1/10/2024

REVIEWED
By Britany Wylie at 4:31 pm, Jan 15, 2024

Worklist: 6649

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION
C2023-2796	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2023-2804	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2023-2815	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2023-2841	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2023-2856	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2024-0030	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ

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

Extraction Date 1/9/24 Analyst: Anne Nord
Plate lot#: 231212 Plate re-test: 06/12/24

Mobile phase A: 0.1% Formic Acid in LCMS Water **Mobile phase B:** 0.1% Formic acid in Acetonitrile
MTBE LCMS Methanol Hexane
Blank Blood Lot: 23J52629 **Urine Blank:** 1324 **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 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. 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 QCs 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:

	1	2	3	4	5	6
a	cal 1	Internal control urine	m2024-0030-1			
b	cal 2	negative blood				
c	cal 3	2796-1				
d	cal 4	2804-1				
e	cal 5	2815-1				
f	cal 6	2841-1				
g	cal 7	2856-1				
h	Internal control (blood)	negative urine				

Plate position 3

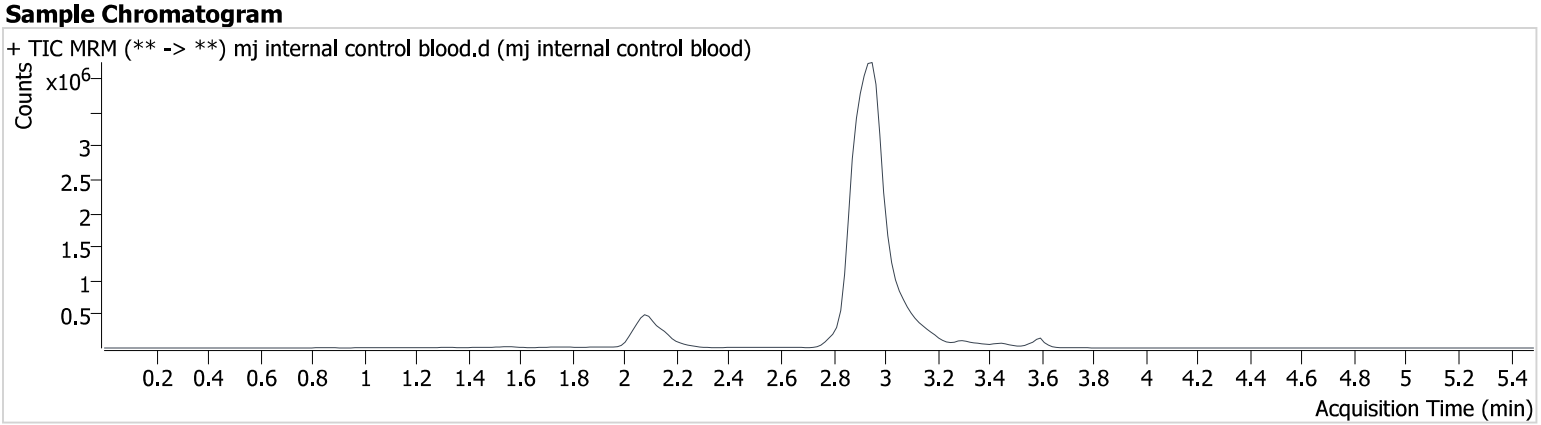
c2023-____-_

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj internal control blood.d
Type	QC	**Sample**	mj internal control blood
Acq. Method	thc quant 50 50.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	1/9/2024 7:14:39 PM		
Sample Info.			

Sample Chromatogram



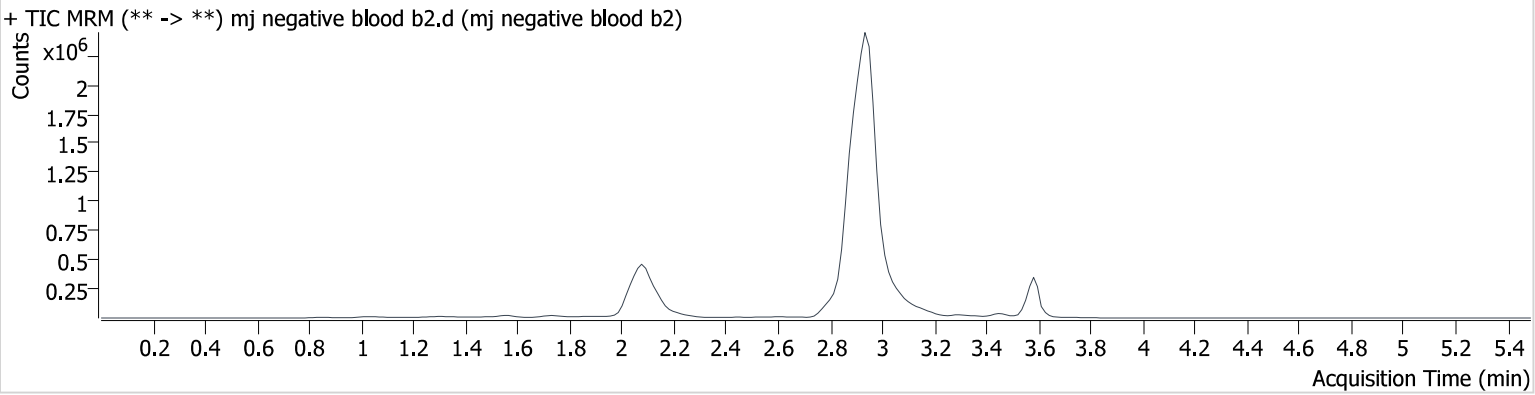
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.081	31109	∞	938.66	∞	2022300	4.674 ng/ml
THC-COOH	2.152	50430	1673.3	265.96	36.4	730682	14.497 ng/ml
THC	3.603	47181	∞	25.55	∞	338361	4.703 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj negative blood b2.d
Type	Sample	**Sample**	mj negative blood b2
Acq. Method	thc quant 50 50.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	1/9/2024 7:21:13 PM		
Sample Info.			

