

REVIEWED

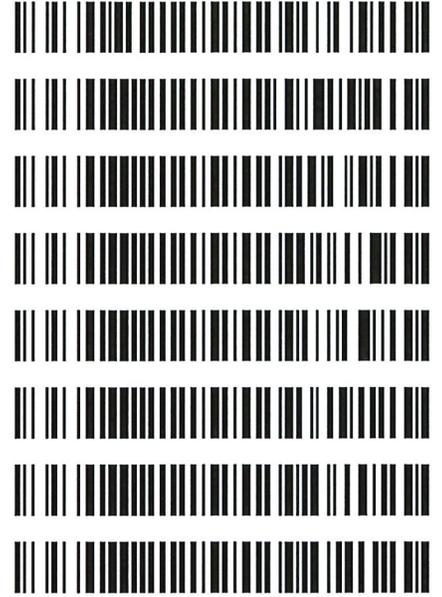
By Celena Shrum at 3:29 pm, Oct 23, 2019

10/11/2019

TS

Worklist: 3753

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
M2019-4169	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2019-2672	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2019-2803	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2019-2857	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2019-2911	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2019-2936	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2019-2959	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2019-2960	1	BCK	AM 27 Blood THC Quant by LC-QQQ



B

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

Extraction Date: 10/15/19
Plate lot#: IDP-108-190716

Analyst: Tamara Salazar
Plate Expiration: 01/16/2020

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 445283-3
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/urine (calibrated pipette) Pipette ID: 3** in wells of analytical (standards) plate.
- 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes. *Shaker ID: 067105*
- 4. Pipette **500µL 0.1% formic acid in water** in wells of analytical plate for blood samples.
- 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) Manifold ID: 067104
- 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.
SPE Dry ID: 067103
- 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.
Worklist path: D:\MassHunter\Data\2019\AM 27\101519 MDQ P1 and P2_THCQ TS
Batch Name: THCQ wklst 3753 TS
- 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: 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: THC-OH 3-100 TS

TS

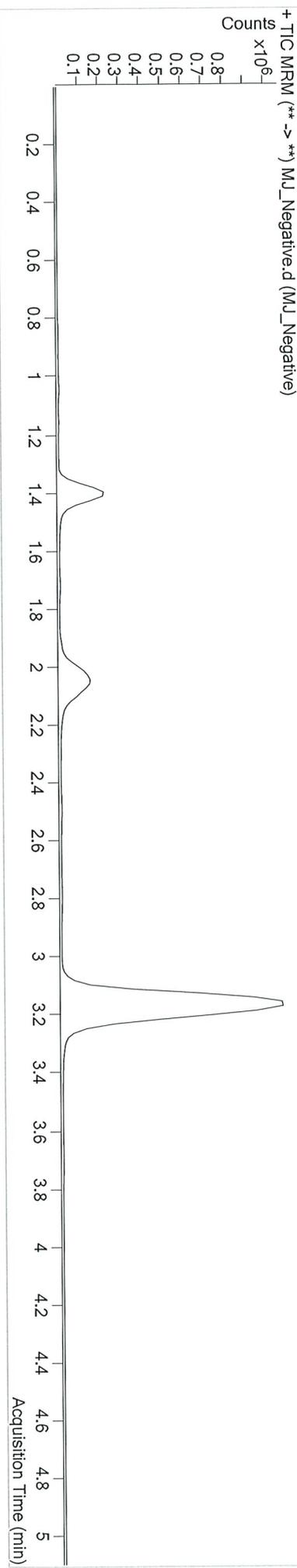
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wk1st 3753 TS.batch.bin
Calibration Last Update 10/16/2019 8:58:41 AM

Instrument	Falco	Data File	MJ_Negative.d
Type	Sample	Sample	MJ_Negative
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-A2		
Injection Volume	10		
Acq. Date-Time	10/16/2019 4:04:40 AM		
Sample Info.			

