



REVIEWED

By Tamara Salazar at 2:45 pm, Oct 06, 2023

Worklist: 6521

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>	
C2023-2144	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2145	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2023-2148	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2163	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2172	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2023-2175	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2179	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2195	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2211	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2218	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2242	1	BLOOD	AM 27 Blood THC Quant by LC-QQQ	
C2023-2246	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2023-2261	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2273	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2023-2274	2	BCK	AM 27 Blood THC Quant by LC-QQQ	



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

Extraction Date 10/05/23
Plate lot#: 230627

Analyst: Anne Nord
Plate re-test: 12/27/2023

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: 23J52629 **Urine Blank:** 8423 **Column:** UCT Selectra DA 100 x 2.1mm 3um
LCMS-QQQ ID: 69679

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. Urine hydrolysis: add 1.5 ml urine to blank plate, add 250 ul 1N KOH mix and incubate at 40 degrees for 15 minutes.
Pipette **1000µL (calibrated pipette) blood or 1000µL hydrolyzed urine** Pipette ID: I41142J 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** **blood sample, 500 ul saturated phosphate buffer in urine** in wells of analytical plate.
- 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 6. Transfer **800µL of blood+acid or urine 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: 66792
- 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: 66819
- 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. SN > 10
- 4. Case sample response for THC 1ng/ml LOD 3ng/ml LOQ, OH-THC 3ng/mL LOD and LOQ, Carboxy-THC: 5 ng/mL (qualitative only). 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 QC's pass for each analyte? (if not is it describe in comments section)
- 6. Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS:

Curve range THC-OH 3-100 dropped cal 1 due to qualifier ion having poor response in relation to baseline.



	1	2	3	4	5	6
a	cal 1	Internal control urine	2211-1 mixing	2246-1		
b	cal 2	negative blood	2218-1	2273-1		
c	cal 3	2144-1	2242-1	2211-1 SLE and injection		
d	cal 4	2148-1	2261-1			
e	cal 5	2163-1	2274-2			
f	cal 6	2175-1	negative urine			
g	cal 7	2179-1	2145-1			
h	Internal control (blood)	2195-1	2172-1			

Plate position 3

c2023-____-_-

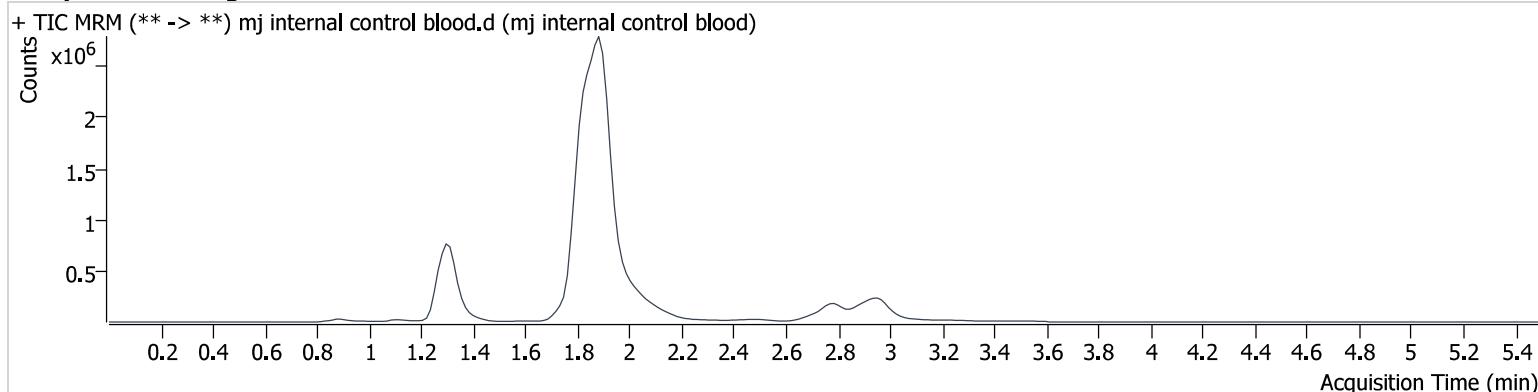


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj internal control blood.d
Type	QC	Sample	mj internal control blood
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-H1	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/5/2023 9:09:02 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.304	29838	∞	861.60	∞	2419804	4.748 ng/ml
THC-COOH	1.327	49055	312.0	294.42	∞	719757	14.033 ng/ml
THC	2.806	61175	3081.8	30.38	∞	560625	4.066 ng/ml

