REVIEWED
By Tamara Salazar at 8:02 am, Dec 19, 2023

12/18/2023

Worklist: 6613

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION	
C2023-2663	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2687	2	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2023-2693	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2701	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2706	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2720	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-2759	4	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2023-2775	1	AVK	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 12/14/23 Analyst: Anne Nord
Plate lot#: 230627 Plate re-test: 12/27/2023

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:** 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 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: The autosampler failed to align on 12/14/23. The plate was placed in the freezer. On 12/15/23 the autosampler was aligned and the samples were injected. The end of run blood control evaporated. On 12/16/23 it was reconstituted and injected that sample failed to inject. On 12/18/23 it was reconstituted and injected that injection was evaluated.

	1	2	3	4	5	6
a	cal 1	Internal control urine	negative urine			
b	cal 2	negative blood	2687-2			
c	cal 3	2663-1	2759-4			
d	cal 4	2693-1	2706-1 SLE and injection plate			
e	cal 5	2701-1 ~~mixing plate~~ 12/18/23				
f	cal 6	2706-1 Mixing plate 12/18/23				
g	cal 7	2720-1				
h	Internal control (blood)	2775-1				

Plate position 3

c2023-____-_

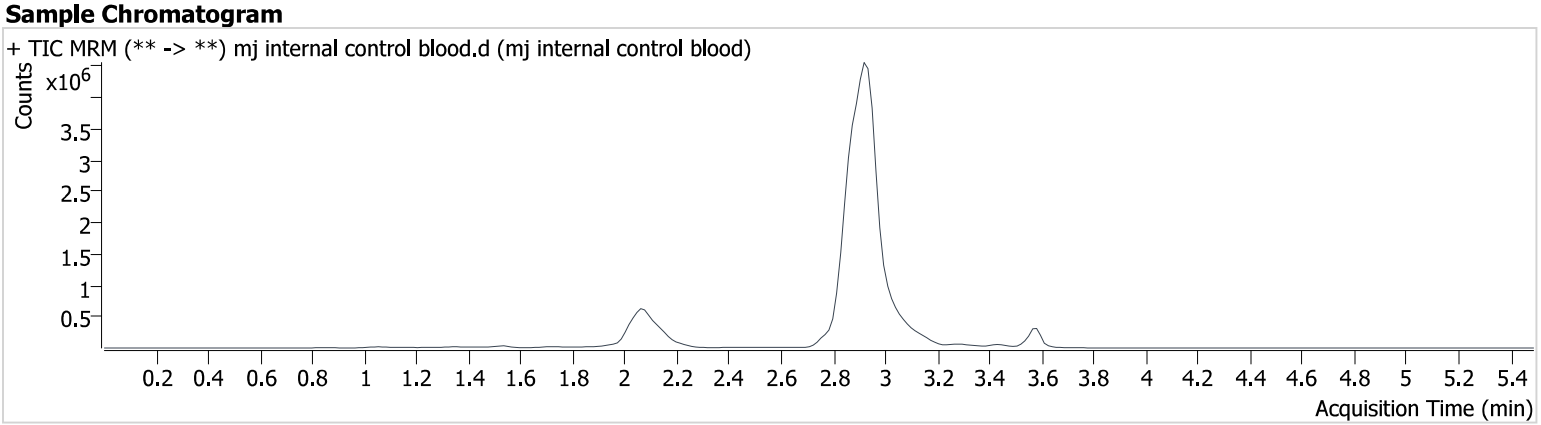
AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

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**	
Injection Volume	10		
Acq. Date-Time	12/15/2023 6:42:57 PM		
Sample Info.			

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

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.066	38010	∞	850.49	∞	2794203	4.565 ng/ml
THC-COOH	2.137	66618	189.4	283.98	37161.5	897421	14.154 ng/ml
THC	3.588	97253	7580.5	24.09	169.9	799150	4.079 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

Instrument	69679	**Data File**	mj negative blood.d
Type	Sample	**Sample**	mj negative blood
Acq. Method	thc quant 50 50.m	**Operator**	Anne Nord
Sample Position	P3-B2	**Comment**	
Injection Volume	10		
Acq. Date-Time	12/15/2023 6:49:31 PM		
Sample Info.			

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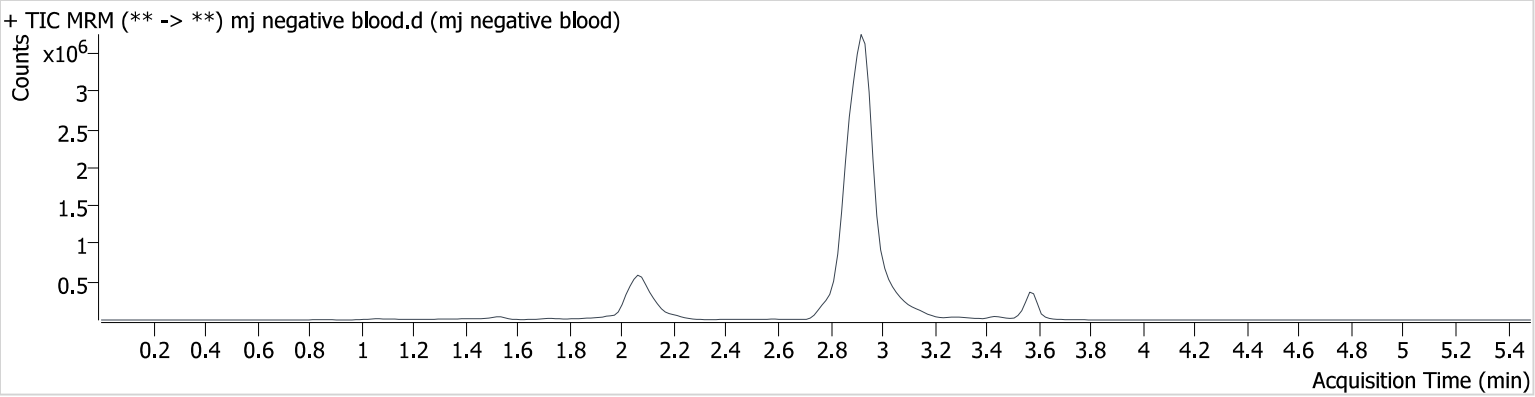
Sample Chromatogram