Sample Chromatogram



TS

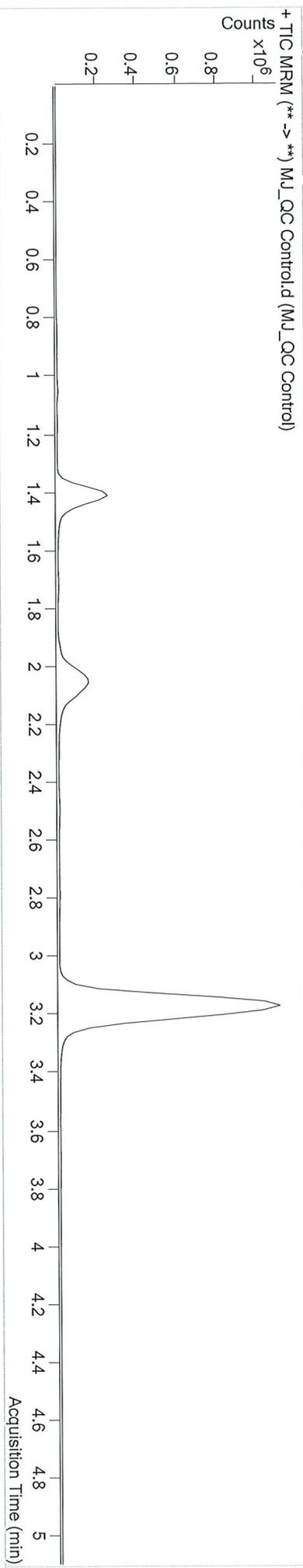
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wk1st 3753 TS.batch.bin
Calibration Last Update 10/16/2019 8:58:41 AM

Instrument	Falco	Data File	MJ_QC Control.d
Type	Sample	Sample	MJ_QC Control
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-H1		
Injection Volume	10		
Acq. Date-Time	10/16/2019 3:49:29 AM		
Sample Info.			

Sample Chromatogram



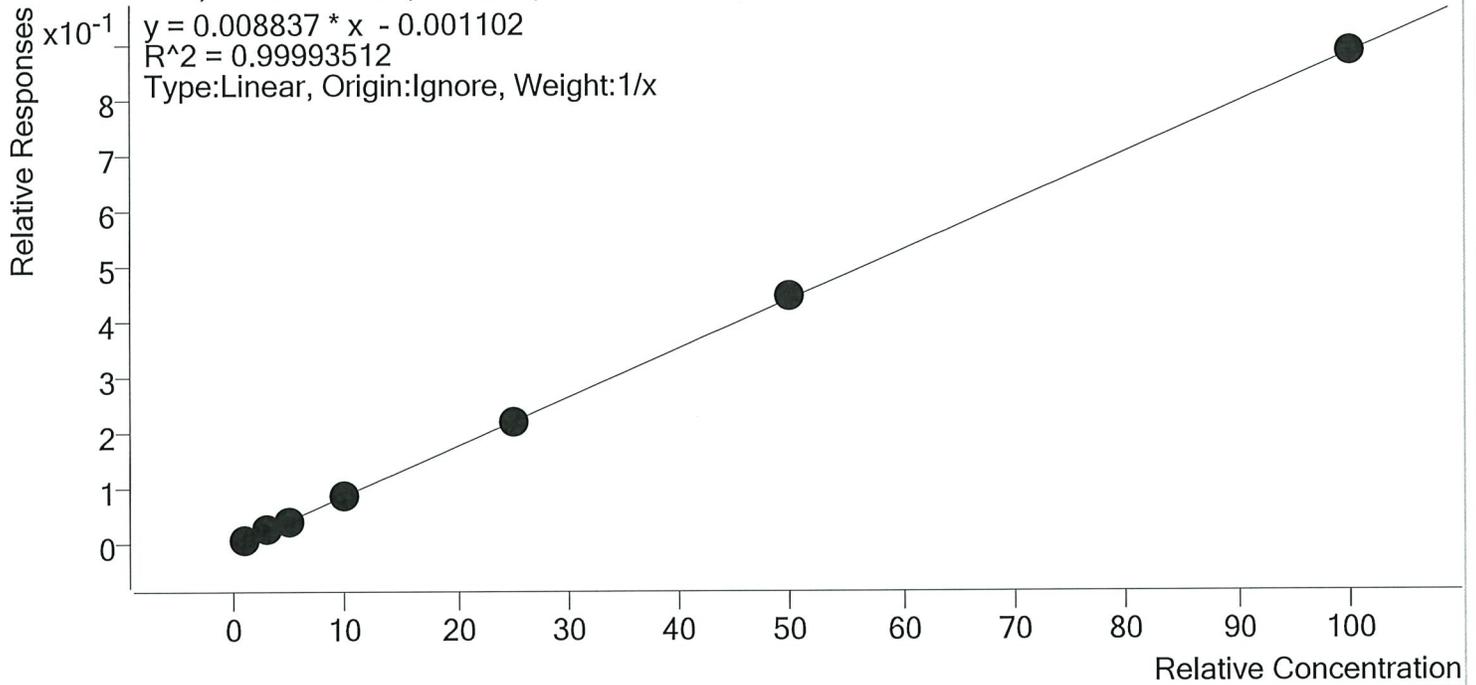
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.194	218961	1279.67	27.6	416.82	5854606	4.3567 ng/ml
THC-COOH	1.444	75620	502.94	59.4	987.79	213886	13.9780 ng/ml
THC-OH	1.423	55109	∞	11.3	135.03	728413	4.4302 ng/ml



