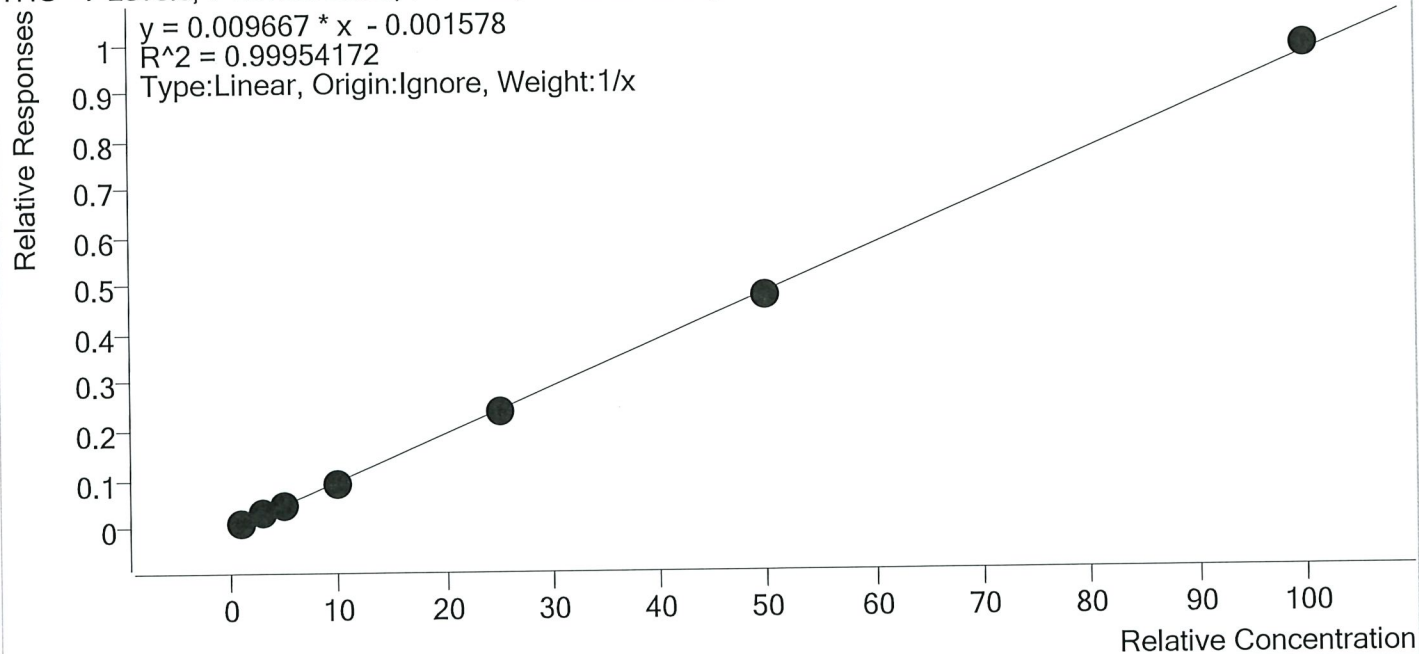
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	150895	∞	9.1	737.80	1170799	4.2719 ng/ml
THC-COOH	1.474	104806	∞	58.0	∞	337838	14.5515 ng/ml
THC	3.285	158589	849.51	26.5	280.75	3983043	4.2822 ng/ml



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
Last Cal. Update 7/1/2020 1:27 PM
Analyst Name ISP\Datastor
Analyte THC **Internal Standard** THC-D3

