

**REVIEWED**

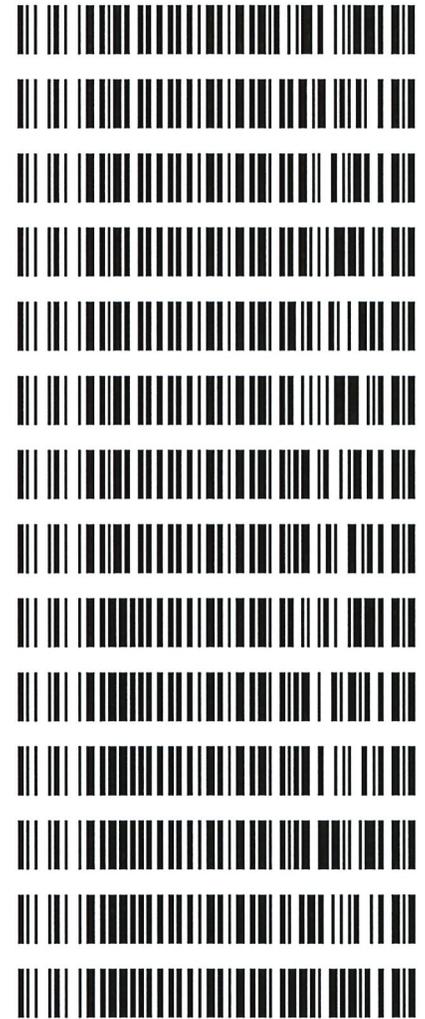
By Anne Nord at 12:43 pm, Jul 16, 2020

AS

7/10/2020

**Worklist: 4353**

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
M2020-2000	2	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-2163	2	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-2166	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-2216	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-2252	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-2272	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-2366	3	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-2374	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1891	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1899	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1900	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1901	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1925	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-1952	1	BCK	AM 27 Blood THC Quant by LC-QQQ



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

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Extraction Date: 07/14/2020  
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  $\geq 0.98$  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 3-100*

TS

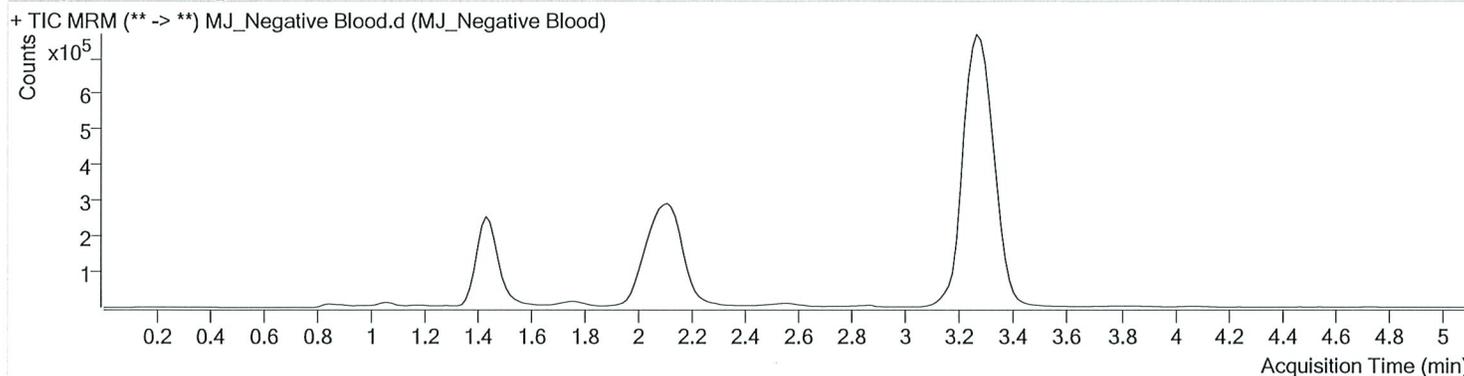


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
Calibration Last Update 7/16/2020 10:03:27 AM

<b>Instrument</b>	Falco	<b>Data File</b>	MJ_Negative Blood.d
<b>Type</b>	Sample	<b>Sample</b>	MJ_Negative Blood
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Tamara Salazar
<b>Sample Position</b>	P3-H5	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/14/2020 1:09:50 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