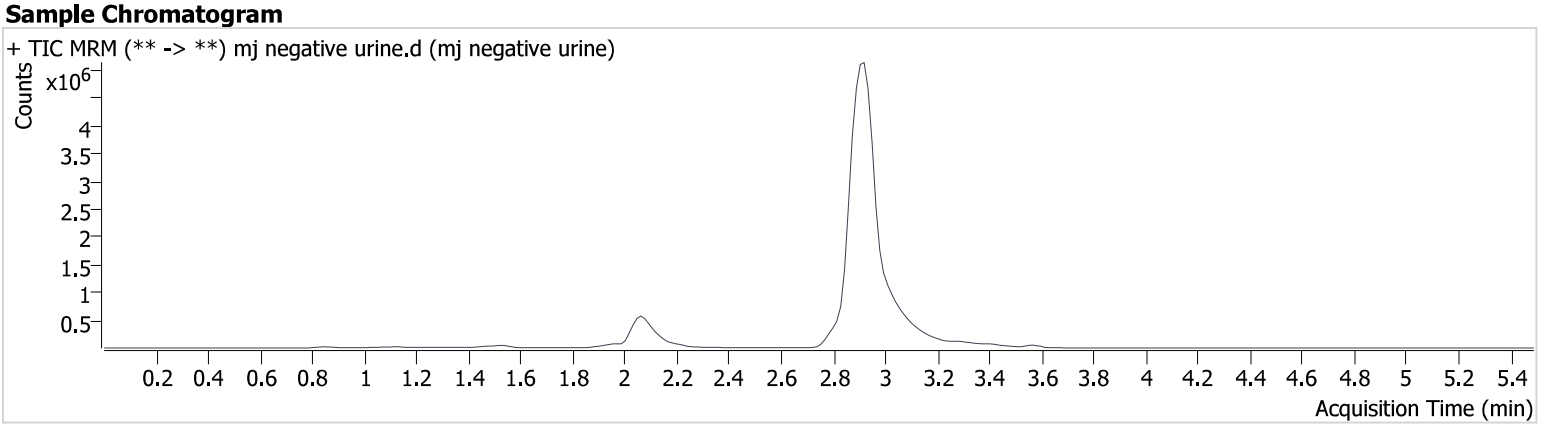


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

Instrument	69679	**Data File**	mj negative urine.d
Type	Sample	**Sample**	mj negative urine
Acq. Method	thc quant 50 50.m	**Operator**	Anne Nord
Sample Position	P3-A3	**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	12/15/2023 8:21:44 PM		
Sample Info.			

Sample Chromatogram

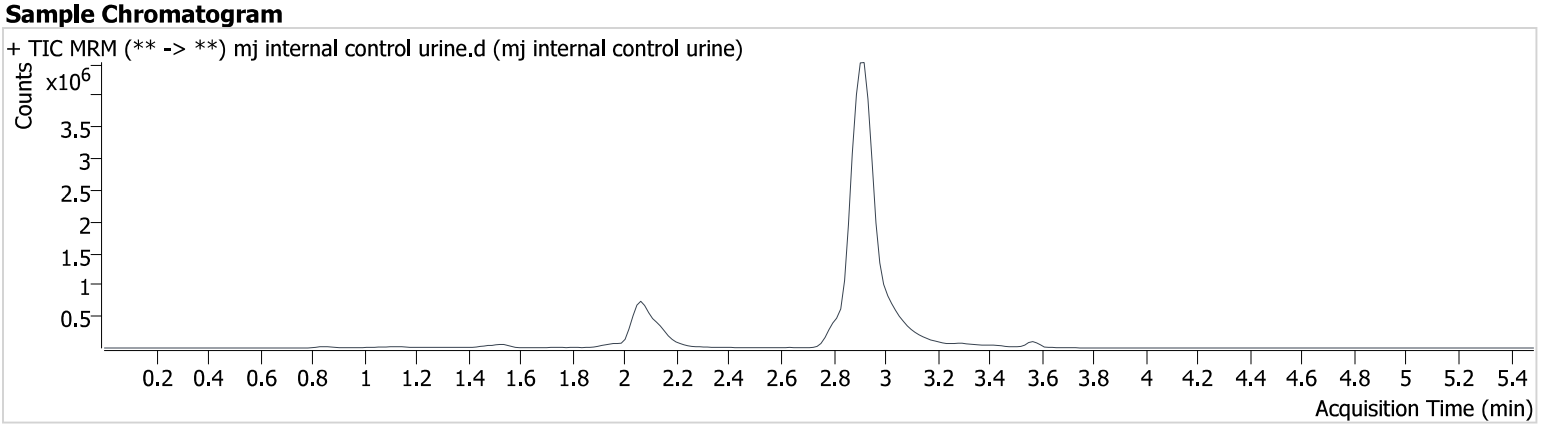


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

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	12/15/2023 8:54:38 PM		
Sample Info.			

Sample Chromatogram



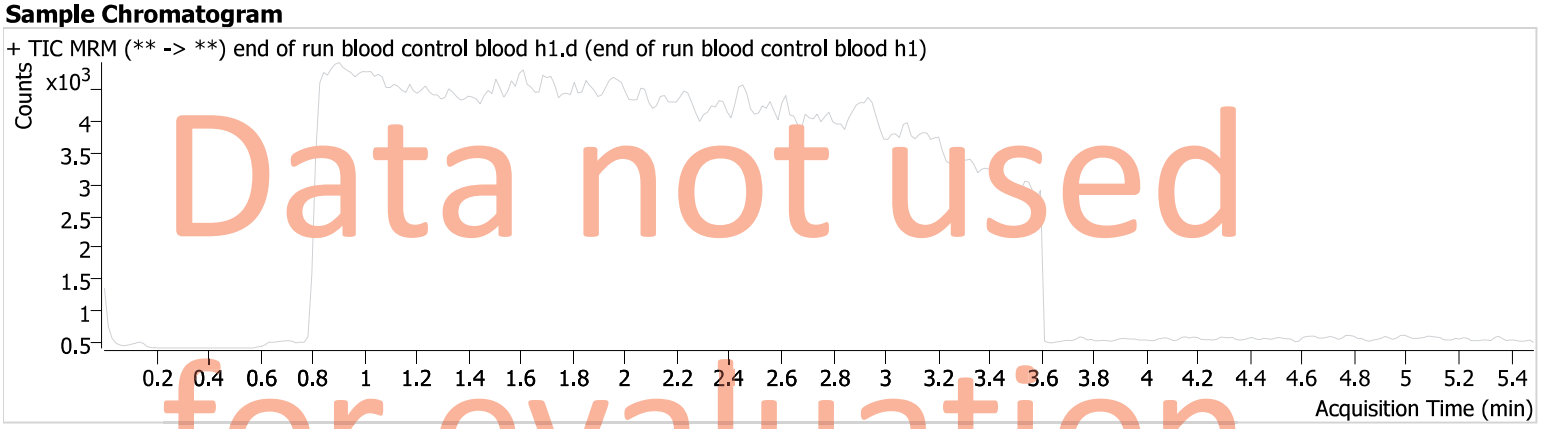
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.066	35048	3049.8	869.22	∞	2775672	4.260 ng/ml
THC-COOH	2.137	64723	69022.4	287.01	426.8	942926	13.180 ng/ml
THC	3.573	28584	467.4	26.67	5043.9	282811	3.449 ng/ml