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



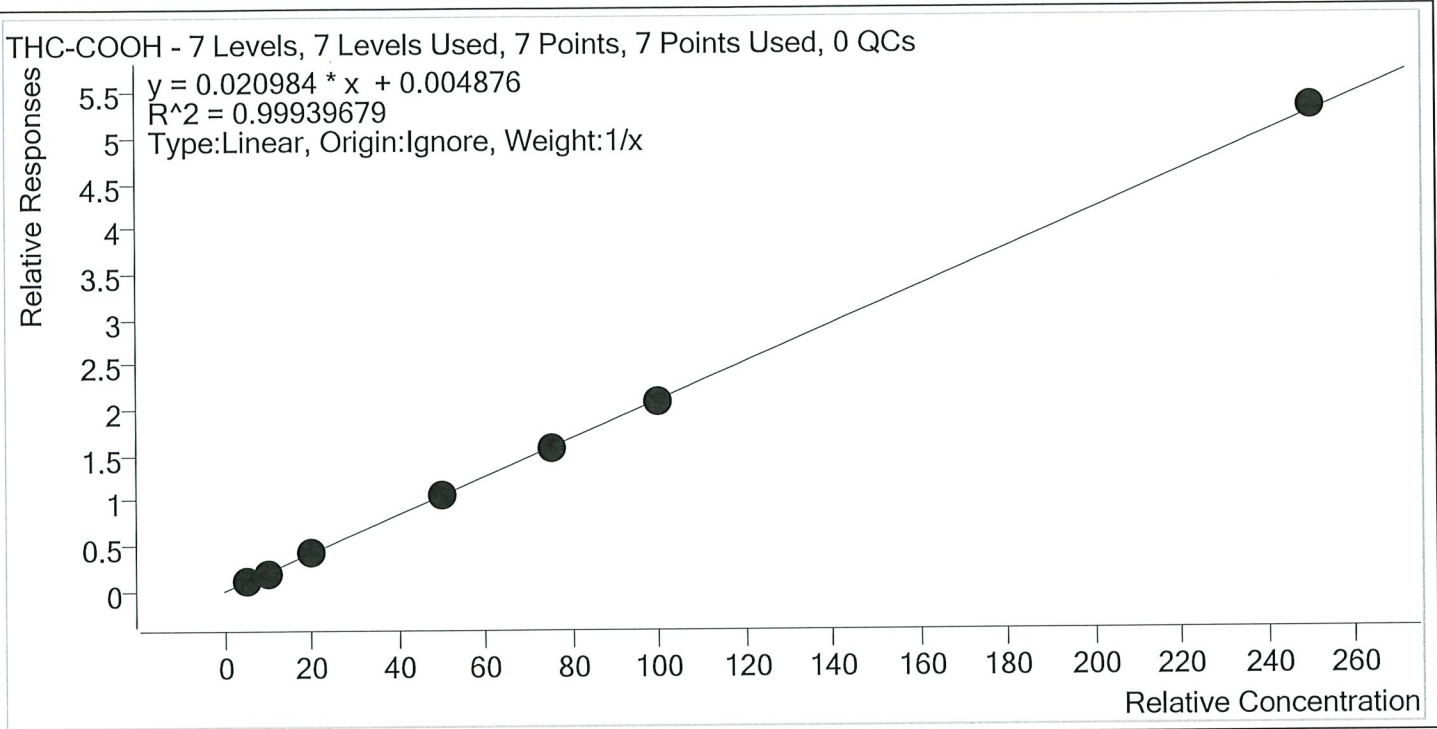
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
TS MJ Cal 1	1	✓	1.0	1.1	109.9
TS MJ Cal 2	2	✓	3.0	2.9	96.8
TS MJ Cal 3	3	✓	5.0	4.9	98.4
TS MJ Cal 4	4	✓	10.0	9.7	97.3
TS MJ Cal 5	5	✓	25.0	24.3	97.2
TS MJ Cal 6	6	✓	50.0	49.4	98.8
TS MJ Cal 7	7	✓	100.0	101.7	101.7

TS



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
Last Cal. Update 7/1/2020 1:27 PM
Analyst Name ISP\Datastor
Analyte THC-COOH **Internal Standard** THC-COOH-D9