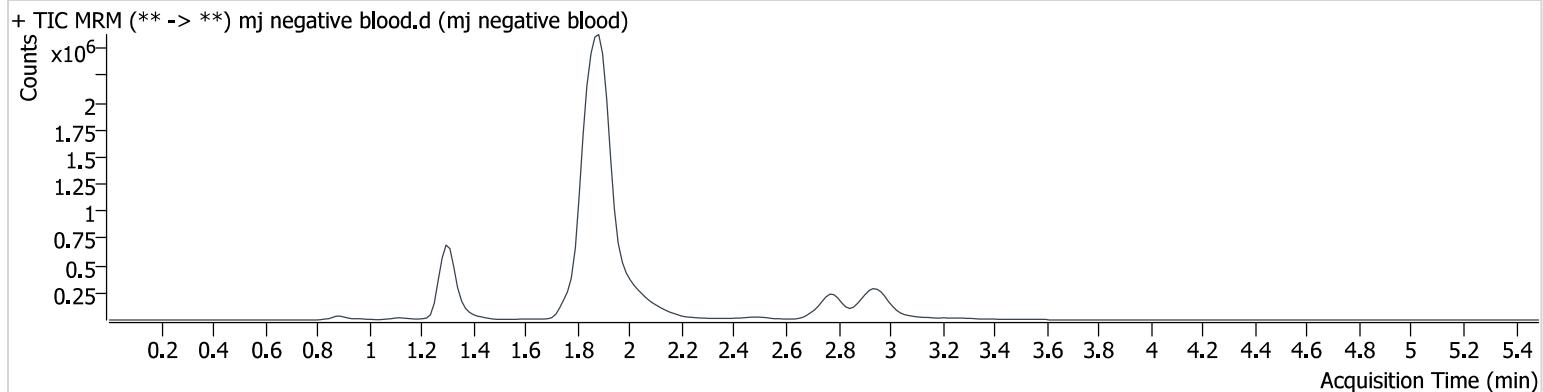


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj negative blood.d
Type	Sample	Sample	mj negative blood
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-B2	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/5/2023 9:15:38 PM		
Sample Info.			

Sample Chromatogram



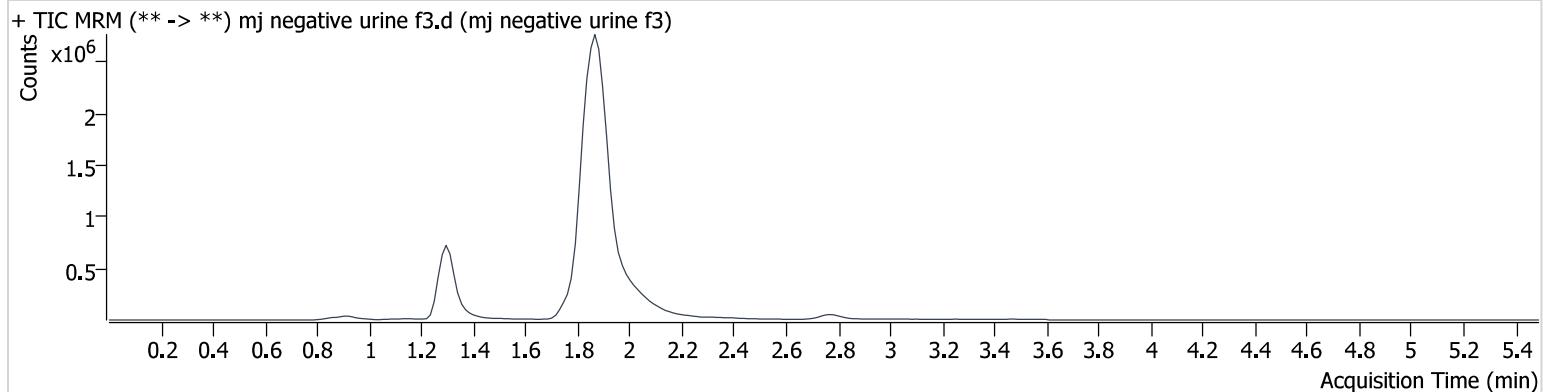


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj negative urine f3.d
Type	Sample	Sample	mj negative urine f3
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-F3	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/5/2023 11:47:28 PM		
Sample Info.			

Sample Chromatogram



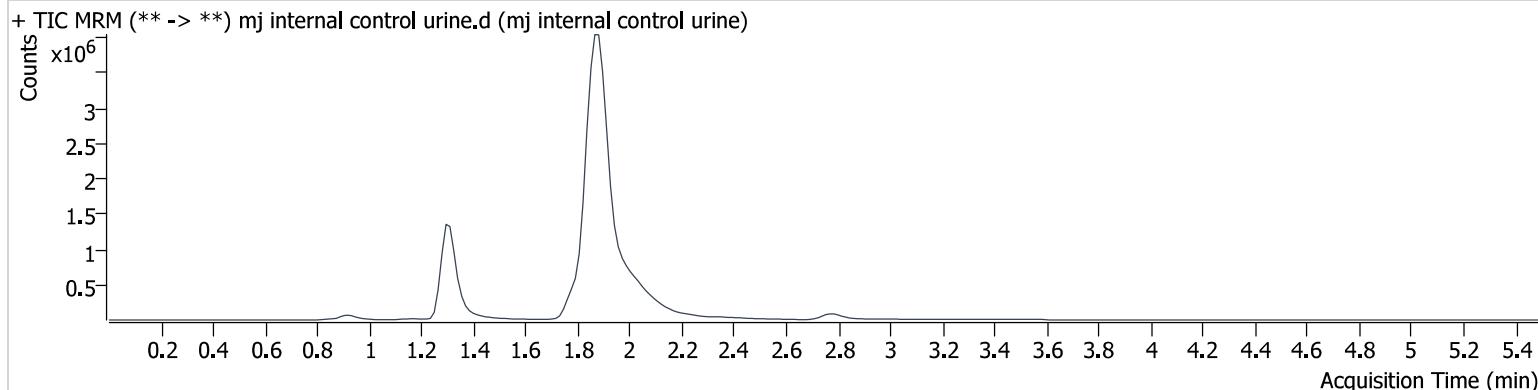


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj internal control urine.d
Type	Sample	Sample	mj internal control urine
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-A2	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/6/2023 12:53:29 AM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.304	49356	∞	887.47	∞	3880420	4.873 ng/ml
THC-COOH	1.327	63241	∞	277.60	781.1	813742	15.856 ng/ml
THC	2.791	47732	∞	29.20	∞	401619	4.387 ng/ml