AM #27 Cannabinoids

Batch results	D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update	12/18/2023 1:08:17 PM

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	12/15/2023 9:01:12 PM		
Sample Info.			

Sample Chromatogram



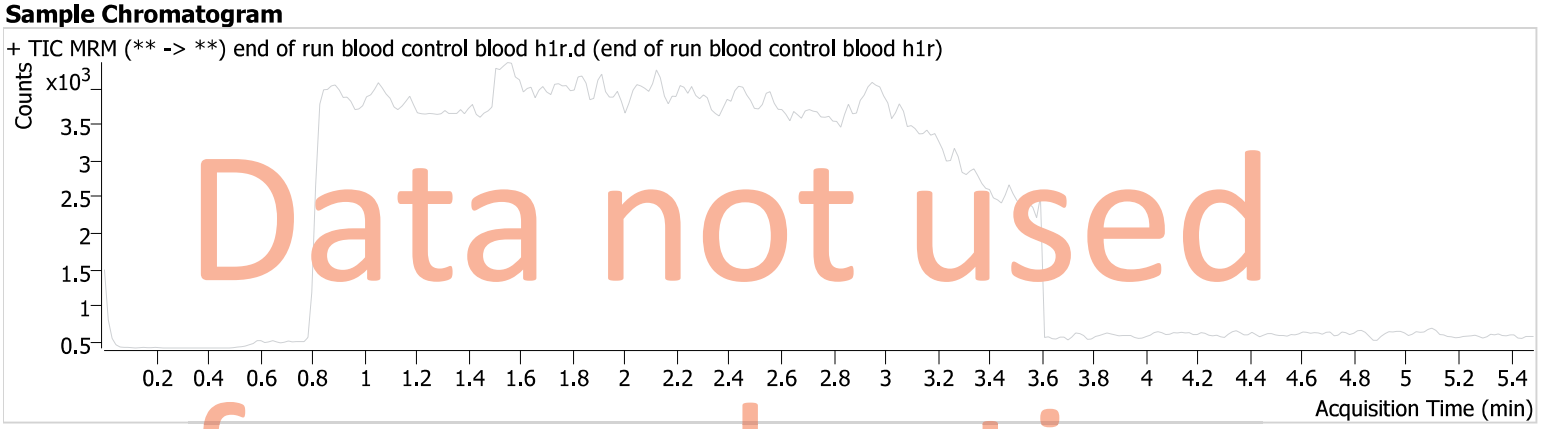
sample evaporated and did not inject

AM #27 Cannabinoids

Batch results	D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update	12/18/2023 1:08:17 PM

Instrument	69679	**Data File**	end of run blood control blood h1r.d
Type	Sample	**Sample**	end of run blood control blood h1r
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	12/16/2023 3:59:45 PM		
Sample Info.			

Sample Chromatogram



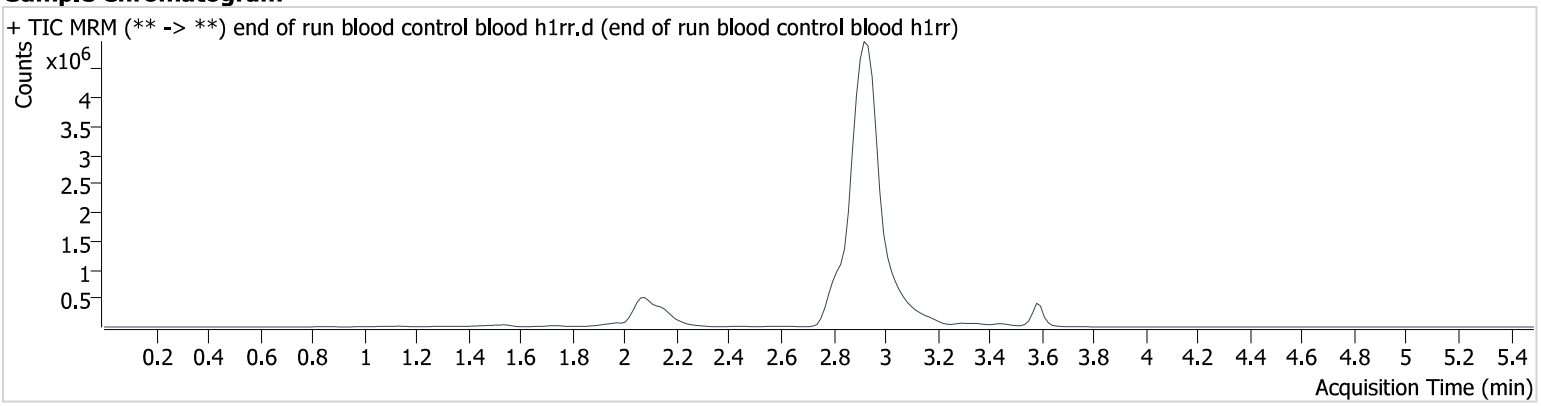
sample reconstituted 12/16/23, it did not inject

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

Instrument	69679	**Data File**	end of run blood control blood h1rr.d
Type	Sample	**Sample**	end of run blood control blood h1rr
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	12/18/2023 11:49:32 AM		
Sample Info.			