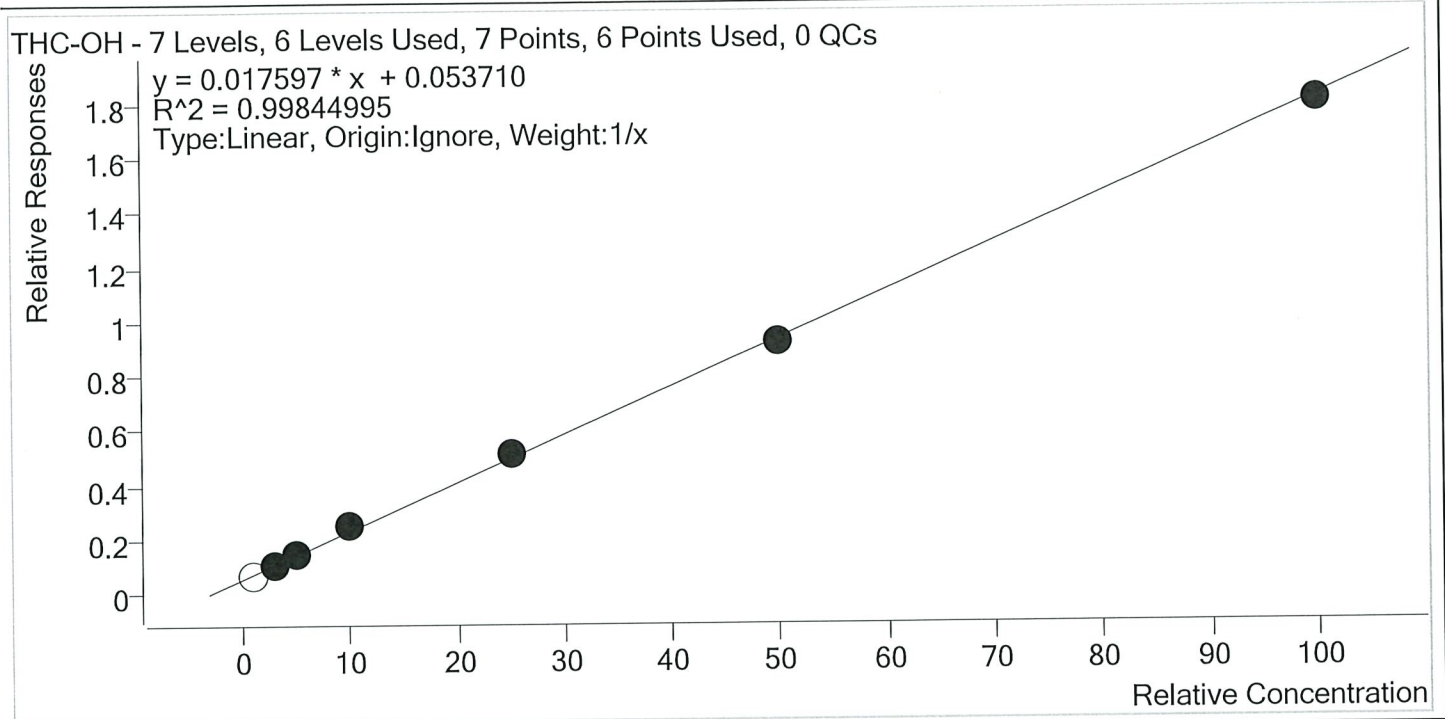
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
TS MJ Cal 1	1	✓	5.0	5.4	107.2
TS MJ Cal 2	2	✓	10.0	9.1	91.1
TS MJ Cal 3	3	✓	20.0	20.9	104.6
TS MJ Cal 4	4	✓	50.0	49.6	99.1
TS MJ Cal 5	5	✓	75.0	73.9	98.6
TS MJ Cal 6	6	✓	100.0	98.2	98.2
TS MJ Cal 7	7	✓	250.0	252.9	101.2

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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
 Last Cal. Update 7/1/2020 1:27 PM
 Analyst Name ISP\Datastor
 Analyte THC-OH Internal Standard THC-OH-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
TS MJ Cal 1	1	×	1.0	0.8	82.1
TS MJ Cal 2	2	✓	3.0	2.7	90.0
TS MJ Cal 3	3	✓	5.0	4.9	98.0
TS MJ Cal 4	4	✓	10.0	11.0	109.6
TS MJ Cal 5	5	✓	25.0	26.3	105.2
TS MJ Cal 6	6	✓	50.0	49.0	98.1
TS MJ Cal 7	7	✓	100.0	99.1	99.1