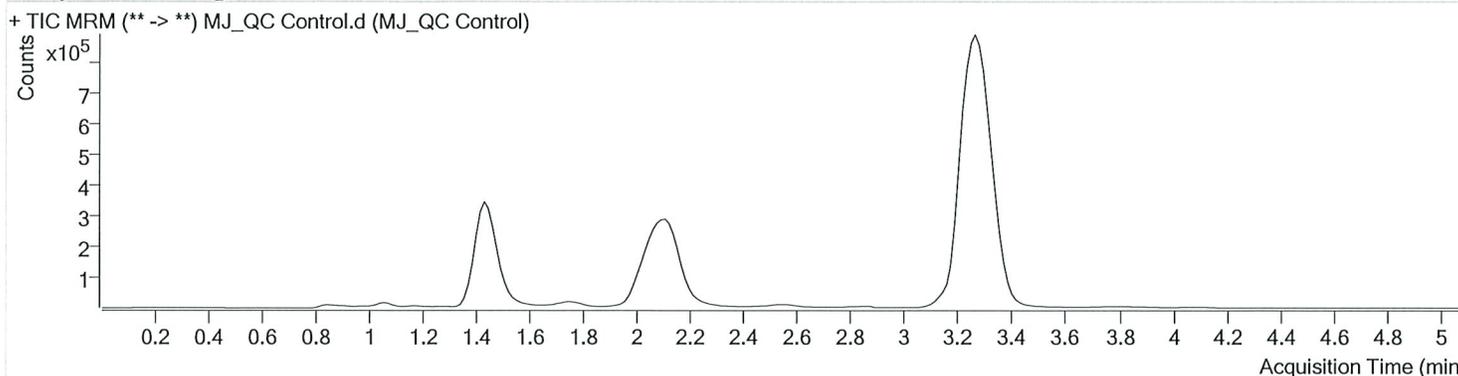


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
 Calibration Last Update 7/16/2020 10:03:27 AM

<b>Instrument</b>	Falco	<b>Data File</b>	MJ_QC Control.d
<b>Type</b>	Sample	<b>Sample</b>	MJ_QC Control
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Tamara Salazar
<b>Sample Position</b>	P3-A6	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/14/2020 12:54:39 PM		

## Sample Chromatogram



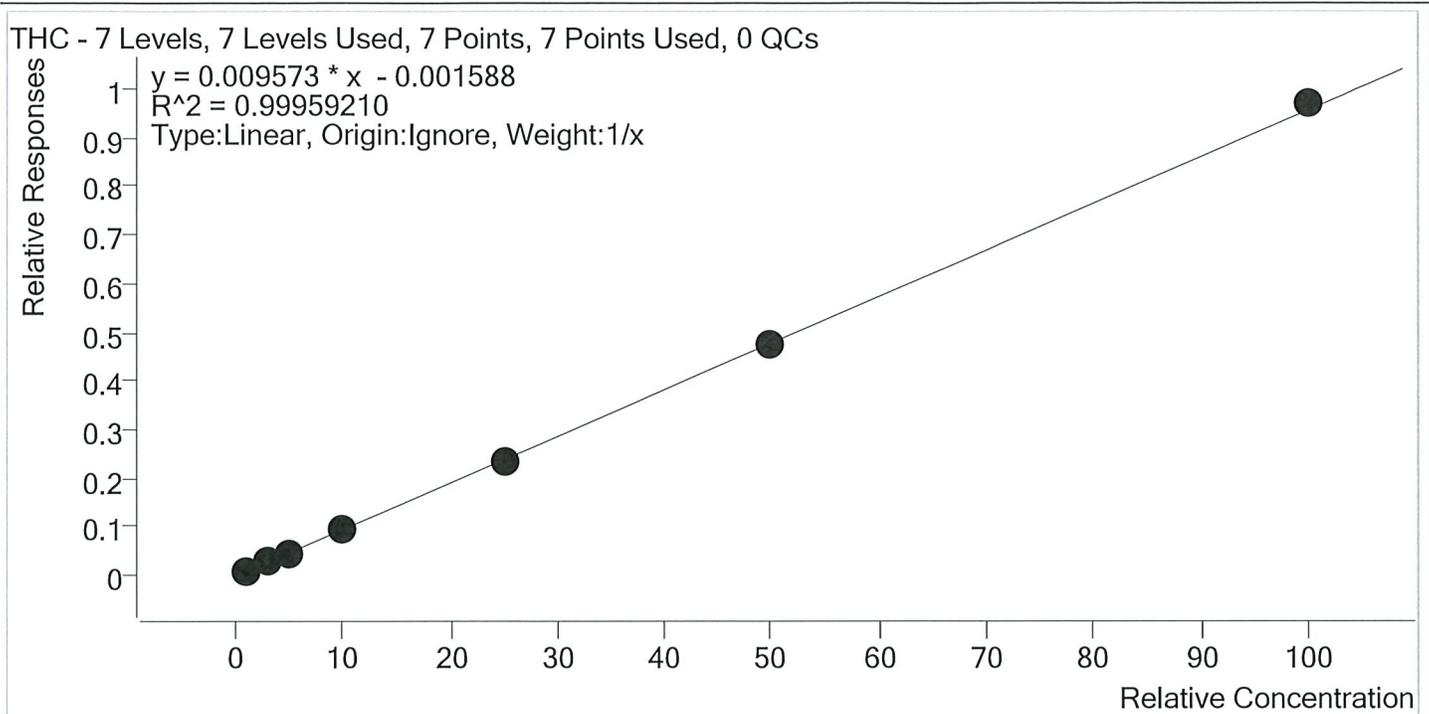
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	171528	∞	9.0	64.75	1162691	4.3019 ng/ml
THC-COOH	1.459	136630	∞	56.4	∞	411156	15.0220 ng/ml
THC	3.285	266619	∞	27.8	224.90	6830524	4.2433 ng/ml