Sample Chromatogram

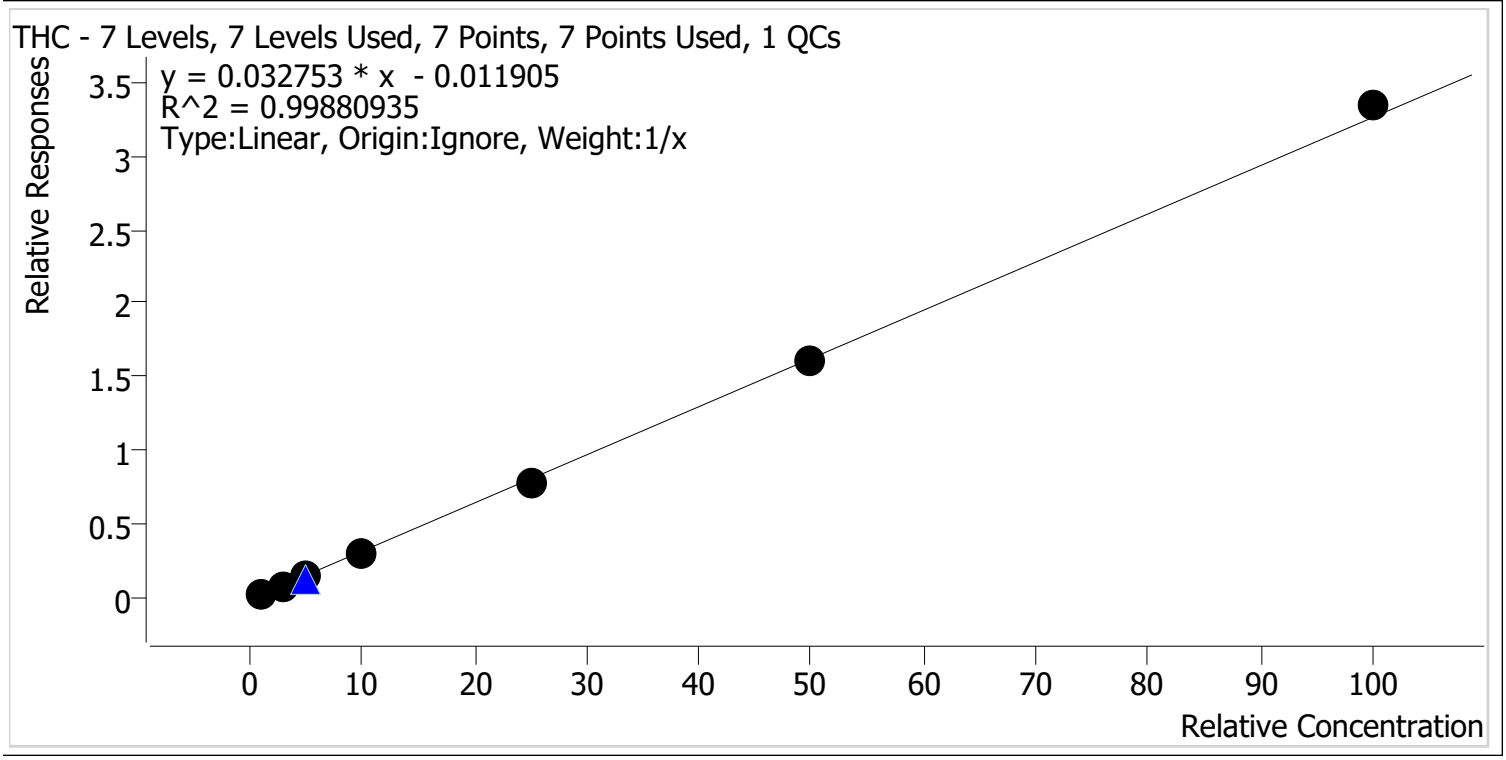


Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.081	30073	∞	754.60	∞	1978618	5.063 ng/ml
THC-COOH	2.152	63228	241.9	269.51	3136.6	812178	14.784 ng/ml
THC	3.588	105038	2406.9	23.98	395.8	867752	4.059 ng/ml

Sample reconstituted and injected 12/18/23 this sample was evaluated.

Compound Calibration Report

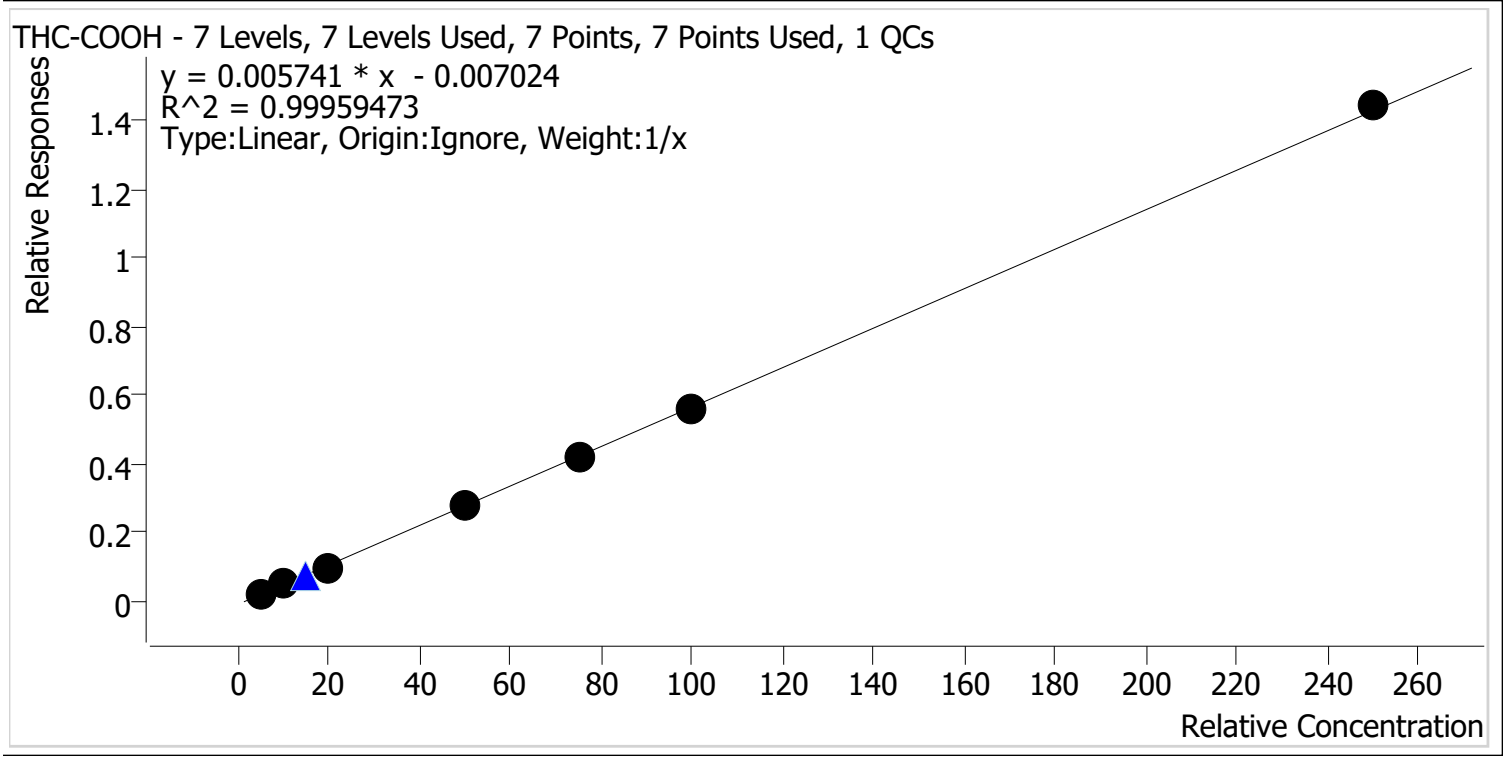
Batch results	D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Last Cal. Update	12/18/2023 1:08 PM
Analyst Name	ISP\datastor
Analyte	THC
Internal Standard	THC-d3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✔	1.0	1.2	118.5
mj cal 2	2	✔	3.0	2.8	94.9
mj cal 3	3	✔	5.0	4.8	96.1
mj cal 4	4	✔	10.0	9.3	92.6
mj cal 5	5	✔	25.0	24.2	97.0
mj cal 6	6	✔	50.0	49.2	98.5
mj cal 7	7	✔	100.0	102.4	102.4