Sample Chromatogram

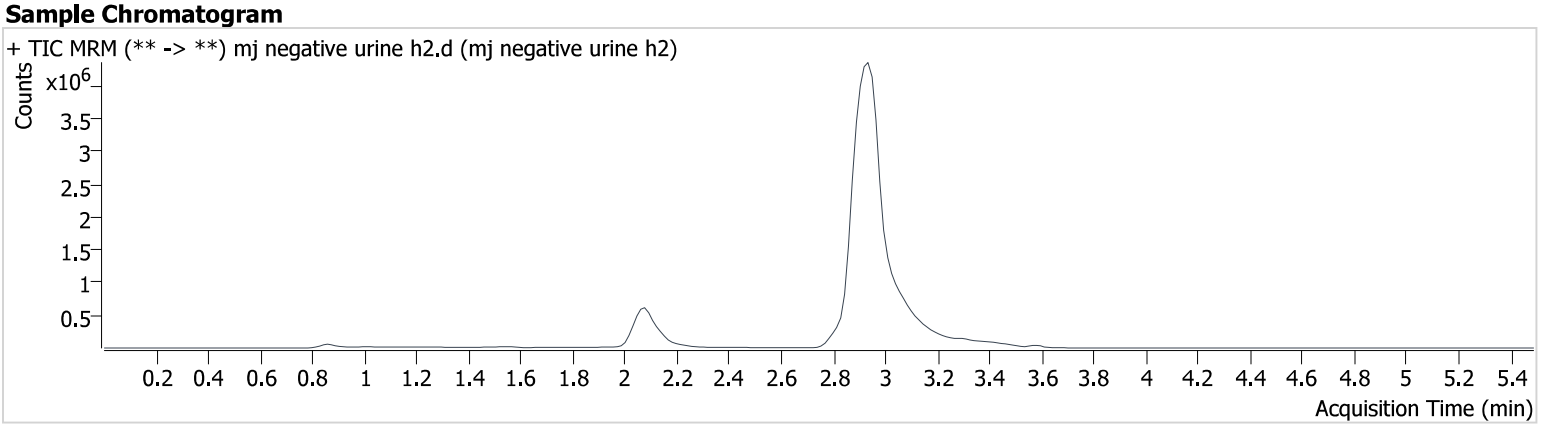


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj negative urine h2.d
Type	Sample	**Sample**	mj negative urine h2
Acq. Method	thc quant 50 50.m	**Operator**	Anne Nord
Sample Position	P3-H2	**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	1/9/2024 8:40:25 PM		
Sample Info.			

Sample Chromatogram

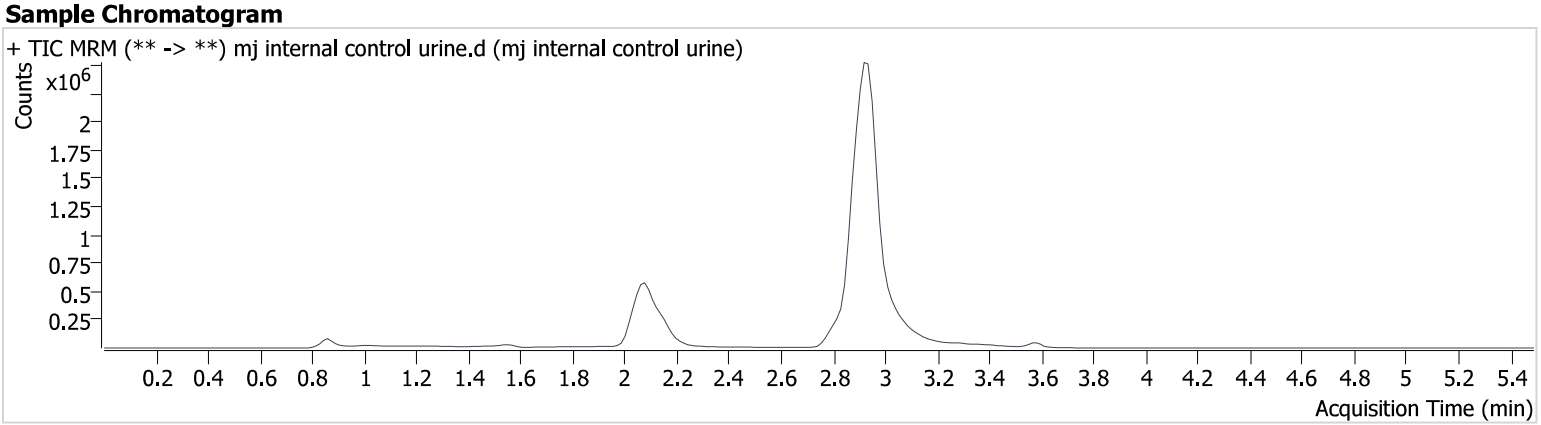


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj internal control urine.d
Type	Sample	**Sample**	mj internal control urine
Acq. Method	thc quant 50 50.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	1/9/2024 9:00:11 PM		
Sample Info.			