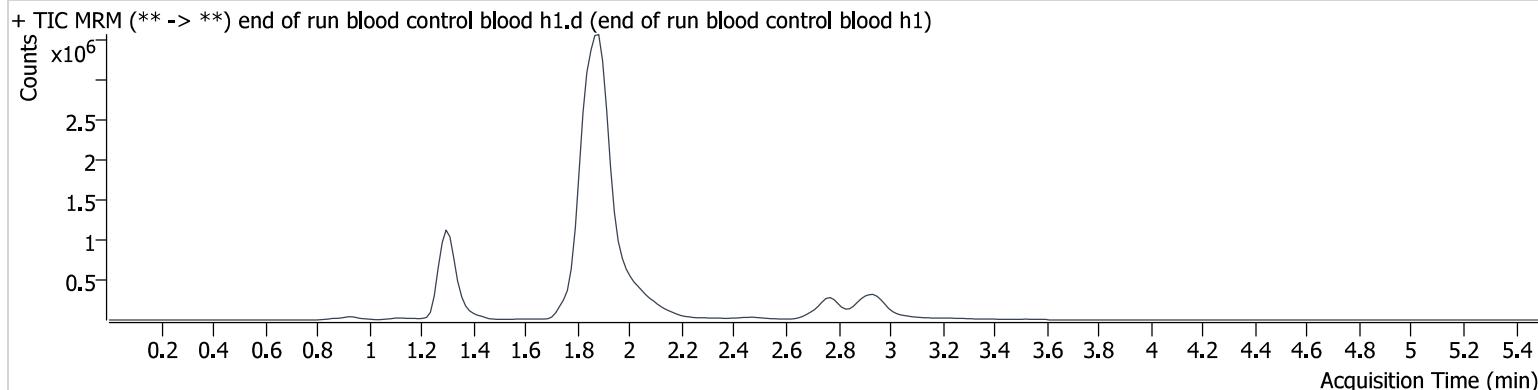


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	end of run blood control blood h1.d
Type	Sample	Sample	end of run blood control blood h1
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-H1	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/6/2023 1:00:05 AM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.304	41242	∞	856.56	∞	3286945	4.817 ng/ml
THC-COOH	1.312	68233	∞	283.94	1573.8	935670	14.942 ng/ml
THC	2.776	103623	1071.2	26.28	∞	929713	4.143 ng/ml