Compound Calibration Report

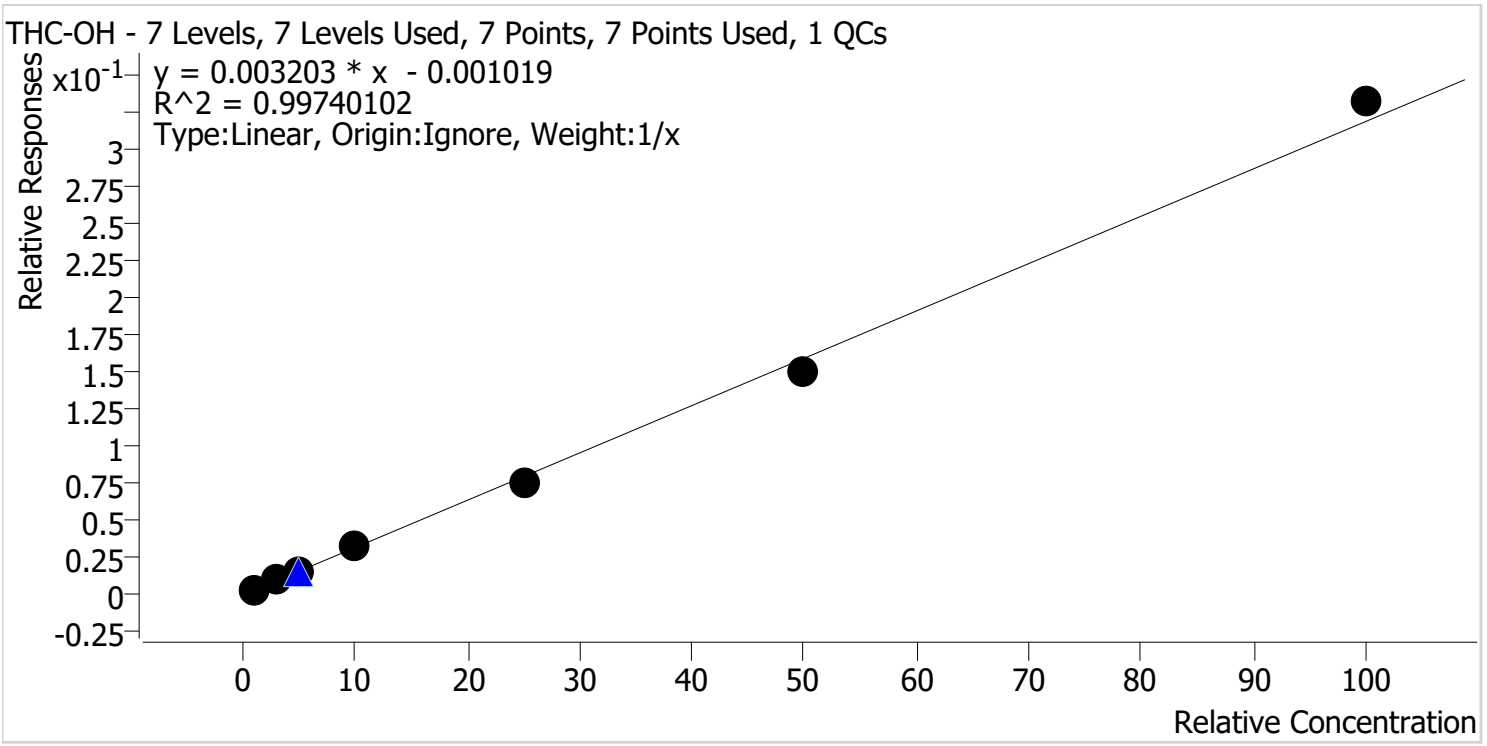
Batch results	D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Last Cal. Update	12/18/2023 1:08 PM
Analyst Name	ISP\datastor
Analyte	THC-COOH
Internal Standard	THC-COOH-d9



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✔	5.0	5.3	106.7
mj cal 2	2	✔	10.0	10.0	100.5
mj cal 3	3	✔	20.0	18.7	93.7
mj cal 4	4	✔	50.0	49.9	99.9
mj cal 5	5	✔	75.0	74.8	99.8
mj cal 6	6	✔	100.0	98.4	98.4
mj cal 7	7	✔	250.0	252.7	101.1