Sample Chromatogram



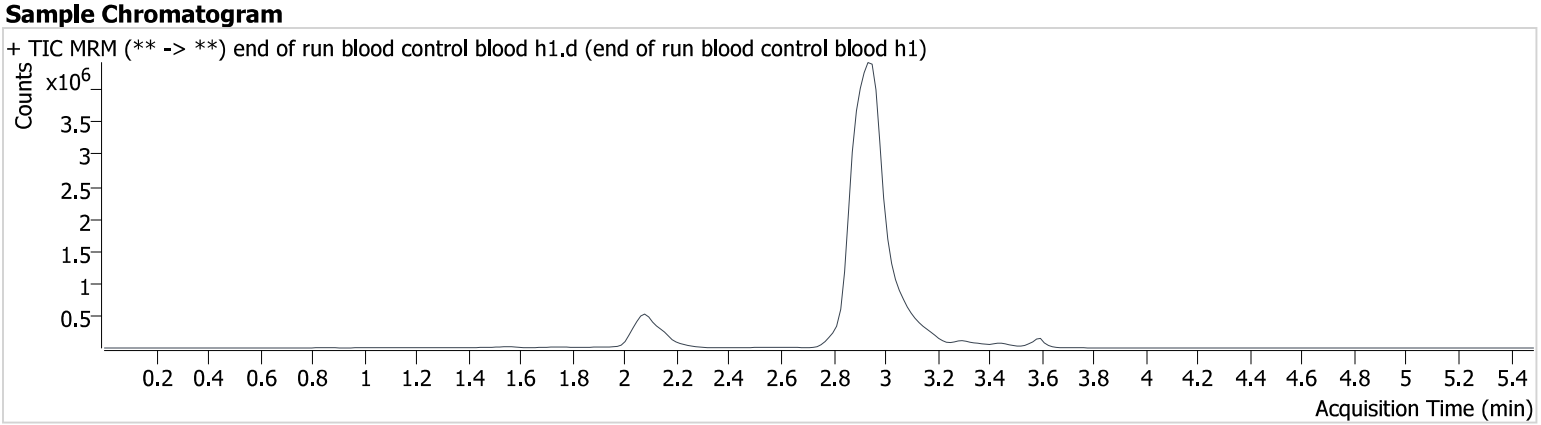
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.081	37599	∞	845.40	∞	2418041	4.721 ng/ml
THC-COOH	2.152	58144	253.4	306.11	471109.6	811601	14.995 ng/ml
THC	3.588	16026	∞	25.31	∞	128414	4.252 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 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	thc quant 50 50.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	1/9/2024 9:06:47 PM		
Sample Info.			

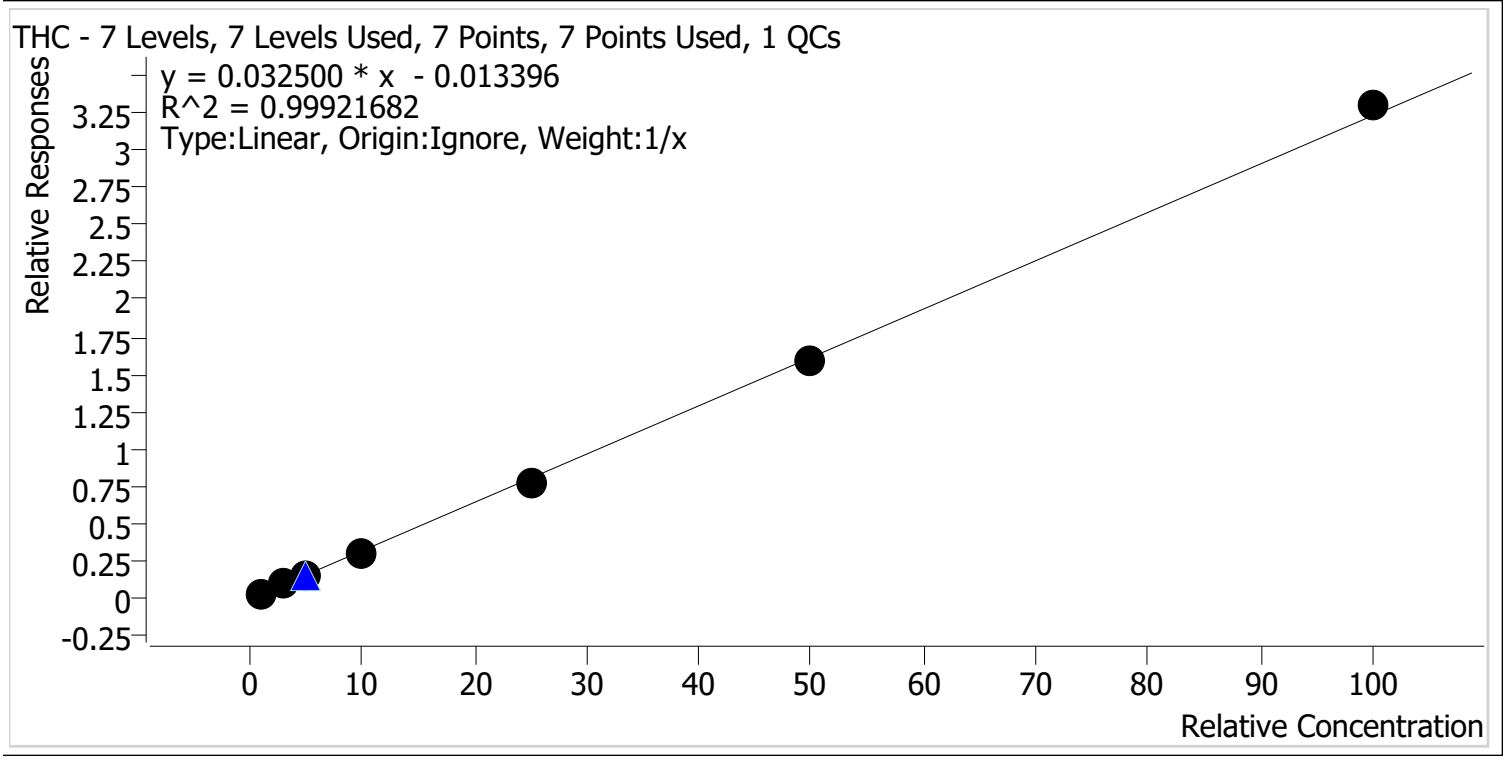
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.081	33427	∞	906.38	469.9	2142625	4.735 ng/ml
THC-COOH	2.152	55022	317.7	275.81	115.8	789551	14.624 ng/ml
THC	3.603	55512	∞	22.42	∞	360621	5.149 ng/ml