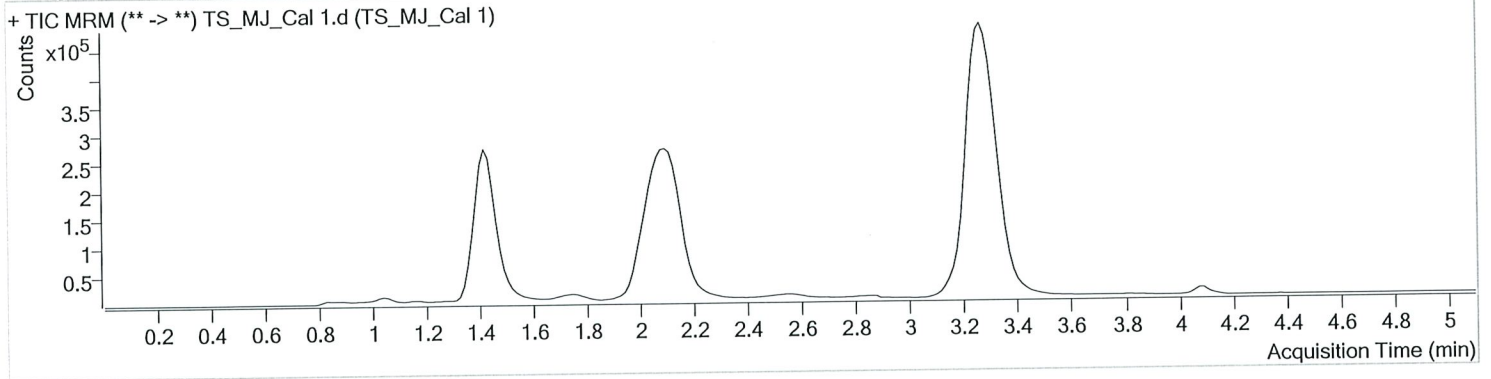
AM #27 Cannabinoid Quant. Results



Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
 Calibration Last Update 7/1/2020 1:27:56 PM

Instrument	Falco	Data File	TS_MJ_Cal 1.d
Type	Cal	Sample	TS_MJ_Cal 1
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P4-H6	Comment	
Injection Volume	10		
Acq. Date-Time	6/30/2020 12:17:46 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.483	76125	∞	6.4 Low	25.72	1116772	0.8214 ng/ml Low
THC-COOH	1.459	37410	34.92	47.9	178.56	318694	5.3617 ng/ml Low
THC	3.285	35098	273.61	29.5	11.08	3879599	1.0991 ng/ml Low

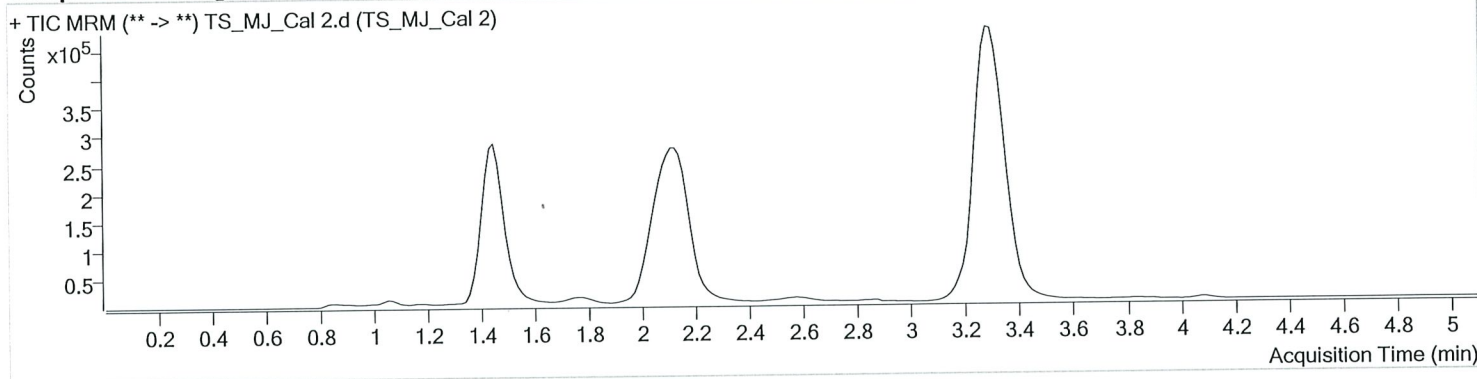


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
 Calibration Last Update 7/1/2020 1:27:56 PM

Instrument	Falco	Data File	TS_MJ_Cal 2.d
Type	Cal	Sample	TS_MJ_Cal 2
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P4-G6	Comment	
Injection Volume	10		
Acq. Date-Time	6/30/2020 12:25:30 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.483	113326	∞	9.5	49.77	1119832	2.6988 ng/ml Low
THC-COOH	1.474	62149	∞	59.2	∞	317057	9.1090 ng/ml Low
THC	3.300	97954	616.79	28.0	90.55	3698748	2.9029 ng/ml Low