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wk1st 3753 TS.batch.bin
Last Cal. Update 10/17/2019 10:15 AM
Analyst Name ISP\datastor
Analyte THC **Internal Standard** THC-D3

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

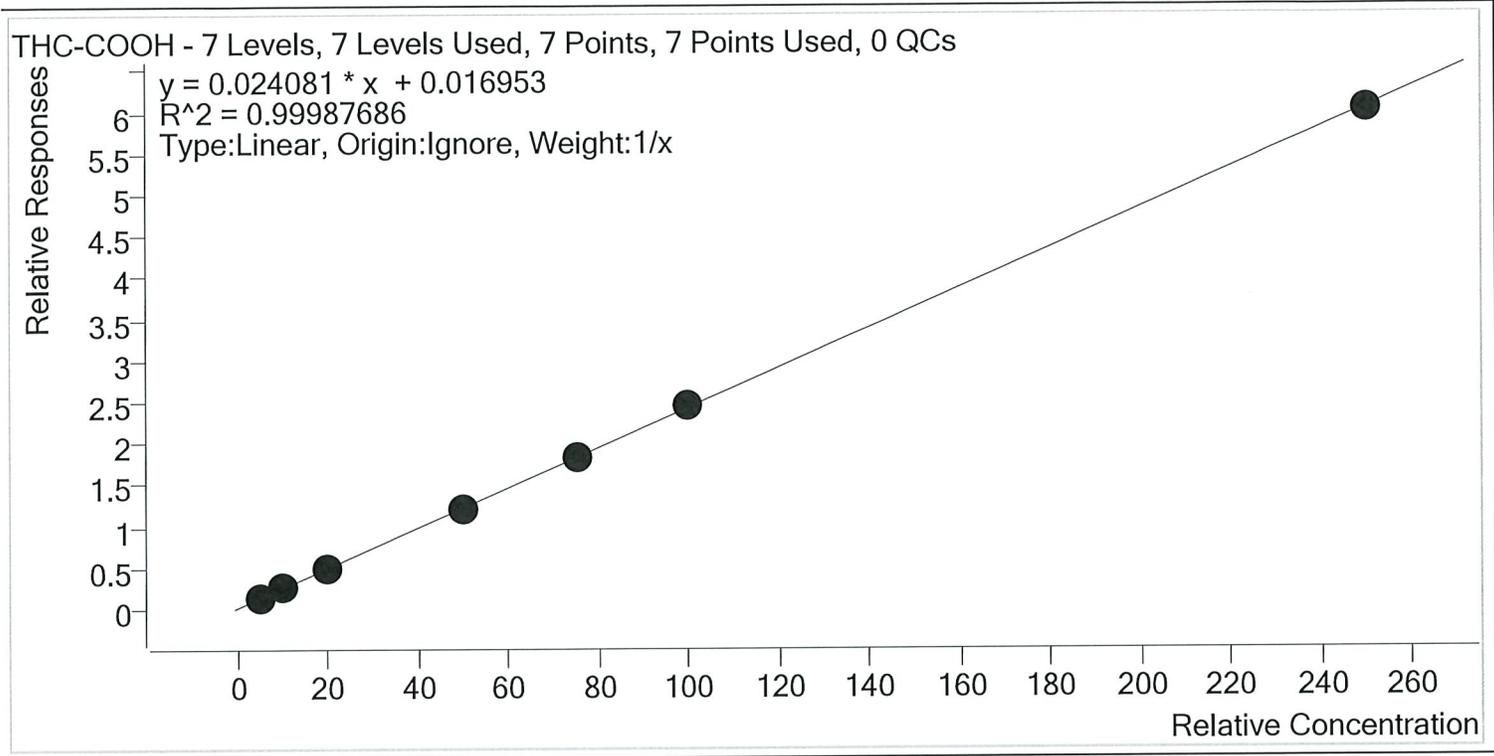


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ Cal 1	1	✓	1.0	1.0	104.9
MJ Cal 2	2	✓	3.0	2.9	98.1
MJ Cal 3	3	✓	5.0	4.9	98.6
MJ Cal 4	4	✓	10.0	9.9	98.7
MJ Cal 5	5	✓	25.0	24.7	99.0
MJ Cal 6	6	✓	50.0	50.3	100.6
MJ Cal 7	7	✓	100.0	100.2	100.2



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wklst
 3753 TS.batch.bin
Last Cal. Update 10/17/2019 10:15 AM
Analyst Name ISP\datastor
Analyte THC-COOH **Internal Standard** THC-COOH-D9