Compound Calibration Report

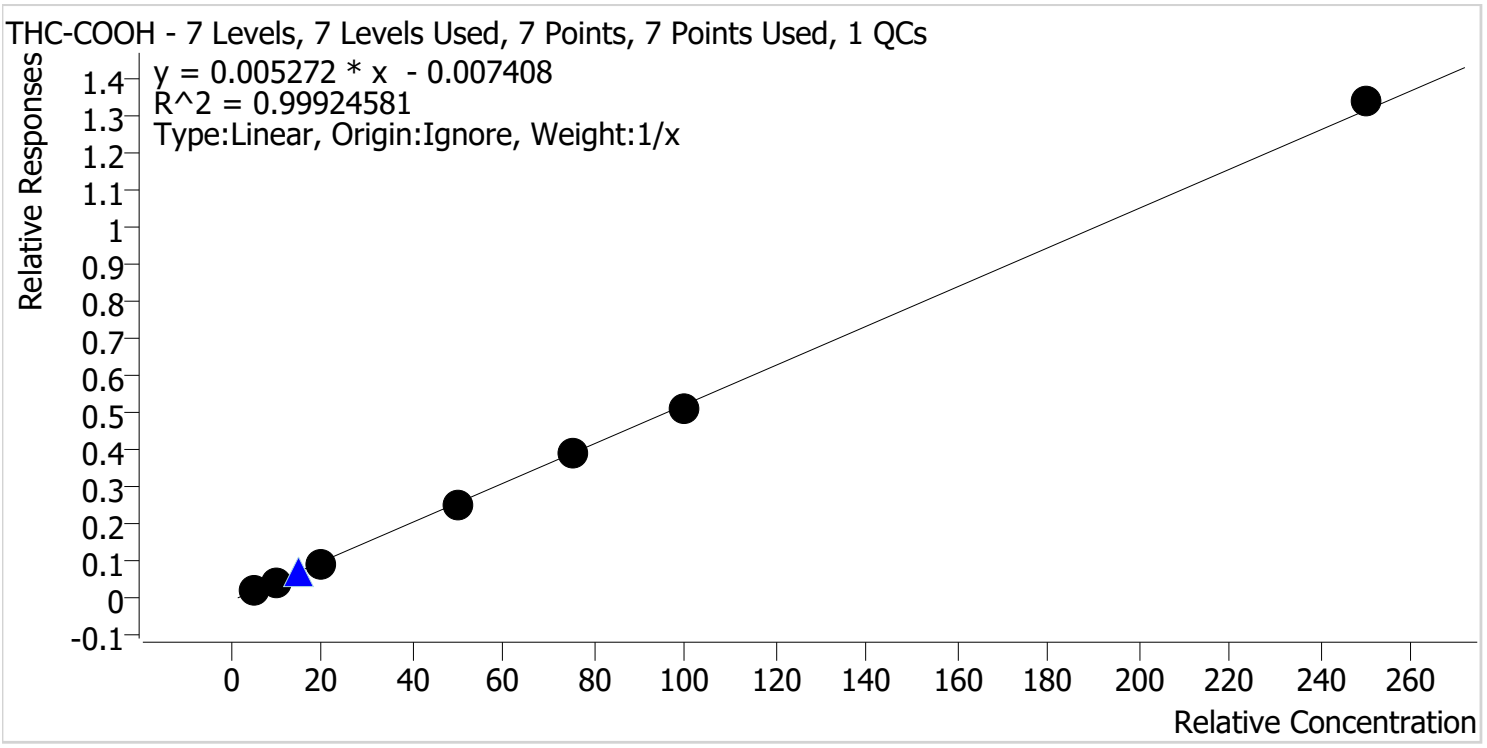
Batch results	D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Last Cal. Update	1/10/2024 10:32 AM
Analyst Name	ISP\datastor
Analyte	THC
Internal Standard	THC-d3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✔	1.0	1.1	112.1
mj cal 2	2	✔	3.0	3.0	98.9
mj cal 3	3	✔	5.0	4.8	95.7
mj cal 4	4	✔	10.0	9.7	96.5
mj cal 5	5	✔	25.0	23.9	95.6
mj cal 6	6	✔	50.0	49.5	99.1
mj cal 7	7	✔	100.0	102.0	102.0