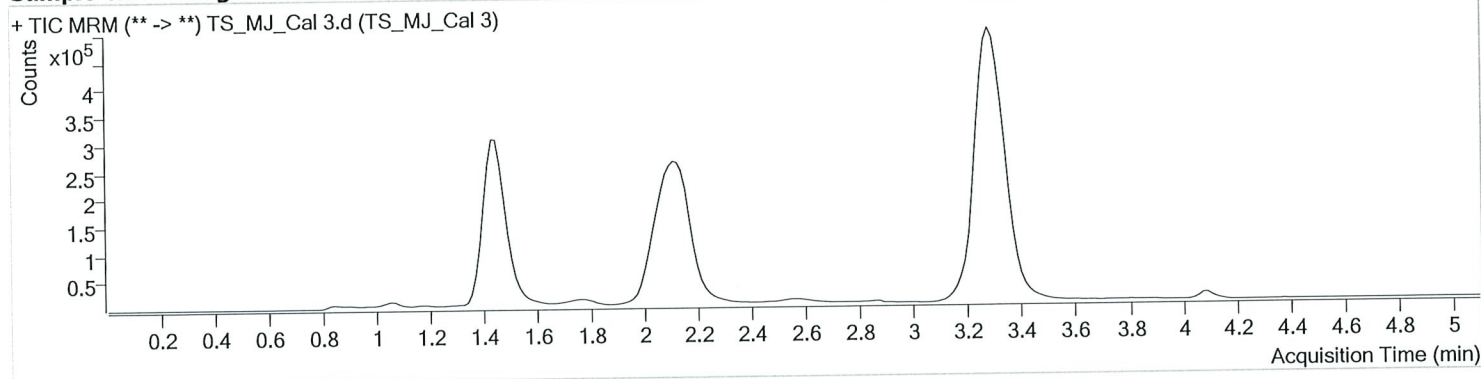


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
Calibration Last Update 7/1/2020 1:27:56 PM

Instrument	Falco	Data File	TS_MJ_Cal 3.d
Type	Cal	Sample	TS_MJ_Cal 3
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P4-F6	Comment	
Injection Volume	10		
Acq. Date-Time	6/30/2020 12:33:06 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	155718	∞	9.3	74.44	1112620	4.9012 ng/ml
THC-COOH	1.474	138756	∞	54.6	765.06	312588	20.9215 ng/ml
THC	3.300	169728	∞	27.7	∞	3692534	4.9183 ng/ml

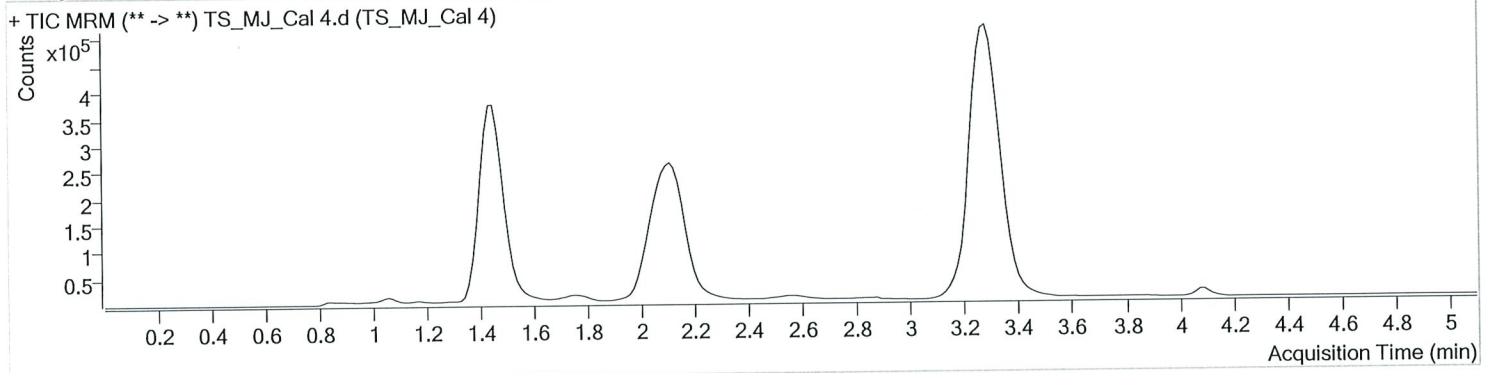


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
Calibration Last Update 7/1/2020 1:27:56 PM