Compound Calibration Report

Batch results

D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin

Last Cal. Update

10/6/2023 8:53 AM

Analyst Name

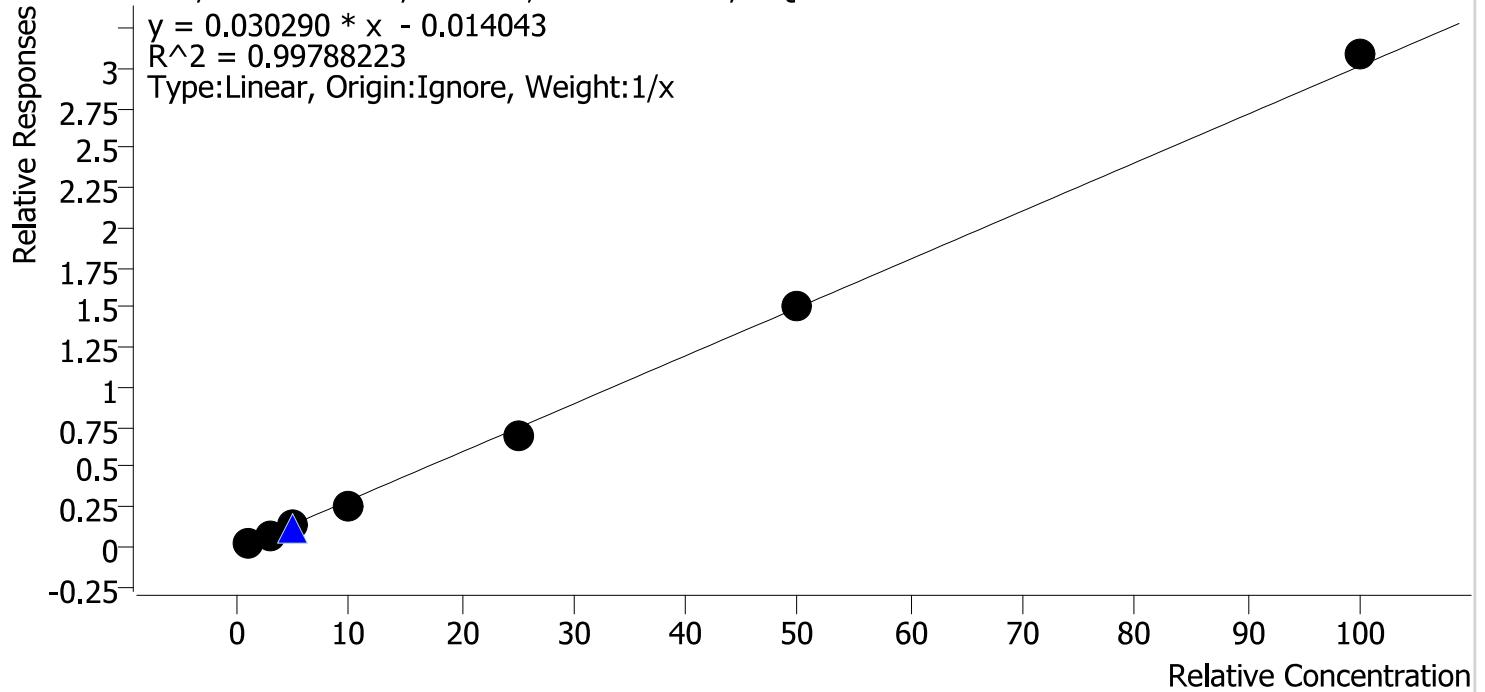
ISP\datastor

Analyte

THC

Internal Standard

THC-d3

THC - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 1 QC


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✓	1.0	1.2	122.5
mj cal 2	2	✓	3.0	2.9	95.7
mj cal 3	3	✓	5.0	4.8	96.2
mj cal 4	4	✓	10.0	8.9	88.8
mj cal 5	5	✓	25.0	23.4	93.6
mj cal 6	6	✓	50.0	50.4	100.9
mj cal 7	7	✓	100.0	102.4	102.4

Compound Calibration Report

Batch results

D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin

Last Cal. Update

10/6/2023 8:53 AM

Analyst Name

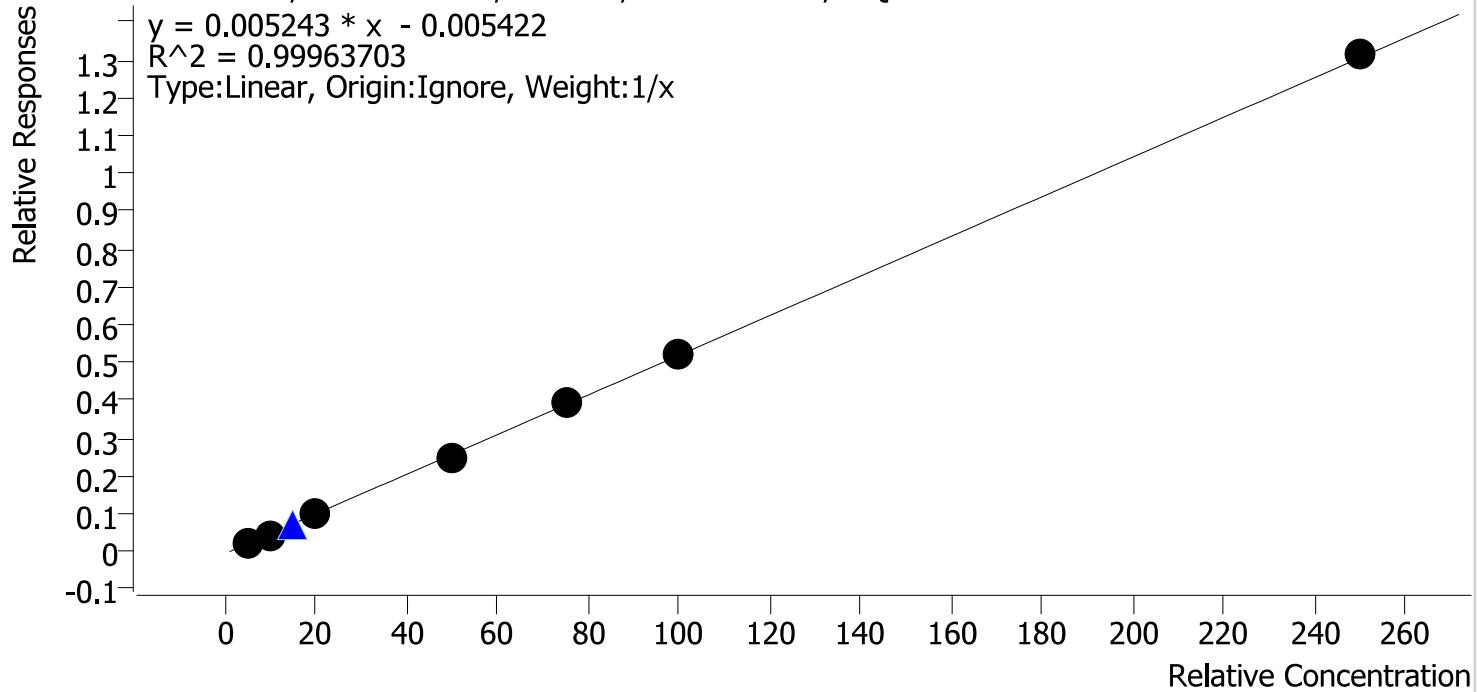
ISP\datastor

Analyte

THC-COOH

Internal Standard

THC-COOH-d9

THC-COOH - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 1 QC


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✓	5.0	5.4	108.6
mj cal 2	2	✓	10.0	9.7	97.3
mj cal 3	3	✓	20.0	19.2	95.9
mj cal 4	4	✓	50.0	48.4	96.8
mj cal 5	5	✓	75.0	75.2	100.3
mj cal 6	6	✓	100.0	100.4	100.4
mj cal 7	7	✓	250.0	251.6	100.6