Compound Calibration Report

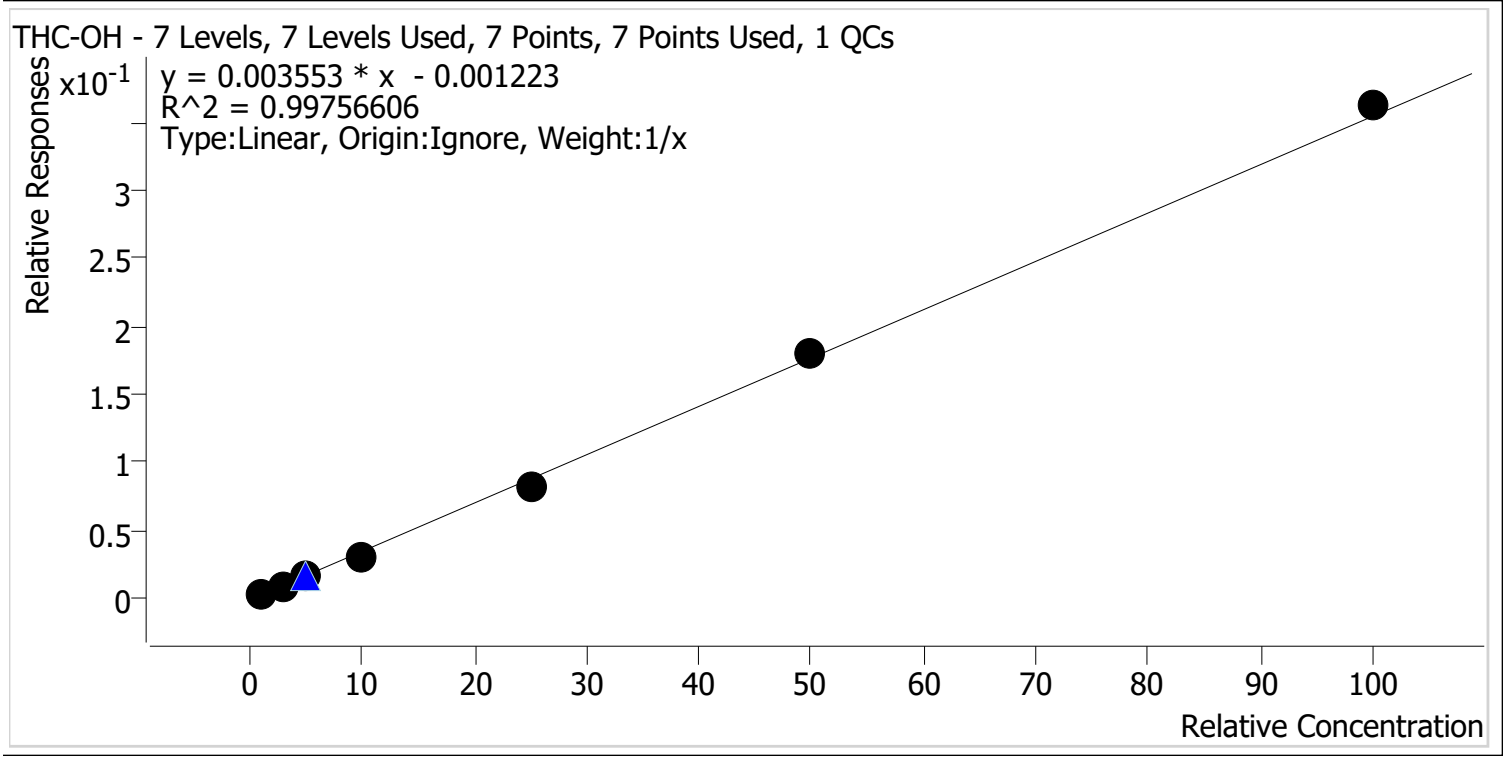
Batch results	D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Last Cal. Update	1/10/2024 10:32 AM
Analyst Name	ISP\datastor
Analyte	THC-COOH
Internal Standard	THC-COOH-d9



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✔	5.0	5.6	111.8
mj cal 2	2	✔	10.0	9.7	97.4
mj cal 3	3	✔	20.0	18.8	94.2
mj cal 4	4	✔	50.0	48.5	97.1
mj cal 5	5	✔	75.0	74.5	99.3
mj cal 6	6	✔	100.0	98.4	98.4
mj cal 7	7	✔	250.0	254.4	101.8

Compound Calibration Report

Batch results	D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Last Cal. Update	1/10/2024 10:32 AM
Analyst Name	ISP\datastor
Analyte	THC-OH
Internal Standard	THC-OH-d3



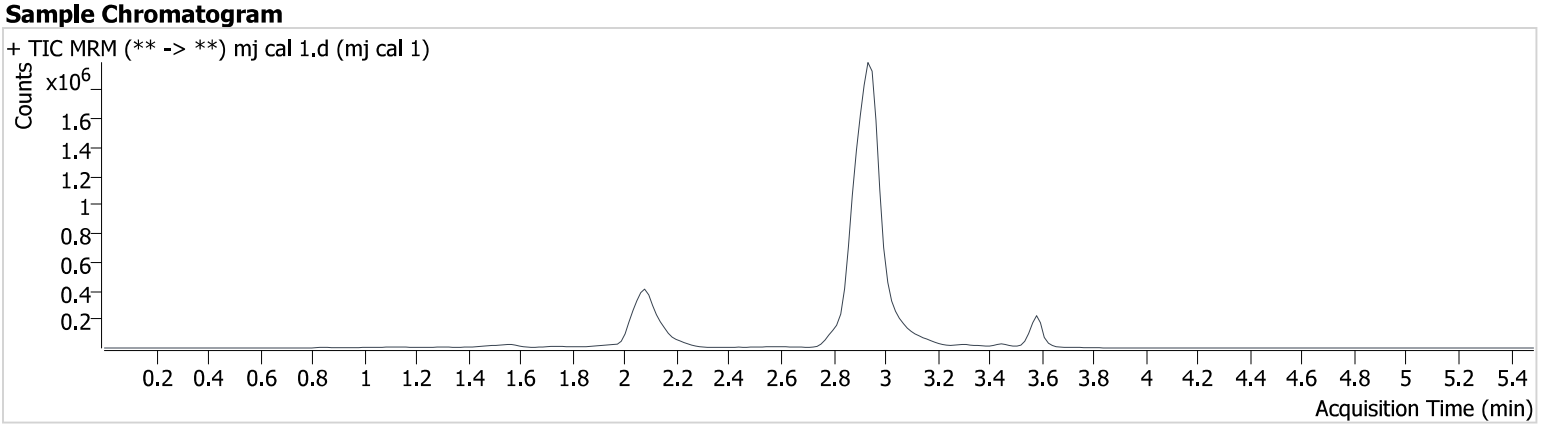
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✔	1.0	1.2	124.4
mj cal 2	2	✔	3.0	2.8	93.5
mj cal 3	3	✔	5.0	4.8	95.5
mj cal 4	4	✔	10.0	9.1	90.5
mj cal 5	5	✔	25.0	23.0	91.9
mj cal 6	6	✔	50.0	51.0	102.0
mj cal 7	7	✔	100.0	102.2	102.2

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj cal 1.d
Type	Cal	**Sample**	mj cal 1
Acq. Method	thc quant 50 50.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	1/9/2024 6:28:23 PM		
Sample Info.			