Instrument	Falco	Data File	TS_MJ_Cal 4.d
Type	Cal	Sample	TS_MJ_Cal 4
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P4-E6	Comment	
Injection Volume	10		
Acq. Date-Time	6/30/2020 12:40:42 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	275110	∞	9.8	253.07	1115609	10.9617 ng/ml
THC-COOH	1.474	325237	∞	59.9	∞	311241	49.5658 ng/ml
THC	3.285	341166	∞	25.7	436.09	3688048	9.7329 ng/ml

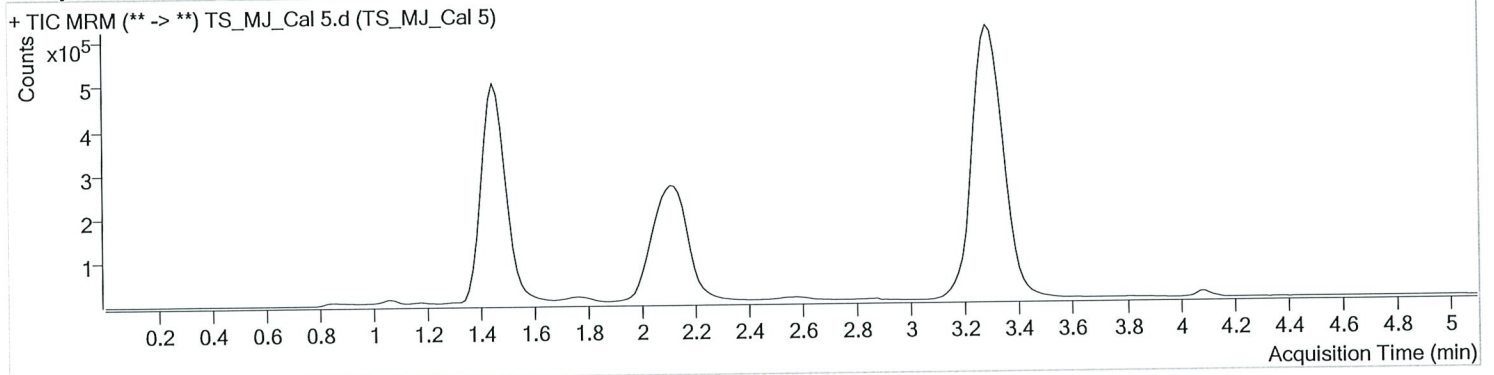


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
Calibration Last Update 7/1/2020 1:27:56 PM

Instrument	Falco	Data File	TS_MJ_Cal 5.d
Type	Cal	Sample	TS_MJ_Cal 5
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P4-D6	Comment	
Injection Volume	10		
Acq. Date-Time	6/30/2020 12:48:18 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	613722	∞	11.4	∞	1187827	26.3096 ng/ml
THC-COOH	1.474	511350	∞	63.1	∞	328589	73.9289 ng/ml
THC	3.300	907791	1765.22	25.5	614.48	3891369	24.2963 ng/ml