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# AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
 Last Cal. Update 7/16/2020 9:30 AM  
 Analyst Name ISP\datastor  
 Analyte THC Internal Standard THC-D3



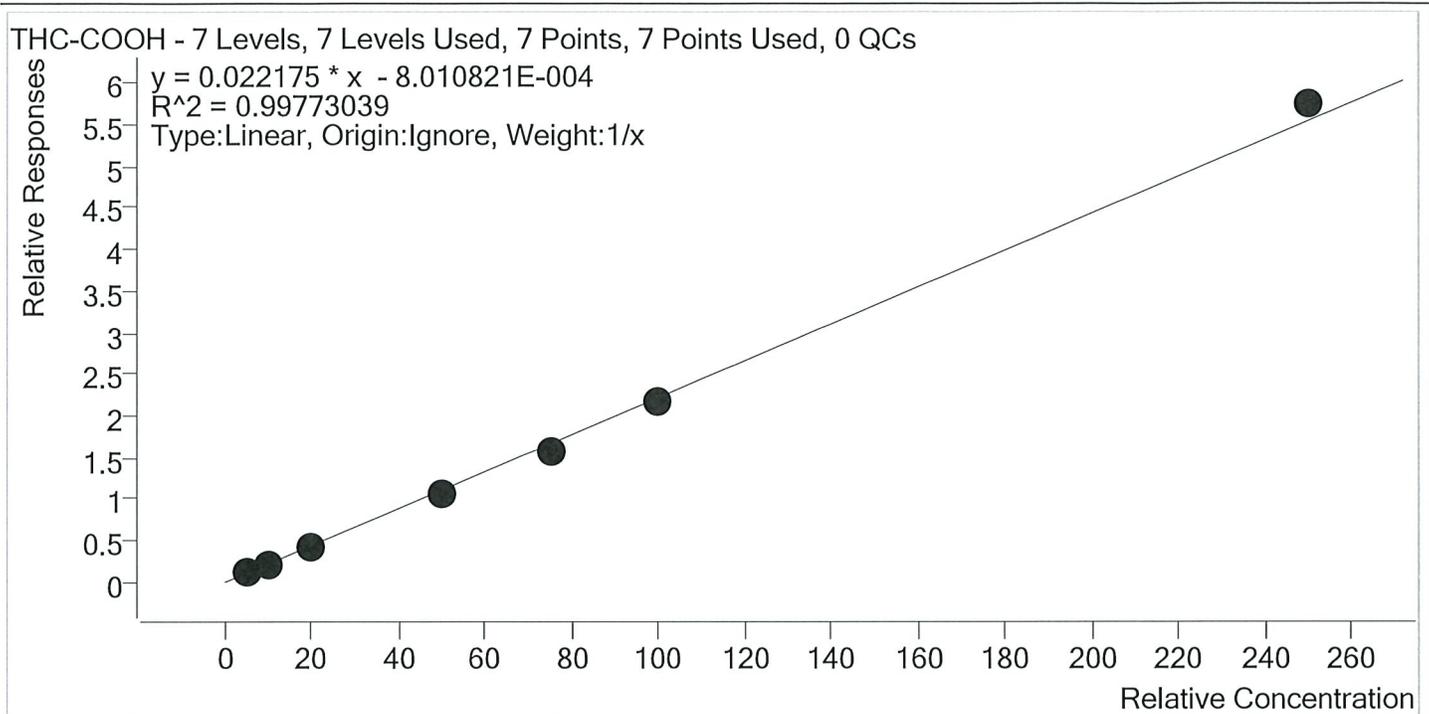
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ Cal 1	1	✓	1.0	1.1	111.5
MJ Cal 2	2	✓	3.0	2.9	96.7
MJ Cal 3	3	✓	5.0	4.8	96.2
MJ Cal 4	4	✓	10.0	9.7	97.5
MJ Cal 5	5	✓	25.0	24.4	97.5
MJ Cal 6	6	✓	50.0	49.6	99.3
MJ Cal 7	7	✓	100.0	101.4	101.4