Sample Chromatogram



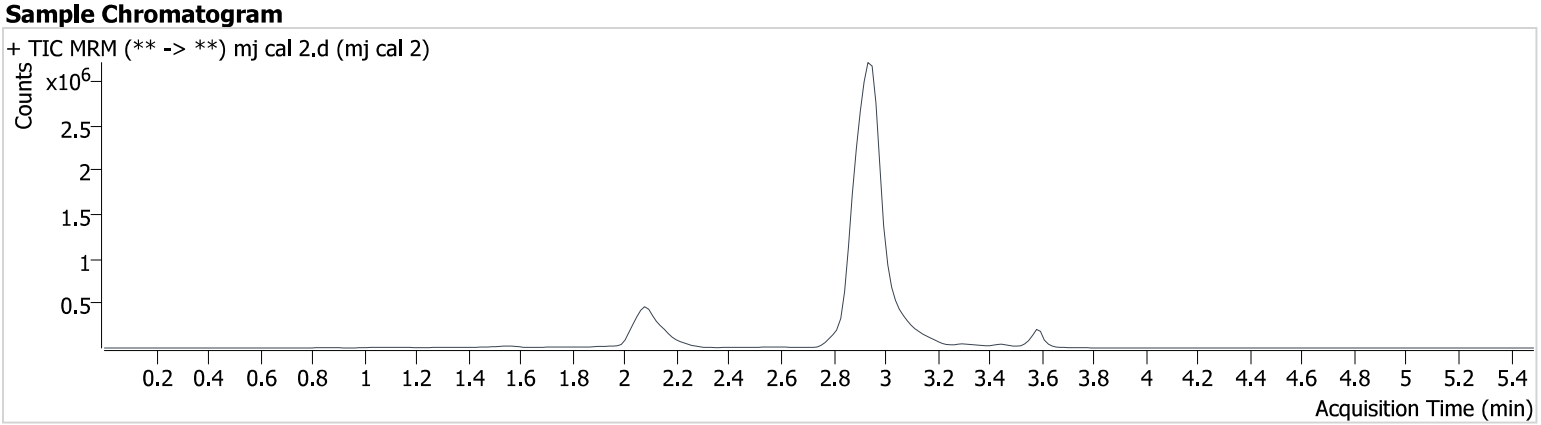
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	2.081	6166	460.6	659.59	∞	1928292	1.244 ng/ml	Low
THC-COOH	2.152	12927	89.4	293.73	260.1	585741	5.592 ng/ml	
THC	3.588	12526	2212.5	22.89	∞	543712	1.121 ng/ml	

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj cal 2.d
Type	Cal	**Sample**	mj cal 2
Acq. Method	thc quant 50 50.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	1/9/2024 6:35:07 PM		
Sample Info.			

Sample Chromatogram



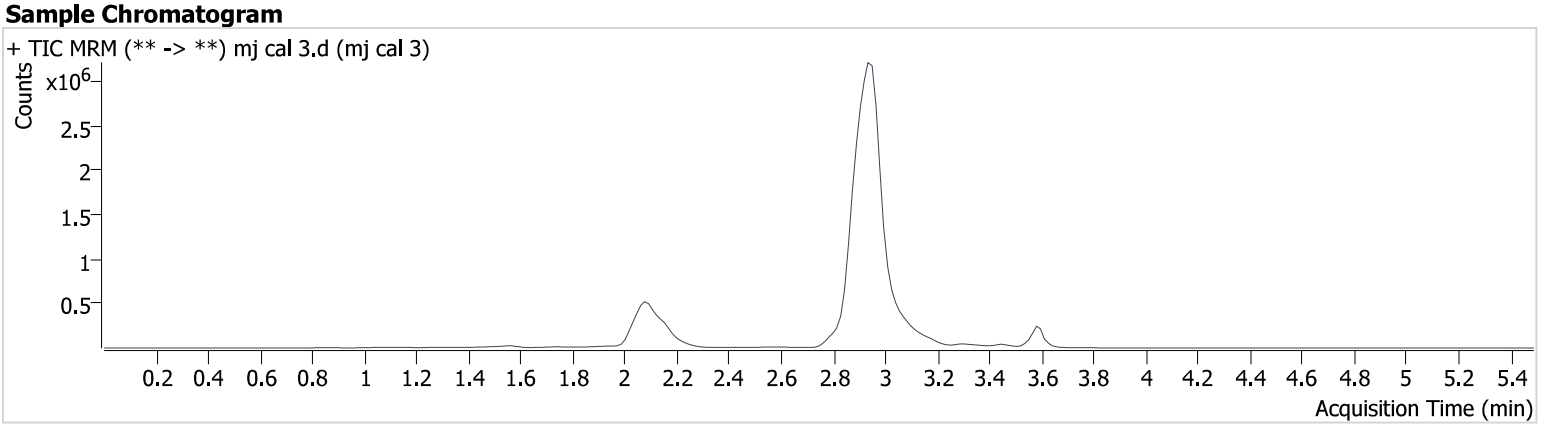
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	2.081	17608	∞	806.11	2563.9	2013891	2.805 ng/ml	Low
THC-COOH	2.152	31541	42339.0	283.90	93.7	717739	9.741 ng/ml	
THC	3.588	40888	∞	22.04	632.1	492475	2.967 ng/ml	

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj cal 3.d
Type	Cal	**Sample**	mj cal 3
Acq. Method	thc quant 50 50.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	1/9/2024 6:41:41 PM		
Sample Info.			