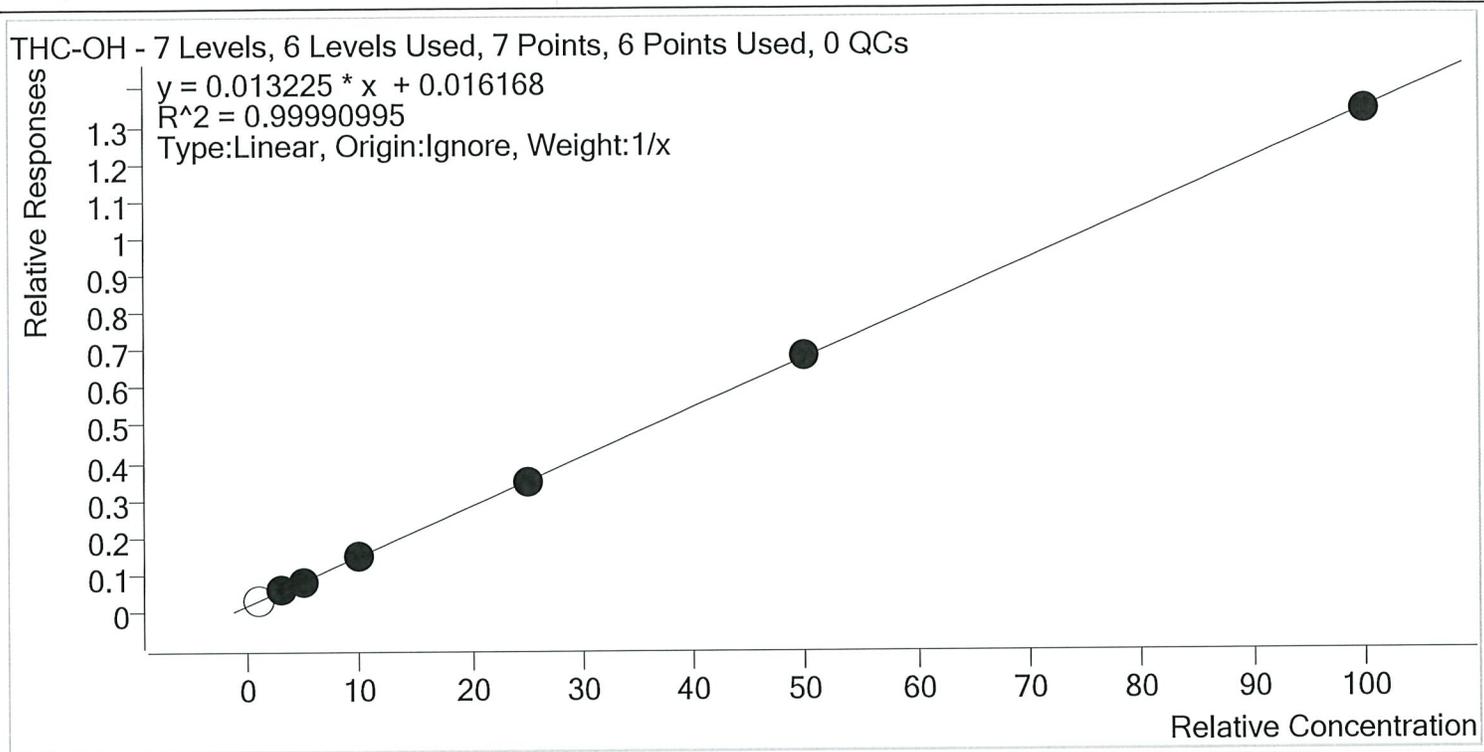


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ Cal 1	1	✓	5.0	5.1	101.8
MJ Cal 2	2	✓	10.0	9.7	96.6
MJ Cal 3	3	✓	20.0	20.3	101.5
MJ Cal 4	4	✓	50.0	49.8	99.6
MJ Cal 5	5	✓	75.0	74.5	99.4
MJ Cal 6	6	✓	100.0	101.5	101.5
MJ Cal 7	7	✓	250.0	249.1	99.6



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wklst
 3753 TS.batch.bin
Last Cal. Update 10/17/2019 10:15 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	1.1	111.7
MJ Cal 2	2	✓	3.0	2.9	98.1
MJ Cal 3	3	✓	5.0	4.9	98.6
MJ Cal 4	4	✓	10.0	10.3	102.7
MJ Cal 5	5	✓	25.0	25.2	100.8
MJ Cal 6	6	✓	50.0	50.1	100.2