TS



# AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
 Last Cal. Update 7/16/2020 9:30 AM  
 Analyst Name ISP\datastor  
 Analyte THC-COOH Internal Standard THC-COOH-D9



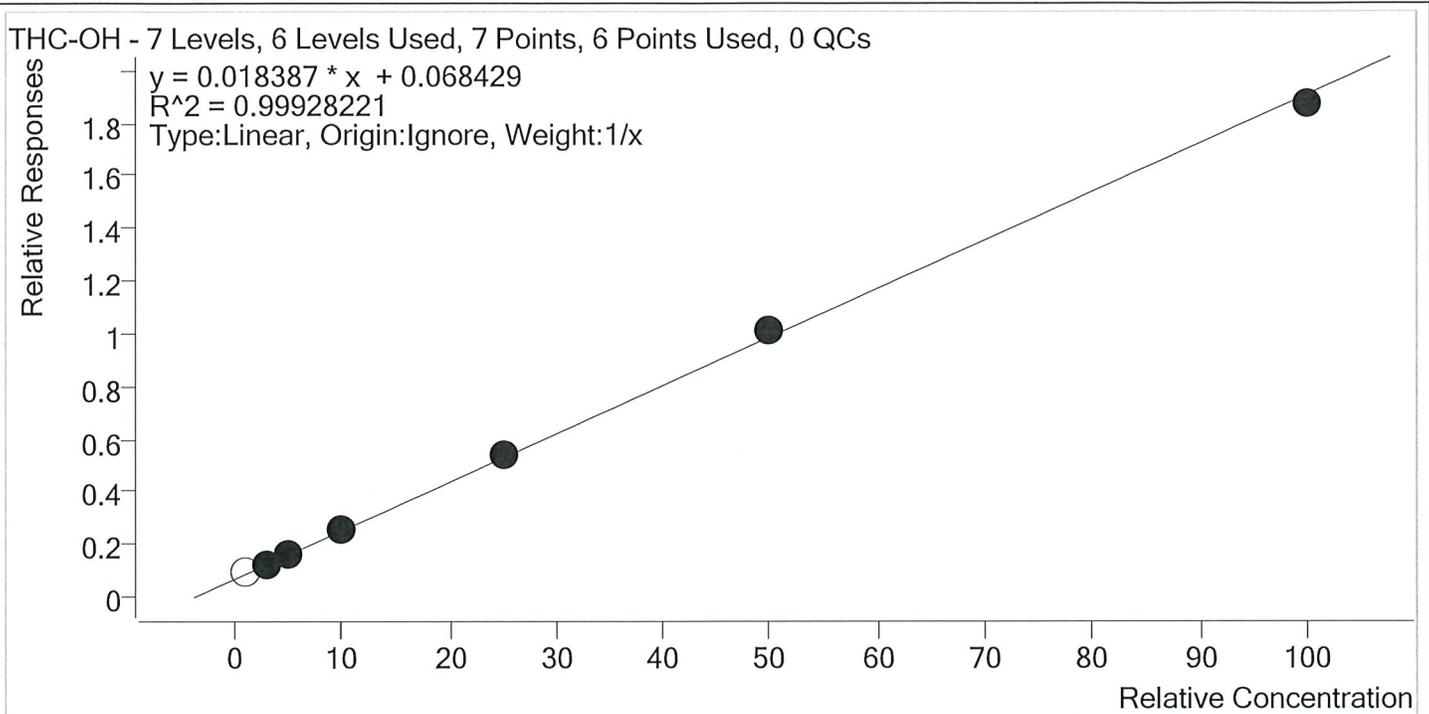
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ Cal 1	1	✓	5.0	5.8	116.7
MJ Cal 2	2	✓	10.0	9.5	95.4
MJ Cal 3	3	✓	20.0	19.2	96.0
MJ Cal 4	4	✓	50.0	48.3	96.6
MJ Cal 5	5	✓	75.0	71.1	94.8
MJ Cal 6	6	✓	100.0	97.0	97.0
MJ Cal 7	7	✓	250.0	259.1	103.6