Sample Chromatogram



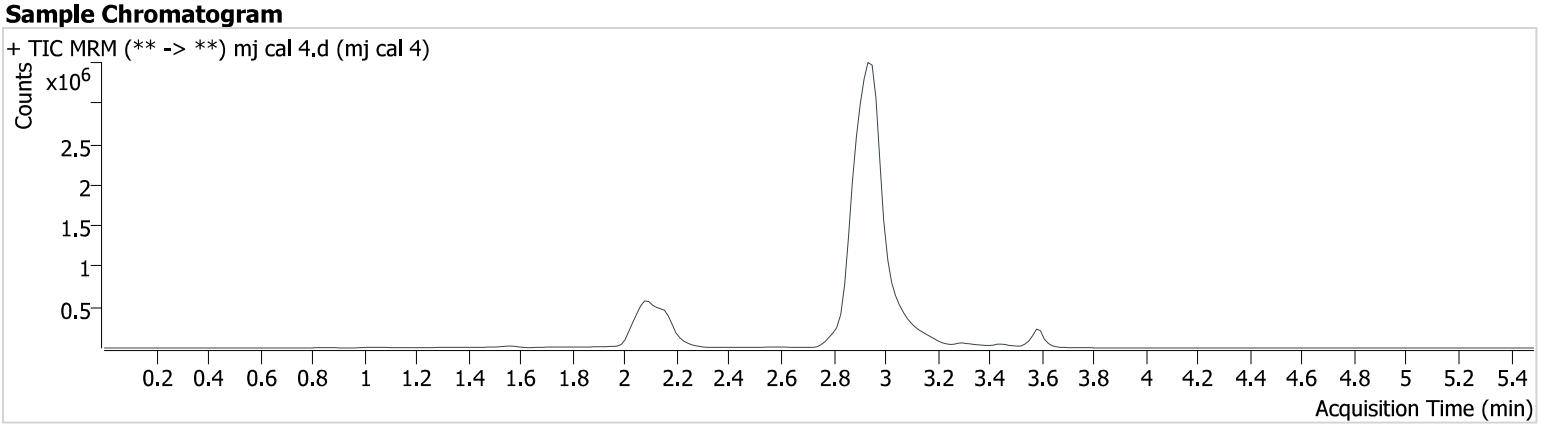
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.081	33975	∞	881.00	∞	2157268	4.777 ng/ml
THC-COOH	2.152	71445	845.5	279.77	152.7	777272	18.841 ng/ml
THC	3.588	77904	∞	25.07	∞	547870	4.787 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj cal 4.d
Type	Cal	**Sample**	mj cal 4
Acq. Method	thc quant 50 50.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	1/9/2024 6:48:17 PM		
Sample Info.			

Sample Chromatogram



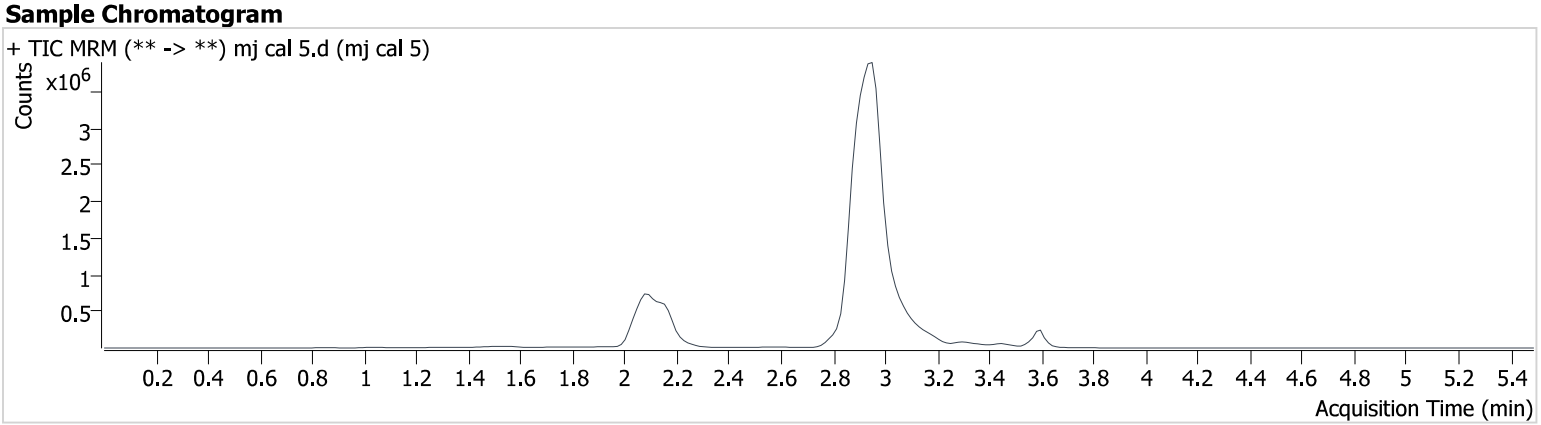
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.081	64776	45675.6	851.68	∞	2093172	9.054 ng/ml
THC-COOH	2.152	186608	752.0	267.75	458.7	751012	48.538 ng/ml
THC	3.588	139661	∞	22.08	∞	465034	9.653 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj cal 5.d
Type	Cal	**Sample**	mj cal 5
Acq. Method	thc quant 50 50.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	1/9/2024 6:54:53 PM		
Sample Info.			