Compound Calibration Report

Batch results

D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin

Last Cal. Update

10/6/2023 8:53 AM

Analyst Name

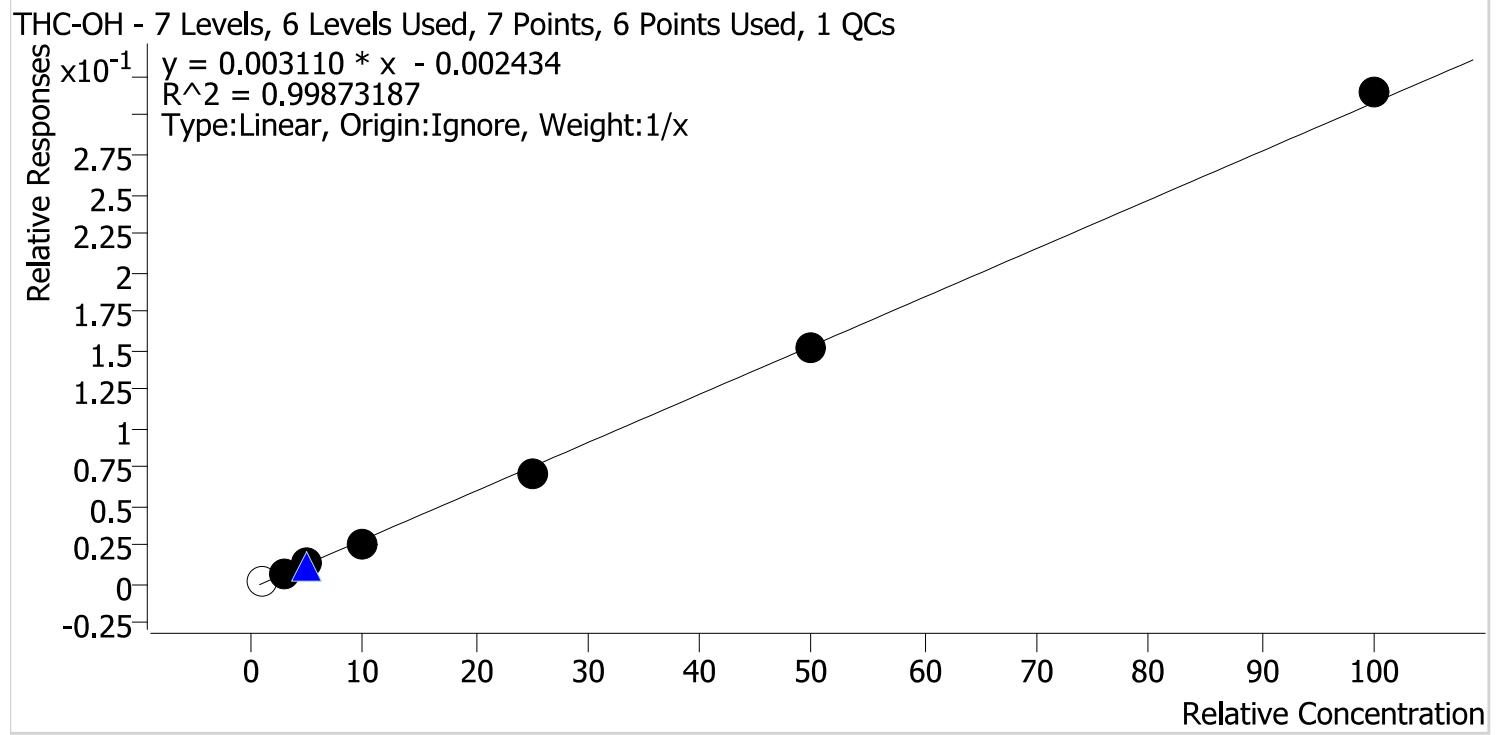
ISP\datastor

Analyte

THC-OH

Internal Standard

THC-OH-d3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	x	1.0	1.6	158.1
mj cal 2	2	✓	3.0	3.3	110.3
mj cal 3	3	✓	5.0	5.0	99.9
mj cal 4	4	✓	10.0	9.4	93.7
mj cal 5	5	✓	25.0	23.8	95.1
mj cal 6	6	✓	50.0	49.5	99.0
mj cal 7	7	✓	100.0	102.1	102.1