TS



# AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
 Last Cal. Update 7/16/2020 9:30 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	1.7	167.6
MJ Cal 2	2	✓	3.0	2.8	94.8
MJ Cal 3	3	✓	5.0	5.1	102.6
MJ Cal 4	4	✓	10.0	10.0	99.9
MJ Cal 5	5	✓	25.0	25.4	101.4
MJ Cal 6	6	✓	50.0	51.6	103.1
MJ Cal 7	7	✓	100.0	98.1	98.1

TS

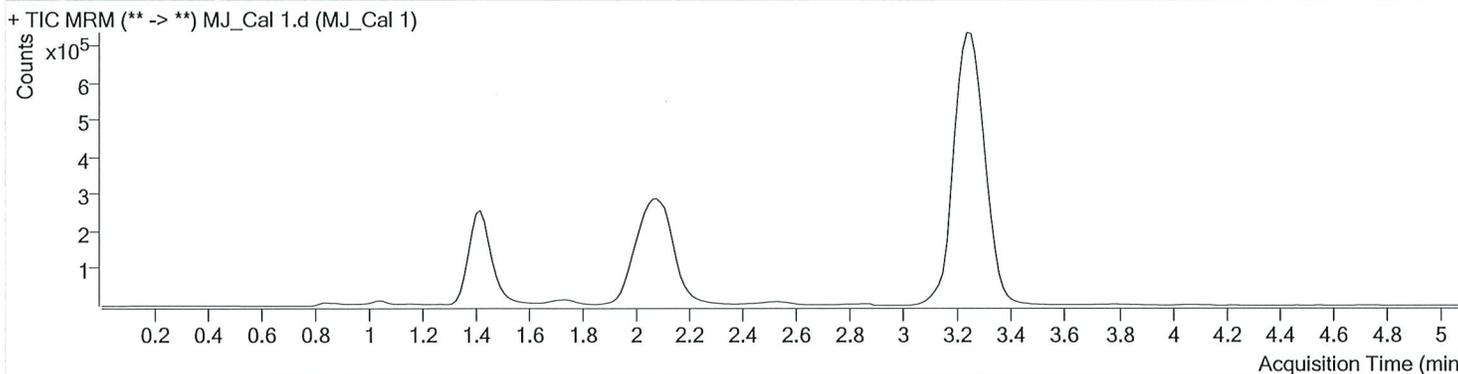


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
Calibration Last Update 7/16/2020 10:03:27 AM

<b>Instrument</b>	Falco	<b>Data File</b>	MJ_Cal 1.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 1
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Tamara Salazar
<b>Sample Position</b>	P3-H6	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/14/2020 11:53:46 AM		

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.468	98559	∞	3.9 <b>Low</b>	17.38	993100	1.6760 ng/ml <b>Low</b>
THC-COOH	1.444	45601	∞	48.0	∞	354735	5.8333 ng/ml
THC	3.254	53653	393.77	28.6	∞	5907189	1.1146 ng/ml <b>Low</b>