MJ Cal 7	7	✓	100.0	99.6	99.6

13

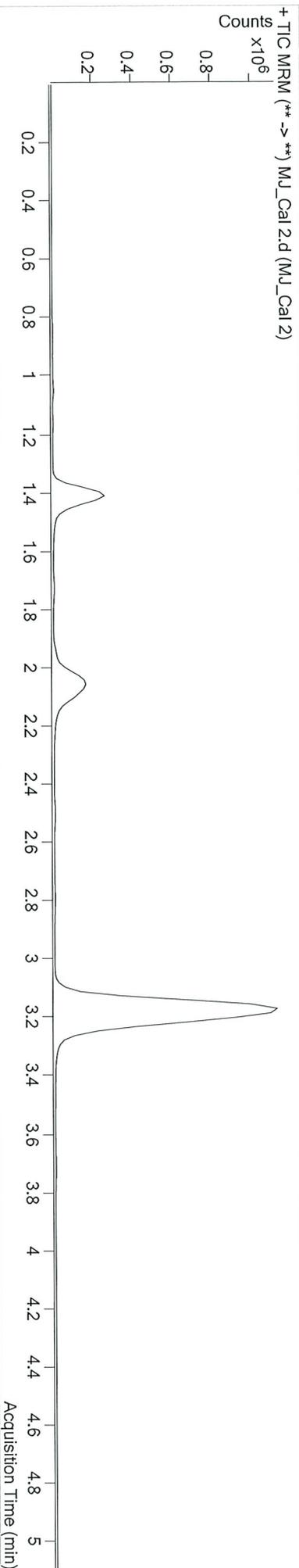
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wk1st 3753 TS.batch.bin
Calibration Last Update 10/16/2019 8:58:41 AM

Instrument	Falco	Data File	MJ_Cal 2.d
Type	Cal	Sample	MJ_Cal 2
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-B1		
Injection Volume	10		
Acq. Date-Time	10/16/2019 3:03:54 AM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.194	149434	509.84	28.6	519.64	5998322	2.9437 ng/ml
THC-COOH	1.444	55286	378.18	57.8	1046.76	221512	9.6605 ng/ml
THC-OH	1.423	41905	∞	11.2	92.65	760580	2.8721 ng/ml