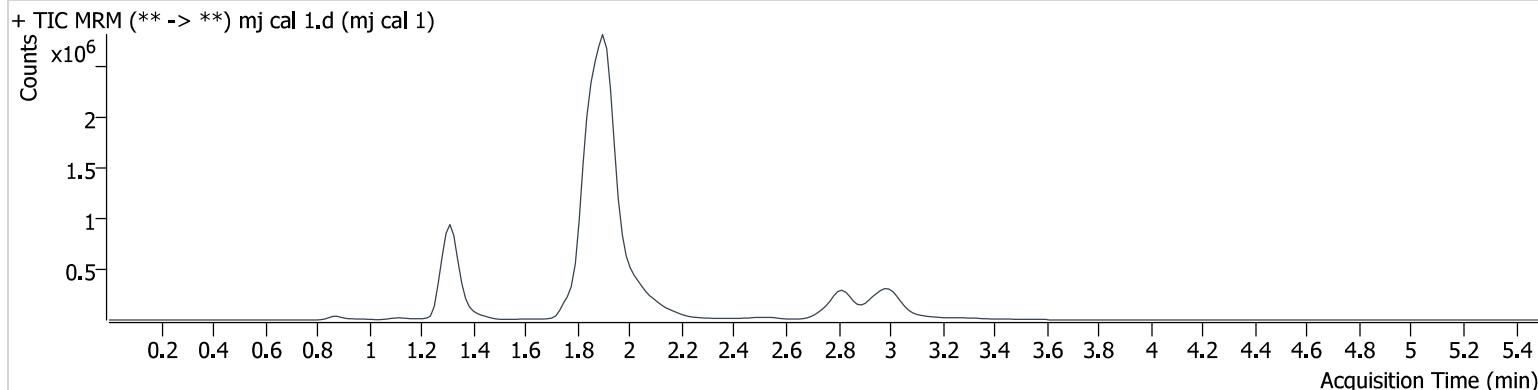


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj cal 1.d
Type	Cal	Sample	mj cal 1
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-A1	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/5/2023 8:22:40 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.319	8161	∞	717.12	∞	3287400	1.581 ng/ml	Low
THC-COOH	1.342	20058	∞	289.89	∞	870359	5.429 ng/ml	
THC	2.836	23721	∞	30.29	58.9	1029265	1.225 ng/ml	

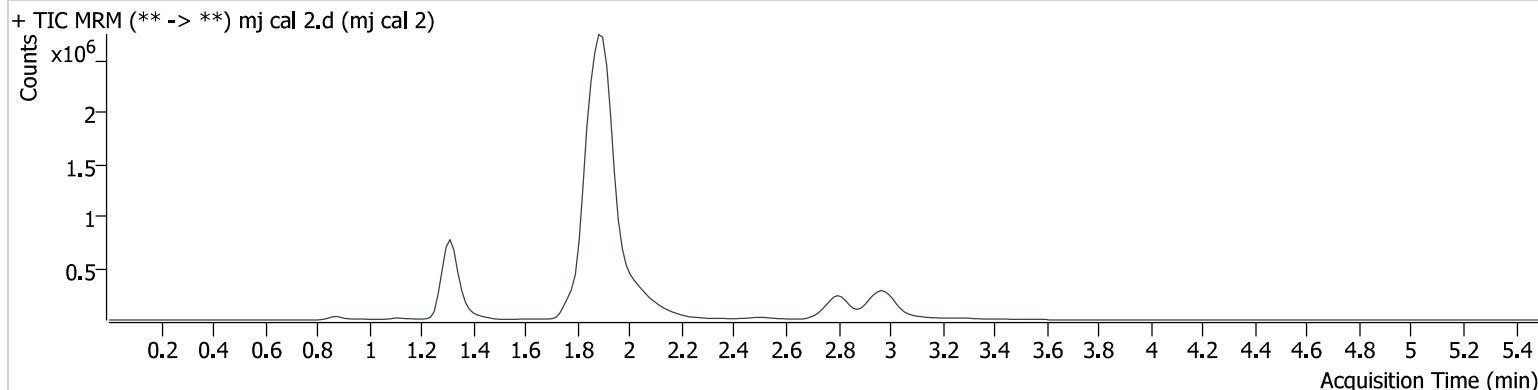


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj cal 2.d
Type	Cal	Sample	mj cal 2
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-B1	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/5/2023 8:29:26 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.319	18489	∞	838.48	∞	2354543	3.308 ng/ml
THC-COOH	1.327	31176	∞	289.24	442.4	683884	9.729 ng/ml
THC	2.821	55670	∞	26.90	∞	763404	2.871 ng/ml