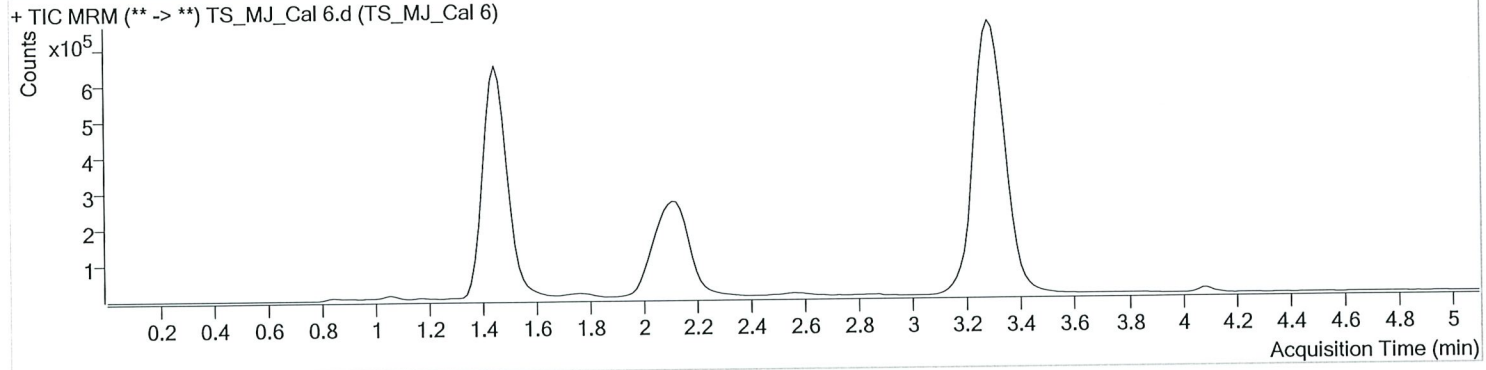


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
Calibration Last Update 7/1/2020 1:27:56 PM

Instrument	Falco	Data File	TS_MJ_Cal 6.d
Type	Cal	Sample	TS_MJ_Cal 6
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P4-C6	Comment	
Injection Volume	10		
Acq. Date-Time	6/30/2020 12:55:53 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	1092950	∞	13.0	286.28	1192474	49.0332 ng/ml
THC-COOH	1.474	678554	∞	62.9	∞	328515	98.2005 ng/ml
THC	3.300	1841940	3998.46	25.6	∞	3870148	49.3987 ng/ml

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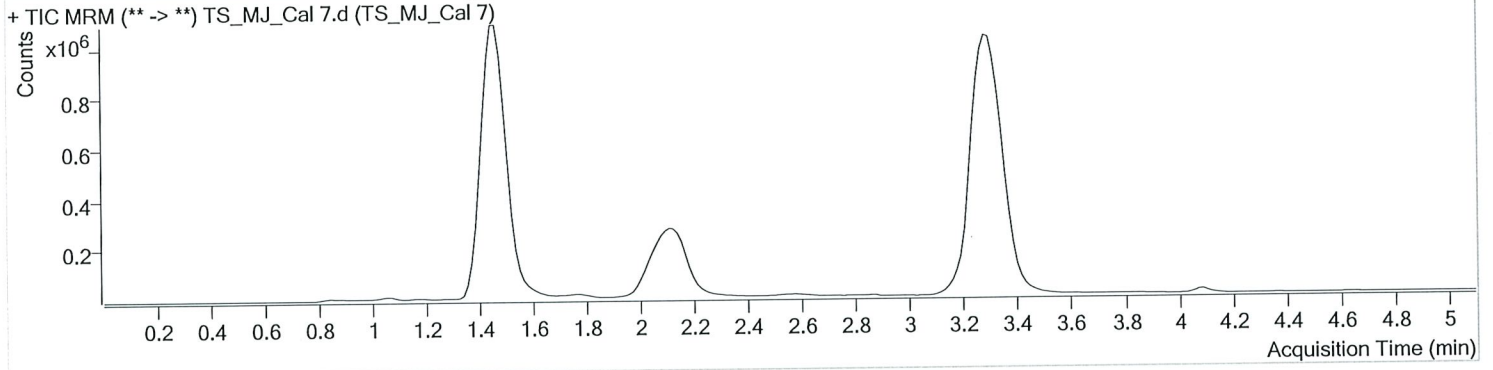


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\062920 AM 27_28 CS TS\QuantResults\TS THCQ.batch.bin
Calibration Last Update 7/1/2020 1:27:56 PM

Instrument	Falco	Data File	TS_MJ_Cal 7.d
Type	Cal	Sample	TS_MJ_Cal 7
Acq. Method	AM 27 THC quant.m	Operator	Tamara Salazar
Sample Position	P4-B6	Comment	
Injection Volume	10		
Acq. Date-Time	6/30/2020 1:03:28 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	2106905	∞	12.9	∞	1172144	99.0955 ng/ml
THC-COOH	1.474	1643765	∞	64.0	∞	309443	252.9126 ng/ml
THC	3.300	3773123	11165.91	25.2	1428.78	3846039	101.6519 ng/ml