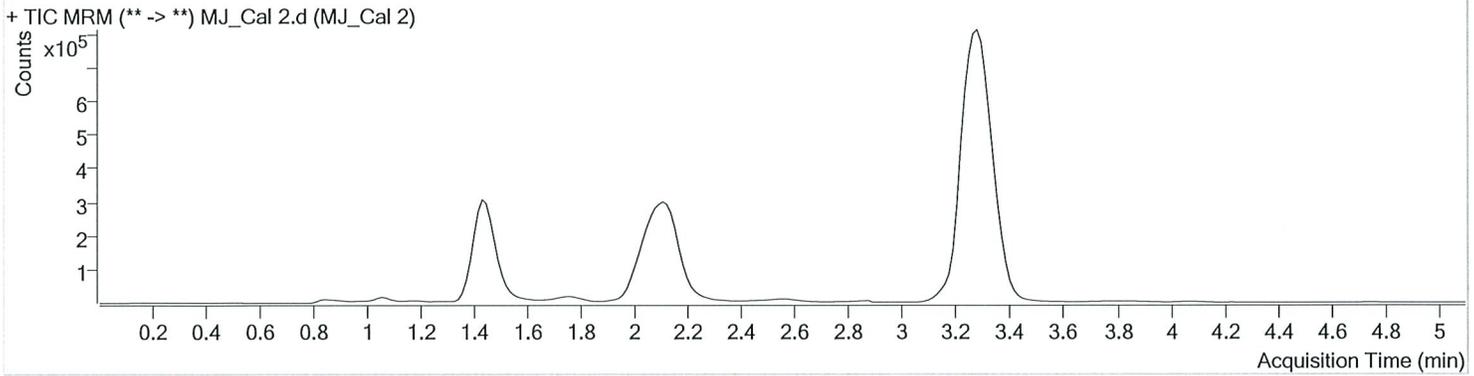


# AM #27 Cannabinoid Quant. Results

**Batch results** D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
**Calibration Last Update** 7/16/2020 10:03:27 AM

<b>Instrument</b>	Falco	<b>Data File</b>	MJ_Cal 2.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 2
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Tamara Salazar
<b>Sample Position</b>	P3-G6	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/14/2020 12:01:30 PM		
<b>Sample Info.</b>			

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.498	136486	∞	8.7	∞	1130666	2.8436 ng/ml <b>Low</b>
THC-COOH	1.474	82601	∞	55.8	647.95	392090	9.5365 ng/ml
THC	3.300	167915	584.94	28.5	160.07	6414070	2.9005 ng/ml <b>Low</b>

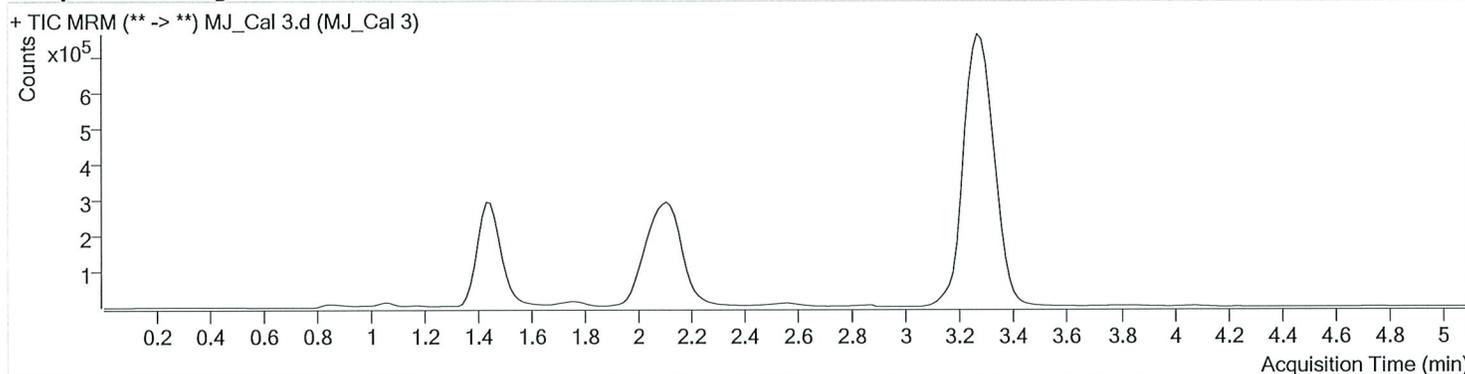


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
 Calibration Last Update 7/16/2020 10:03:27 AM

<b>Instrument</b>	Falco	<b>Data File</b>	MJ_Cal 3.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 3
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Tamara Salazar
<b>Sample Position</b>	P3-F6	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/14/2020 12:09:05 PM		

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	163310	∞	9.2	∞	1003146	5.1325 ng/ml
THC-COOH	1.474	151890	∞	56.9	∞	357286	19.2077 ng/ml
THC	3.285	256838	782.38	25.5	385.31	5779938	4.8076 ng/ml