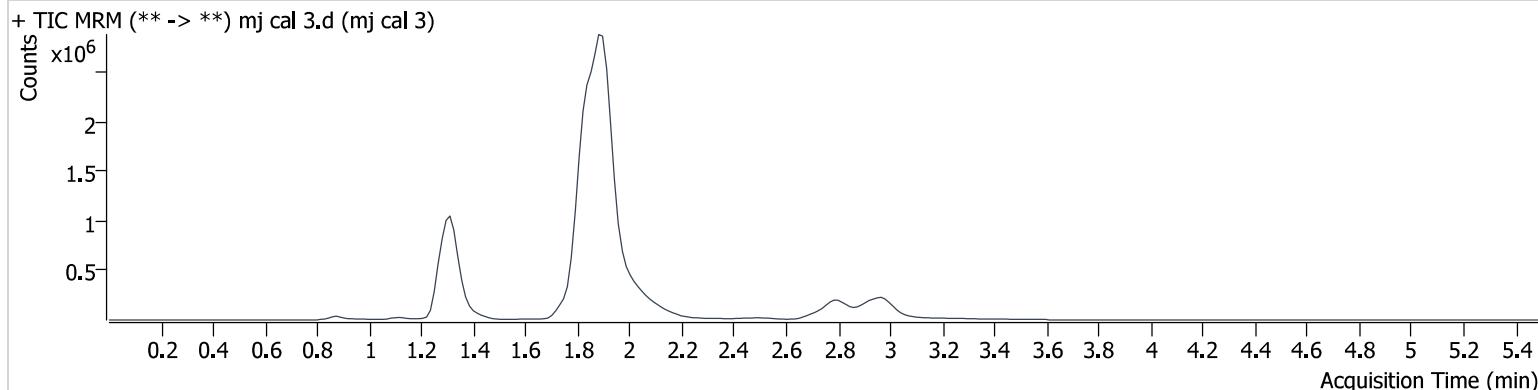


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj cal 3.d
Type	Cal	Sample	mj cal 3
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-C1	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/5/2023 8:36:02 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.304	43061	∞	855.14	∞	3285552	4.997 ng/ml
THC-COOH	1.327	89884	∞	277.58	493.2	944372	19.187 ng/ml
THC	2.821	89236	∞	27.84	∞	677846	4.810 ng/ml

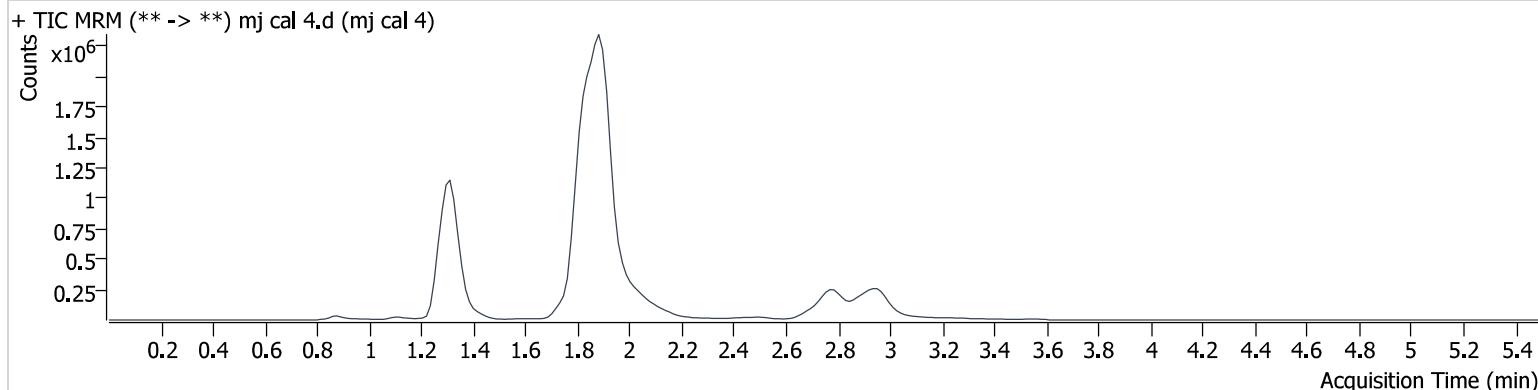


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj cal 4.d
Type	Cal	Sample	mj cal 4
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-D1	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/5/2023 8:42:38 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.304	76276	∞	888.90	∞	2856766	9.368 ng/ml
THC-COOH	1.327	194858	1119.3	276.23	∞	784427	48.411 ng/ml
THC	2.806	197244	∞	25.10	∞	774106	8.876 ng/ml

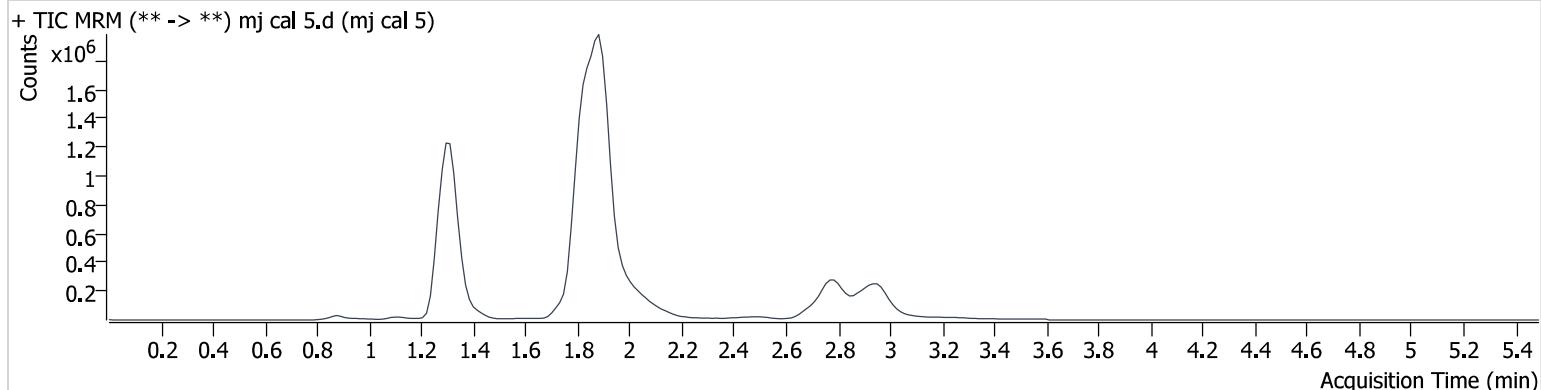


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj cal 5.d
Type	Cal	Sample	mj cal 5
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-E1	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/5/2023 8:49:14 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.304	171175	∞	859.93	∞	2395251	23.763 ng/ml
THC-COOH	1.327	240408	1914.5	272.81	1325.1	617814	75.249 ng/ml
THC	2.806	471973	4749.9	25.25	∞	679074	23.410 ng/ml