Compound Calibration Report

Batch results	D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Last Cal. Update	12/18/2023 1:08 PM
Analyst Name	ISP\datastor
Analyte	THC-OH
Internal Standard	THC-OH-d3



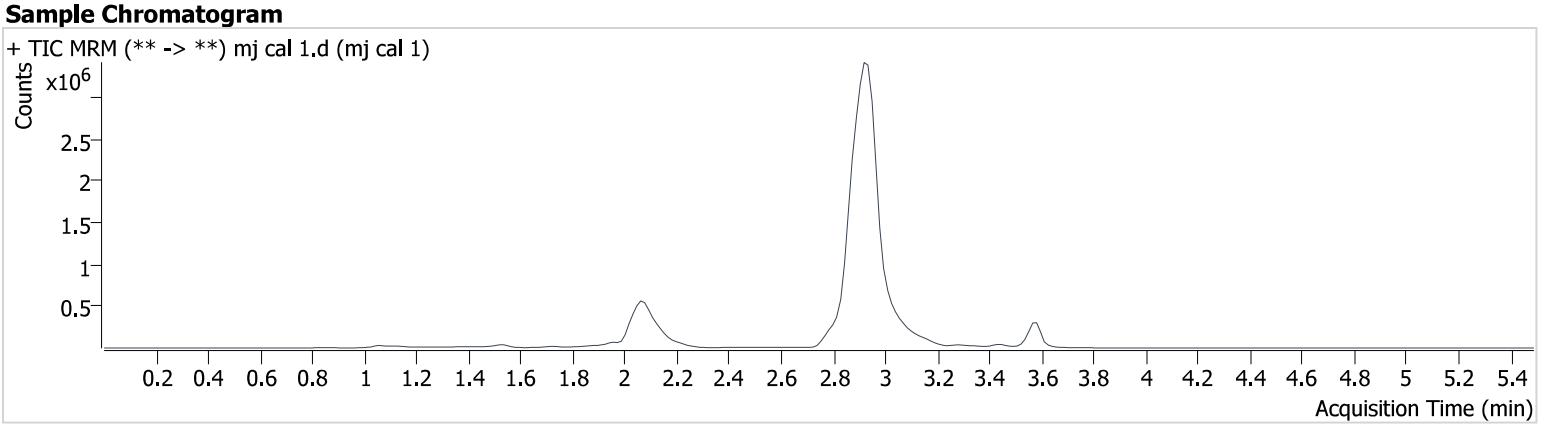
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✔	1.0	1.1	110.7
mj cal 2	2	✔	3.0	3.1	101.9
mj cal 3	3	✔	5.0	4.7	93.6
mj cal 4	4	✔	10.0	10.1	100.7
mj cal 5	5	✔	25.0	23.5	94.1
mj cal 6	6	✔	50.0	47.4	94.8
mj cal 7	7	✔	100.0	104.2	104.2

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

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	12/15/2023 5:56:49 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	2.081	6386	207.8	743.60	∞	2527725	1.107 ng/ml	Low
THC-COOH	2.137	19195	34074.7	292.11	80674.1	813323	5.334 ng/ml	
THC	3.588	22244	∞	24.47	42.1	826949	1.185 ng/ml	

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

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**	
Injection Volume	10		
Acq. Date-Time	12/15/2023 6:03:33 PM		
Sample Info.			

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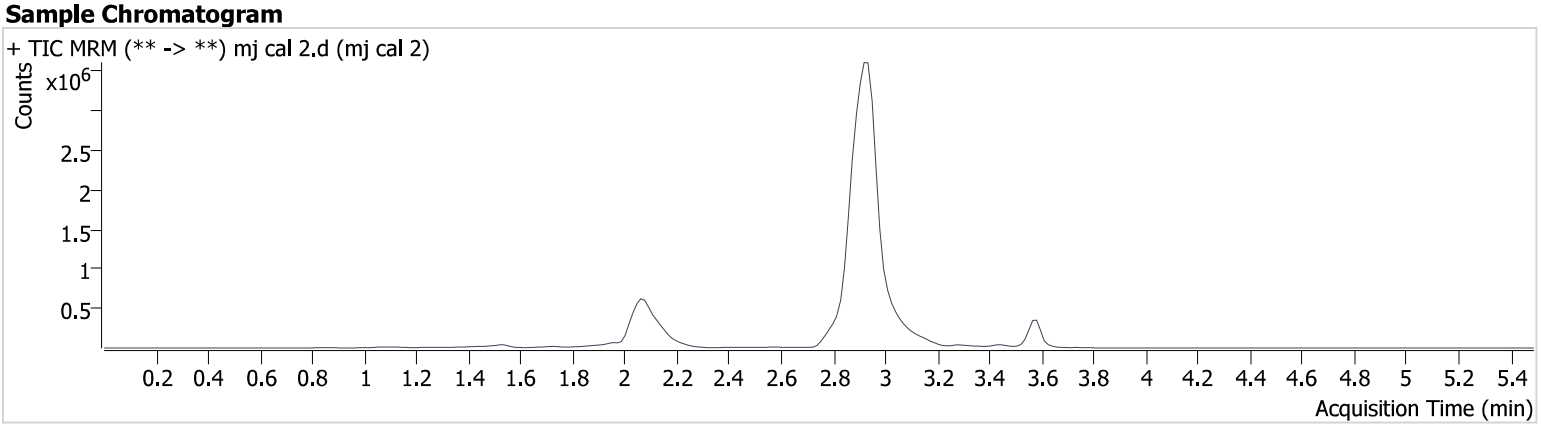
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.066	22997	∞	861.01	∞	2621221	3.057 ng/ml
THC-COOH	2.137	44779	155.3	261.25	168.7	883793	10.049 ng/ml
THC	3.588	73821	∞	24.56	∞	907684	2.847 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

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**	
Injection Volume	10		
Acq. Date-Time	12/15/2023 6:10:07 PM		
Sample Info.			