Sample Chromatogram



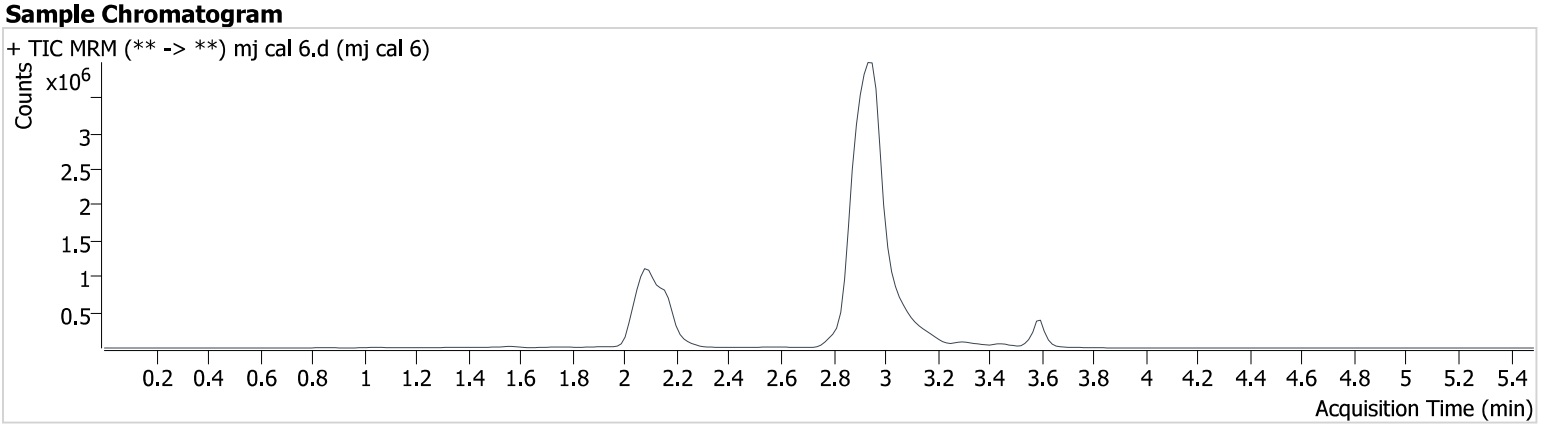
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.081	159615	∞	845.79	∞	1985549	22.969 ng/ml
THC-COOH	2.152	269871	4583.6	265.05	940.1	700608	74.472 ng/ml
THC	3.603	277180	∞	22.74	∞	362992	23.907 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj cal 6.d
Type	Cal	**Sample**	mj cal 6
Acq. Method	thc quant 50 50.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	1/9/2024 7:01:29 PM		
Sample Info.			

Sample Chromatogram



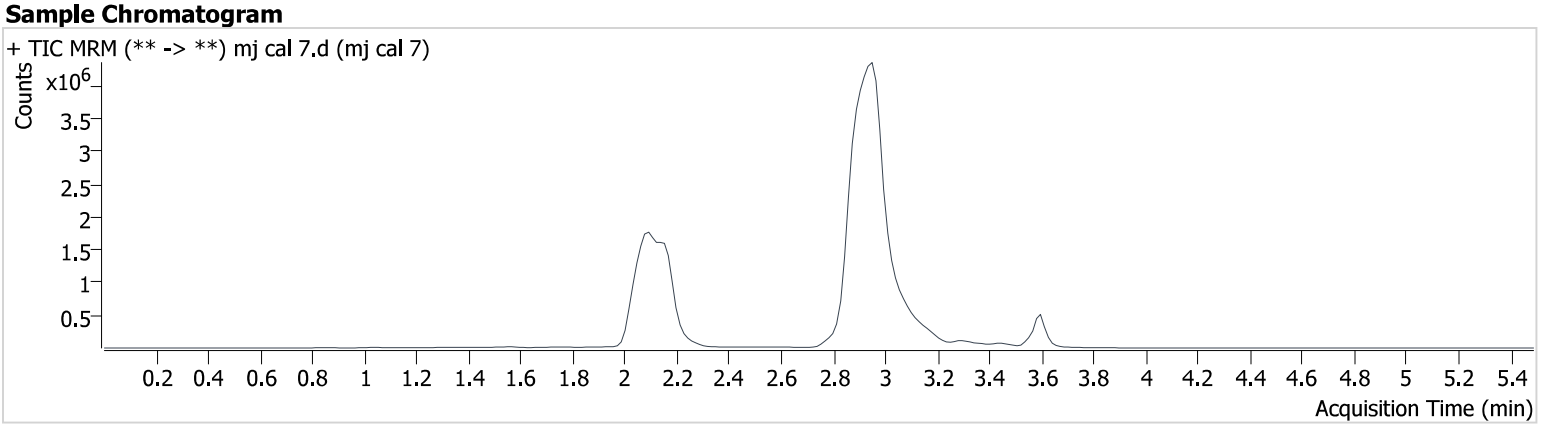
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.081	377476	5009.4	790.94	∞	2098278	50.975 ng/ml
THC-COOH	2.152	380625	3074.8	267.89	1853.9	744113	98.433 ng/ml
THC	3.603	635442	∞	24.87	∞	398036	49.533 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2024\am 27-28\010924\QuantResults\am27.batch.bin
Calibration Last Update 1/10/2024 10:32:05 AM

Instrument	69679	**Data File**	mj cal 7.d
Type	Cal	**Sample**	mj cal 7
Acq. Method	thc quant 50 50.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	1/9/2024 7:08:05 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.081	743128	∞	779.67	∞	2053856	102.176 ng/ml
THC-COOH	2.152	908055	1565.4	266.97	838059 5.0	680868	254.385 ng/ml
THC	3.603	1027304	∞	24.93	∞	311053	102.032 ng/ml