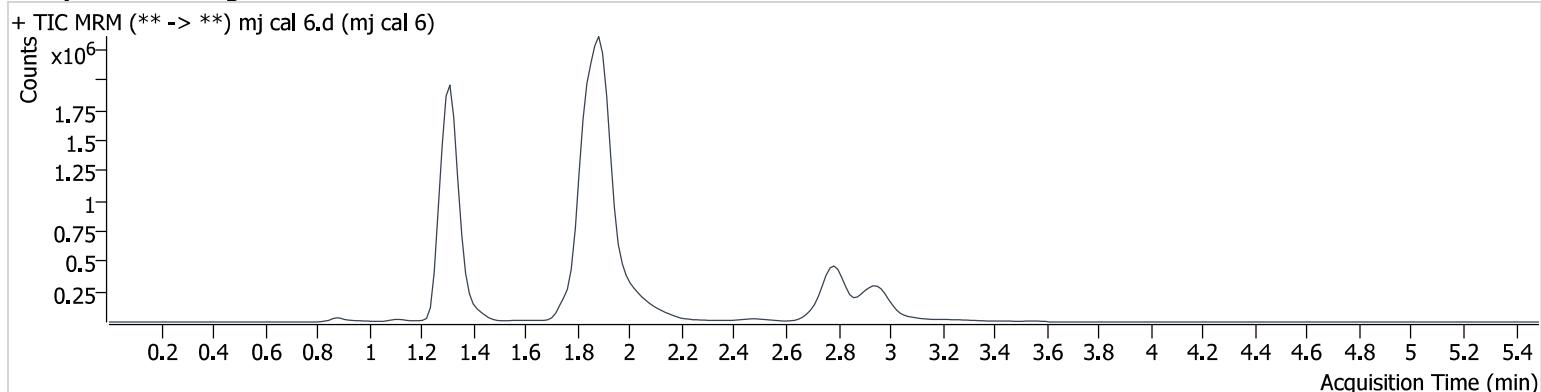


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj cal 6.d
Type	Cal	Sample	mj cal 6
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-F1	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/5/2023 8:55:50 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.304	384092	∞	843.86	∞	2535115	49.503 ng/ml
THC-COOH	1.327	371462	3478.1	273.58	∞	712983	100.400 ng/ml
THC	2.806	1255158	∞	24.15	∞	829156	50.441 ng/ml

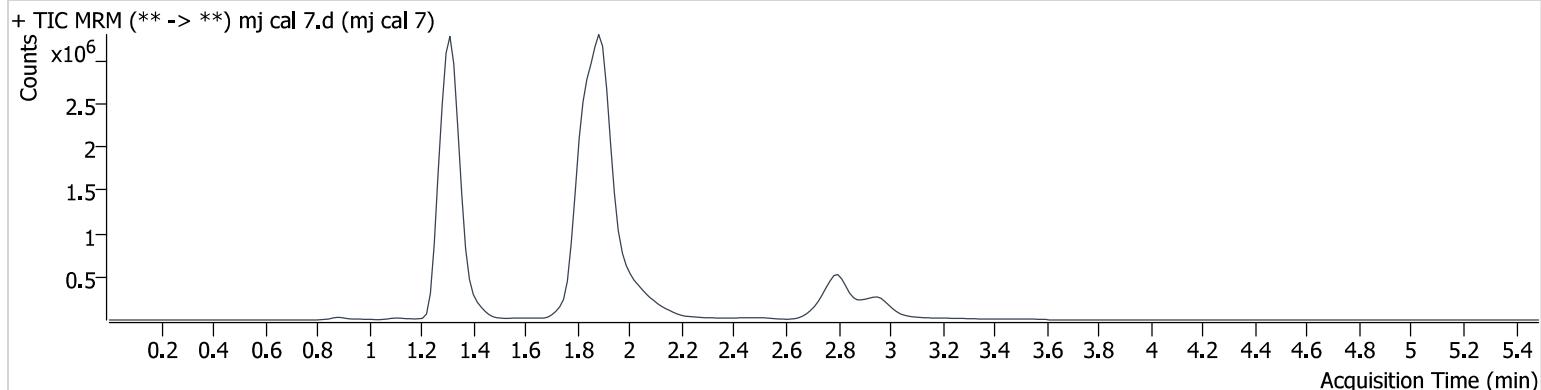


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\100523\QuantResults\cann.batch.bin
Calibration Last Update 10/6/2023 8:53:31 AM

Instrument	69679	Data File	mj cal 7.d
Type	Cal	Sample	mj cal 7
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-G1	Comment	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods
Injection Volume	10		
Acq. Date-Time	10/5/2023 9:02:26 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.304	813749	∞	826.39	∞	2583702	102.061 ng/ml
THC-COOH	1.327	887579	3863.6	272.18	∞	675607	251.595 ng/ml
THC	2.806	1931626	∞	25.19	7244.9	625801	102.368 ng/ml