TS

AM #27 Cannabinoids Quant. Results

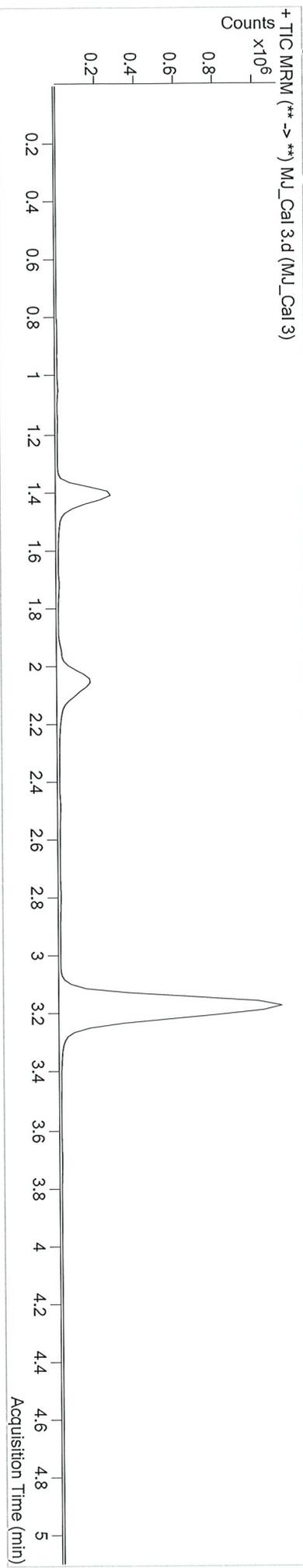


Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wk1st 3753 TS_batch.bin
Calibration Last Update 10/16/2019 8:58:41 AM

Instrument Falco
Type Cal
Acq. Method AM 27 THC quant.m
Sample Position P3-C1
Injection Volume 10
Acq. Date-Time 10/16/2019 3:11:30 AM
Sample Info.

Data File MJ_Cal 3.d
Sample MJ_Cal 3
Comment

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.194	233050	731.76	27.7	509.29	5488124	4.9298 ng/ml
THC-COOH	1.444	103235	599.32	59.1	∞	204199	20.2904 ng/ml
THC-OH	1.423	56829	∞	11.2	397.74	698158	4.8653 ng/ml

TS

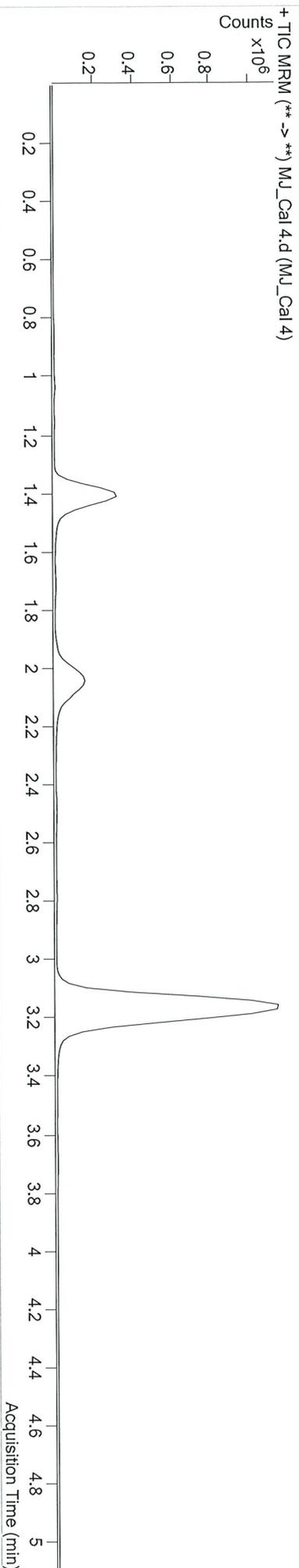
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wk1st 3753 TS.batch.bin
Calibration Last Update 10/16/2019 8:58:41 AM

Instrument	Falco	Data File	MJ_Cal 4.d
Type	Cal	Sample	MJ_Cal 4
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-D1		
Injection Volume	10		
Acq. Date-Time	10/16/2019 3:19:07 AM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.179	525571	4157.81	26.6	409.72	6104303	9.8672 ng/ml
THC-COOH	1.429	267072	442.36	60.8	3069.82	219554	49.8105 ng/ml
THC-OH	1.408	115679	∞	12.6	∞	760779	10.2195 ng/ml