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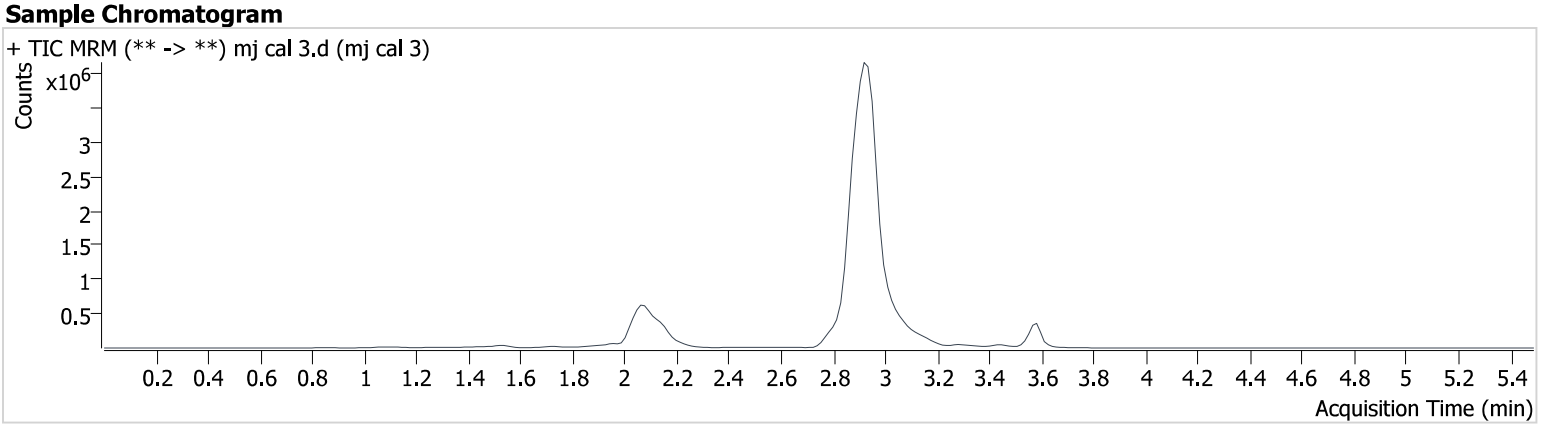
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.066	34882	∞	938.59	∞	2495110	4.682 ng/ml
THC-COOH	2.137	86114	290.2	280.70	457.1	856334	18.740 ng/ml
THC	3.588	122976	∞	25.52	∞	844979	4.807 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

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**	
Injection Volume	10		
Acq. Date-Time	12/15/2023 6:16:41 PM		
Sample Info.			

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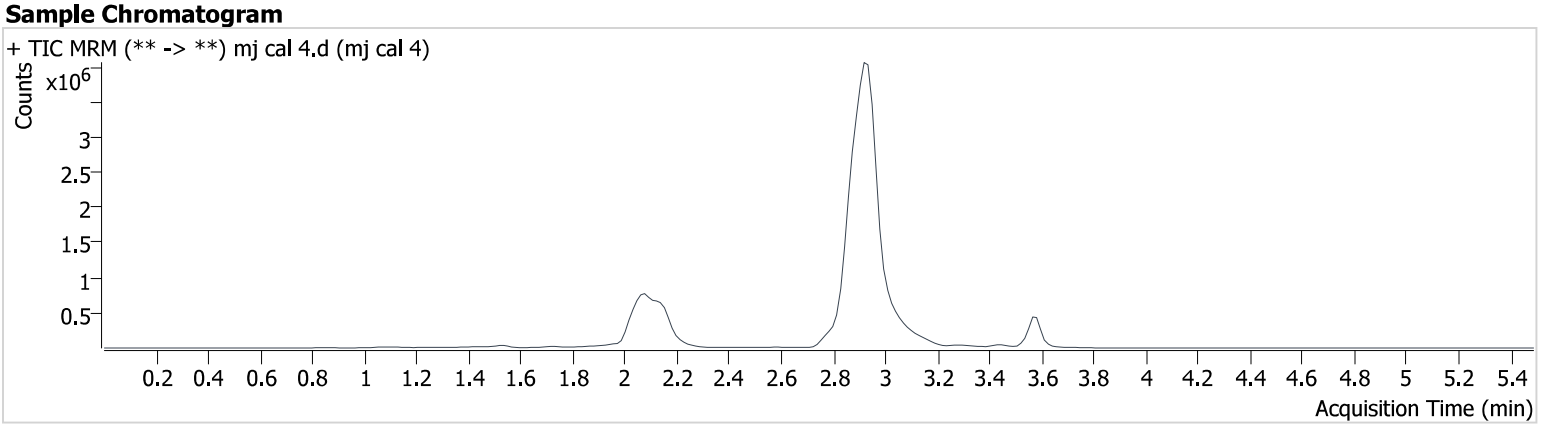
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.066	87142	736.5	857.47	∞	2788603	10.073 ng/ml
THC-COOH	2.137	259303	387346.2	266.26	1934.2	927323	49.930 ng/ml
THC	3.588	282306	∞	25.09	∞	968458	9.264 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

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**	
Injection Volume	10		
Acq. Date-Time	12/15/2023 6:23:15 PM		
Sample Info.			

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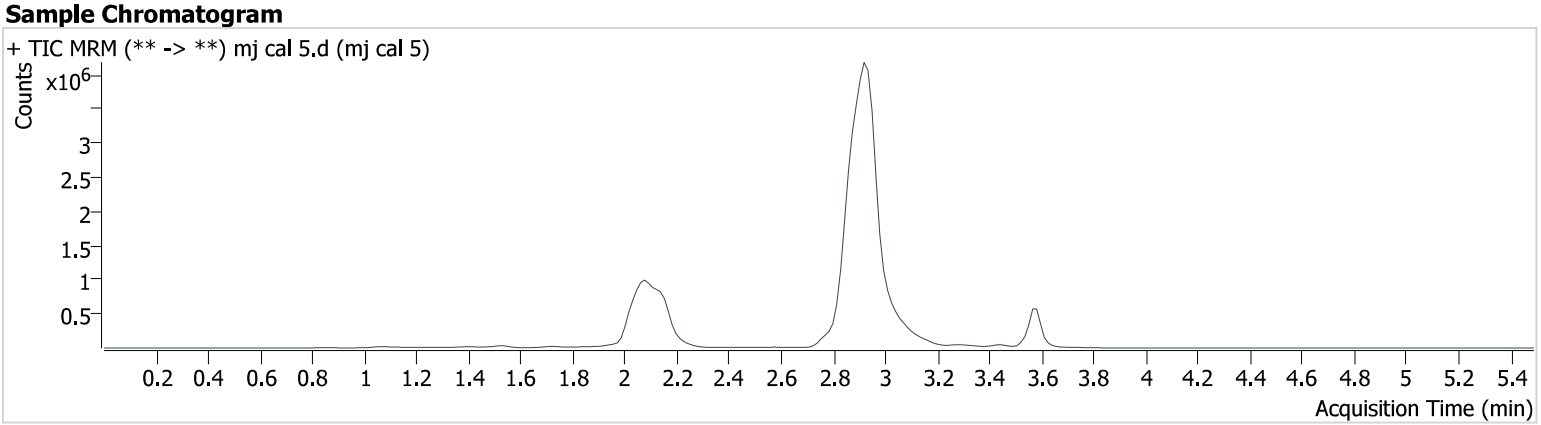
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.066	212102	17115.4	893.91	∞	2853397	23.523 ng/ml
THC-COOH	2.137	380422	1852.7	264.39	1839.0	900397	74.817 ng/ml
THC	3.588	711497	17010.0	25.25	788.2	909673	24.244 ng/ml