TS

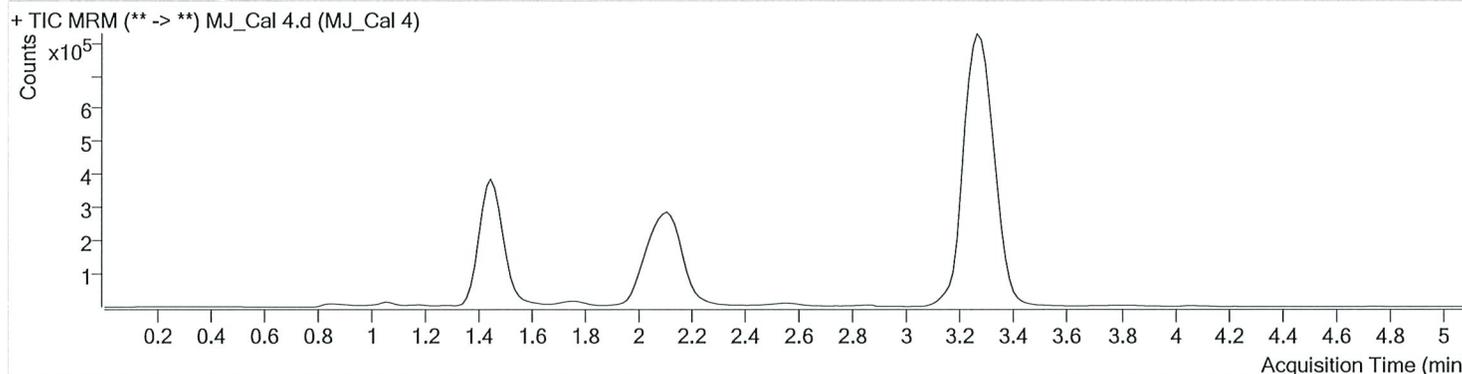


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
Calibration Last Update 7/16/2020 10:03:27 AM

<b>Instrument</b>	Falco	<b>Data File</b>	MJ_Cal 4.d
<b>Type</b>	Cal	<b>Sample</b>	MJ_Cal 4
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Operator</b>	Tamara Salazar
<b>Sample Position</b>	P3-E6	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/14/2020 12:16:40 PM		

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	254987	∞	11.1	128.71	1011583	9.9876 ng/ml
THC-COOH	1.474	388553	∞	59.7	∞	363233	48.2762 ng/ml
THC	3.285	544245	2965.69	25.9	∞	5933834	9.7468 ng/ml

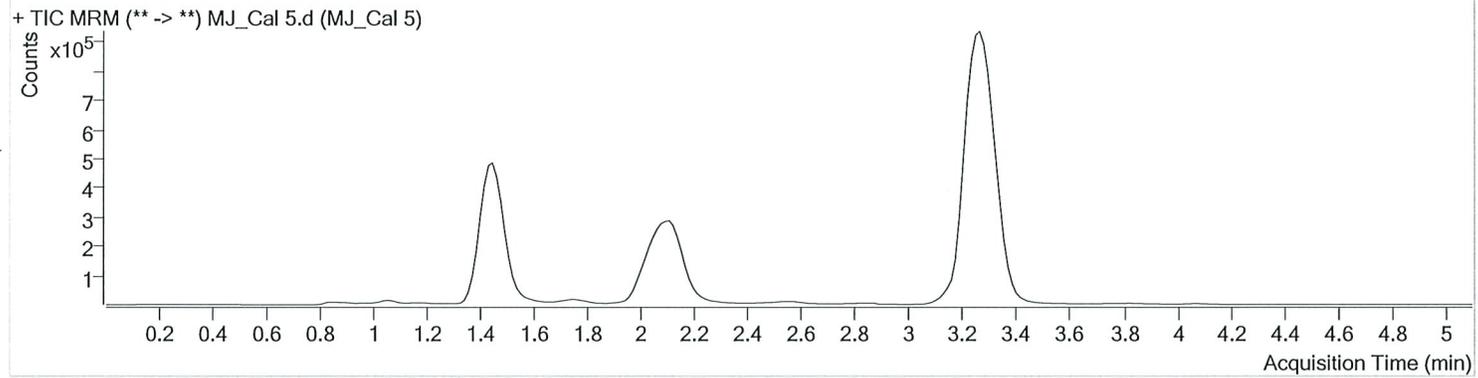


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
Calibration Last Update 7/16/2020 10:03:27 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 P3-D6 Comment  
Injection Volume 10  
Acq. Date-Time 7/14/2020 12:24:17 PM  
Sample Info.