TS

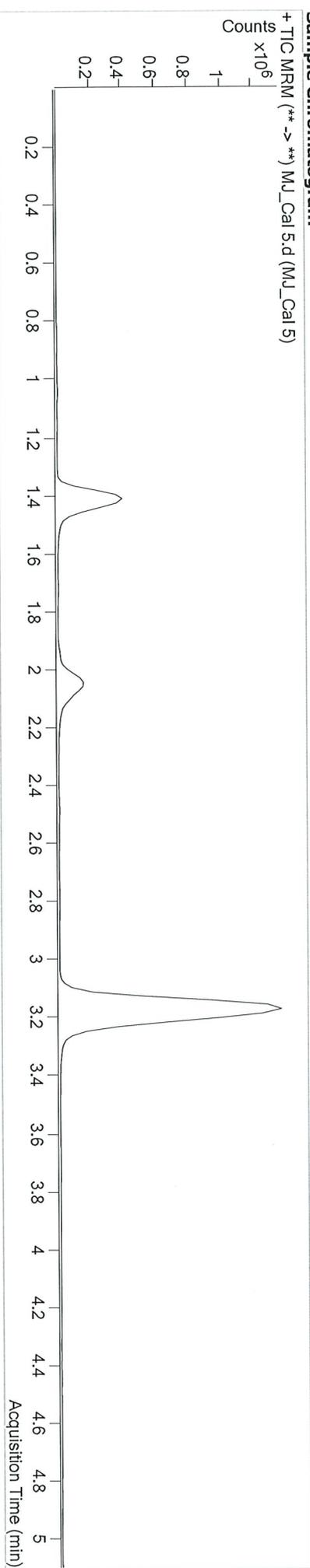
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wk1st 3753 TS.batch.bin
Calibration Last Update 10/16/2019 8:58:41 AM

Instrument	Falco	Data File	MJ_Cal 5.d
Type	Cal	Sample	MJ_Cal 5
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-E1		
Injection Volume	10		
Acq. Date-Time	10/16/2019 3:26:44 AM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.194	1226241	∞	26.7	∞	5635825	24.7451 ng/ml
THC-COOH	1.429	367991	2046.47	61.4	2910.70	203138	74.5233 ng/ml
THC-OH	1.408	244776	∞	12.9	734.89	700791	25.1658 ng/ml

15

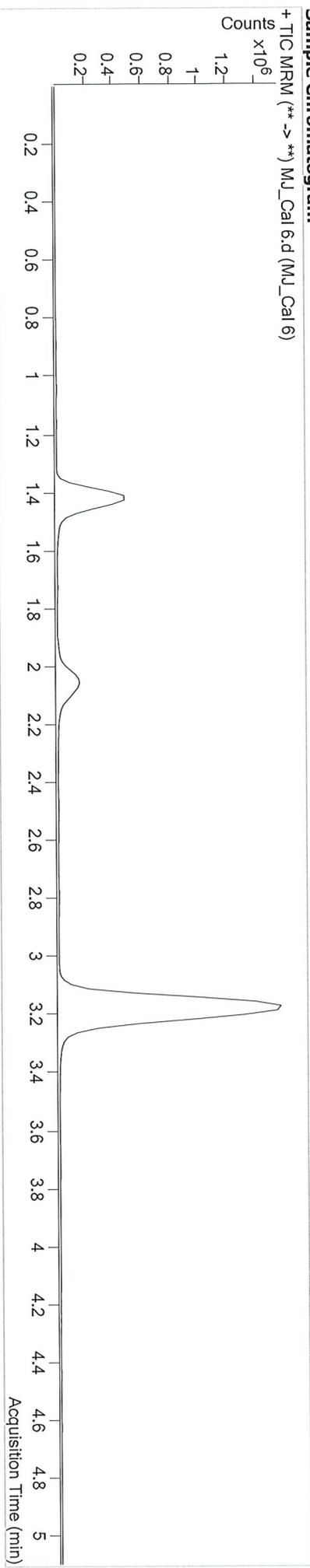
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wk1st 3753 TS.batch.bin
Calibration Last Update 10/16/2019 8:58:41 AM

Instrument	Falco	Data File	MJ_Cal 6.d
Type	Cal	Sample	MJ_Cal 6
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-F1		
Injection Volume	10		
Acq. Date-Time	10/16/2019 3:34:19 AM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.194	2497902	17703.35	27.0	3711.15	5635220	50.2830 ng/ml
THC-COOH	1.444	493609	54984.93	60.3	4191.83	200510	101.5259 ng/ml
THC-OH	1.423	483400	∞	13.8	954.12	712534	50.1079 ng/ml