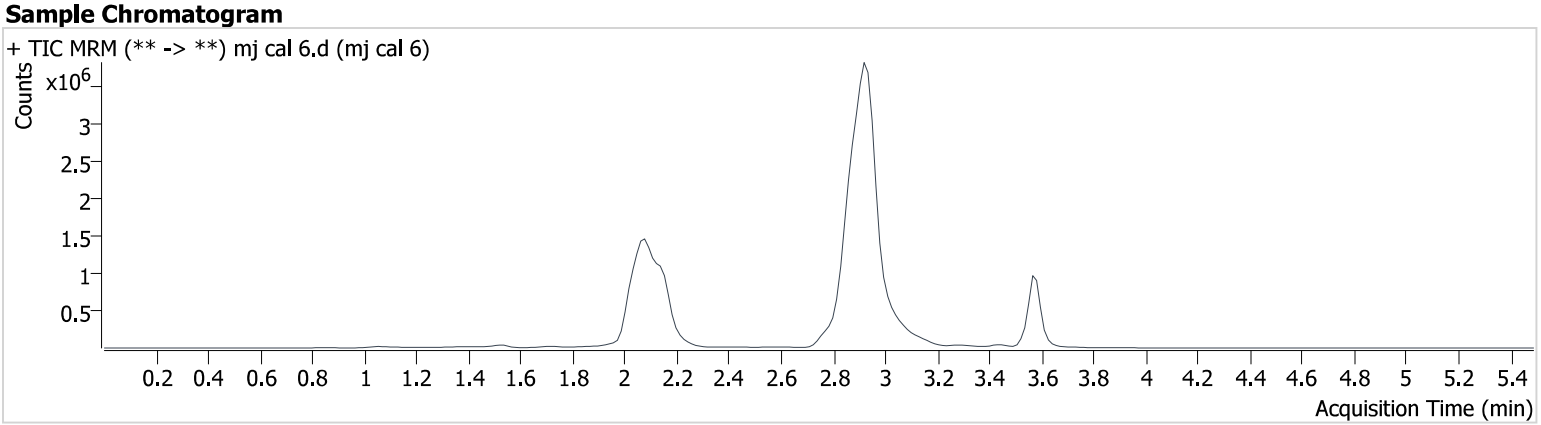
AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

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**	
Injection Volume	10		
Acq. Date-Time	12/15/2023 6:29:49 PM		
Sample Info.			

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

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.066	459267	11787.3	886.31	∞	3045861	47.389 ng/ml
THC-COOH	2.137	521447	485.4	267.87	2400.7	934453	98.423 ng/ml
THC	3.573	1642353	44410.6	25.52	∞	1026090	49.232 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\121423\QuantResults\am 27.batch.bin
Calibration Last Update 12/18/2023 1:08:17 PM

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**	
Injection Volume	10		
Acq. Date-Time	12/15/2023 6:36:23 PM		
Sample Info.			

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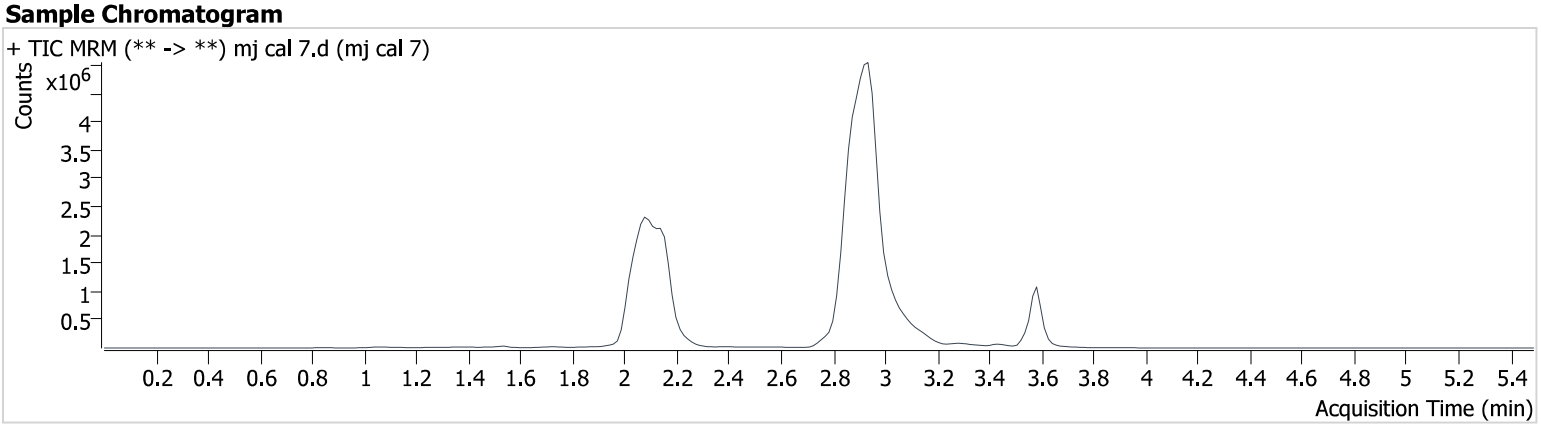
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	2.066	968870	12468.3	818.27	∞	2912426	104.168 ng/ml
THC-COOH	2.137	1201517	1383.4	265.29	3351.2	832203	252.707 ng/ml
THC	3.588	2272979	9098257410 98078.0	26.37	7387.3	679985	102.422 ng/ml