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	544773	∞	11.1	∞	1018774	25.3612 ng/ml
THC-COOH	1.459	565101	∞	63.3	∞	358684	71.0849 ng/ml
THC	3.270	1348697	13393.22	25.6	1701.38	5817083	24.3850 ng/ml

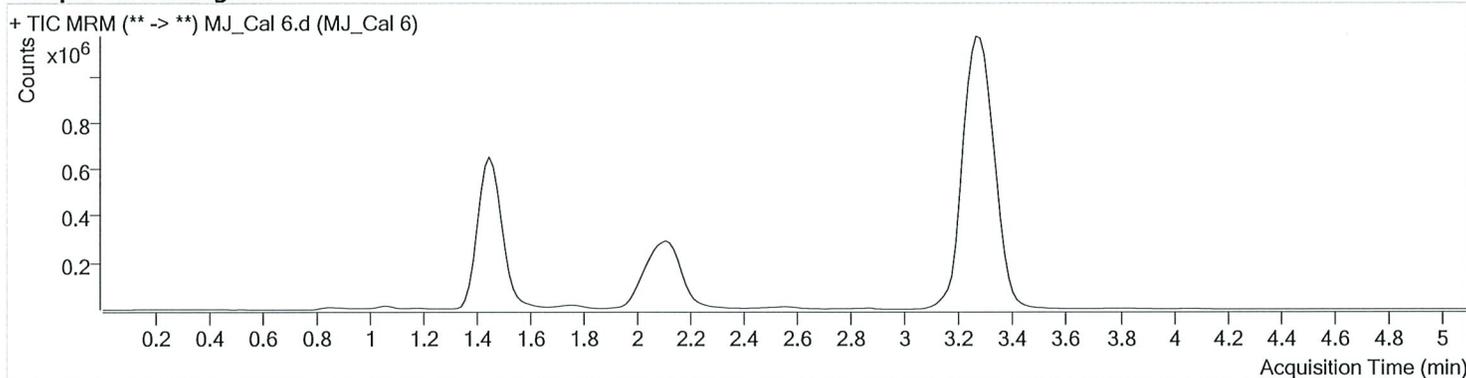


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
Calibration Last Update 7/16/2020 10:03:27 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 P3-C6 Comment  
Injection Volume 10  
Acq. Date-Time 7/14/2020 12:31:52 PM  
Sample Info.

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	1064375	∞	12.3	∞	1047016	51.5677 ng/ml
THC-COOH	1.474	782293	∞	58.9	∞	363992	96.9578 ng/ml
THC	3.285	2844505	3553.33	25.9	1018.24	6005047	49.6470 ng/ml

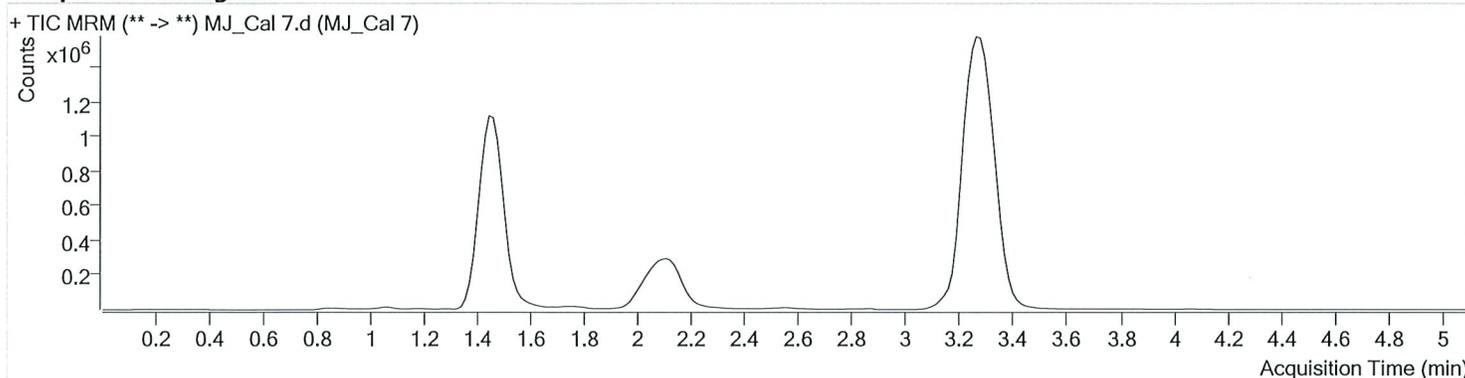


# AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\071420 AM 27 28 TS\QuantResults\AM 27.batch.bin  
Calibration Last Update 7/16/2020 10:03:27 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	P3-B6	Comment	
Injection Volume	10		
Acq. Date-Time	7/14/2020 12:39:27 PM		

## Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.438	1919878	∞	12.8	2066.60	1025420	98.1074 ng/ml
THC-COOH	1.474	1969237	∞	56.7	∞	342790	259.1037 ng/ml
THC	3.285	5744816	∞	25.9	∞	5927964	101.3984 ng/ml