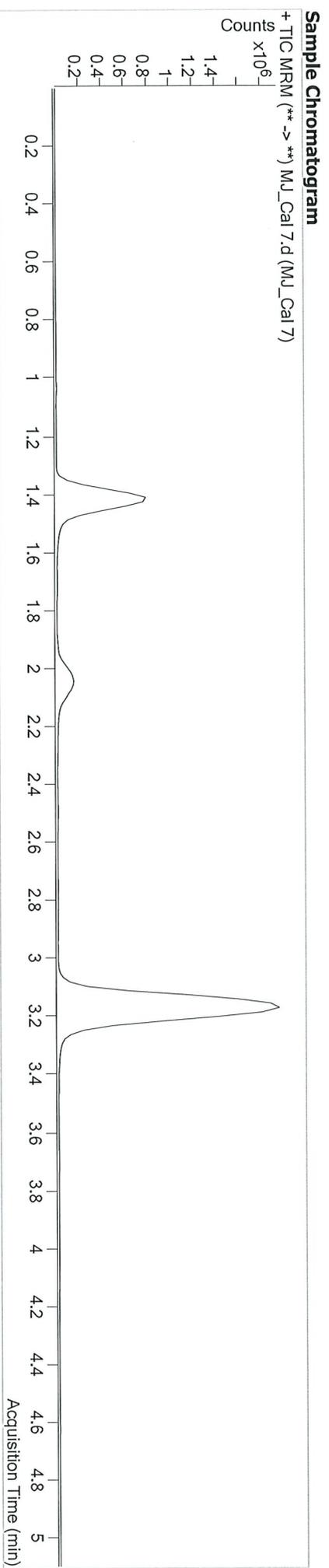
15

AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCOQ TS\QuantResults\THCOQ wk1st 3753 TS.batch.bin
Calibration Last Update 10/16/2019 8:58:41 AM

Instrument	Falco	Data File	MJ_Cal 7.d
Type	Cal	Sample	MJ_Cal 7
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-G1		
Injection Volume	10		
Acq. Date-Time	10/16/2019 3:41:54 AM		
Sample Info.			



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.179	4785137	∞	26.7	∞	5411553	100.1824 ng/ml
THC-COOH	1.429	1135572	7821.75	62.1	6754.94	188776	249.0997 ng/ml
THC-OH	1.408	920534	∞	13.7	2618.18	690464	99.7278 ng/ml



AM #27 Cannabinoids Quant. Results

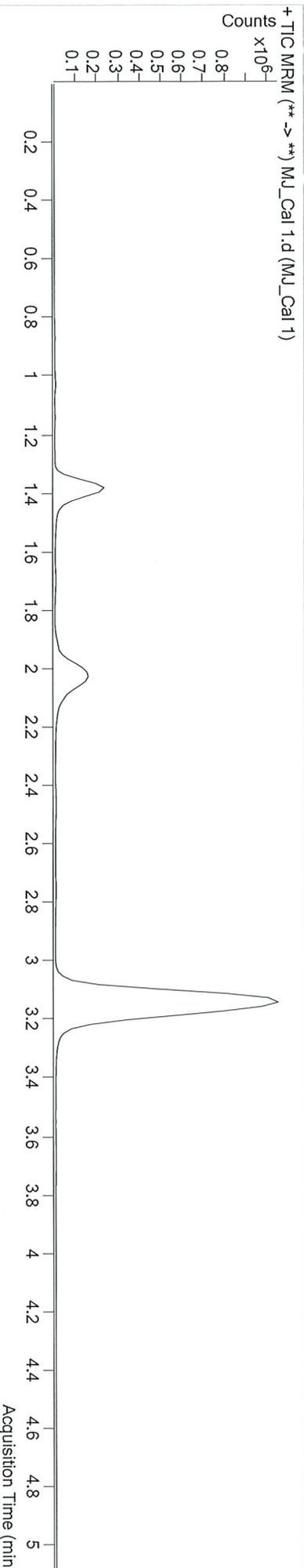


Batch results D:\MassHunter\Data\2019\AM 28\101519 MDQ P1 and P2_THCQ TS\QuantResults\THCQ wk1st 3753 TS.batch.bin
Calibration Last Update 10/16/2019 8:58:41 AM

Instrument Falco
Type Cal
Acq. Method AM 27 THC quant.m
Sample Position P3-A1
Injection Volume 10
Acq. Date-Time 10/16/2019 2:56:09 AM
Sample Info.

Data File MJ_Cal 1.d
Sample MJ_Cal 1
Comment

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.164	48821	238.18	29.7	39.41	5977901	1.0488 ng/ml
THC-COOH	1.414	29887	477.09	50.9	231.19	214220	5.0896 ng/ml
THC-OH	1.453	22819	∞	6.8 Low	11.31	737525	1.0416 ng/ml