REVIEWED
By Amber Gerheart at 6:05 pm, Jun 08, 2023

6/7/2023

Worklist: 6398

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION	
C2023-0991	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2023-1048	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2023-1083	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1095	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1106	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1133	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1141	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1144	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2023-1168	2	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1185	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1188	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1212	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1213	2	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1225	6	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2023-1231	1	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 6/6/23 Analyst: Anne Nord
Plate lot#: 230113 Plate re-test: 7/13/23

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: 22B52016-1 **Urine Blank:** 12522 **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: K52558G** 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: . *Carboxy-THC was not evaluated in this run due to an interfering peak in the blank blood causing the ratios to be out of range on the lower concentrations. On 6/7/23 I made and adjustment to the acquisition method to tried and resolve it after running a few calibrators it was clear that would not resolve it. I did not evaluate or print those additional injections.*

	1	2	3	4	5	6
a	cal 1	Internal control urine	1185-1	1048-1		
b	cal 2	negative blood	1188-1 mixing plate	1144-1		
c	cal 3	1083-1	1212-1	1188-1 SLE and injection		
d	cal 4	1095-1	1213-2			
e	Cal 5	1106-1	1225-6			
f	cal 6	1133-1	1231-1			
g	cal 7	1141-1	negative urine			
h	Internal control (blood)	1168-2	991-1			

Plate position 3

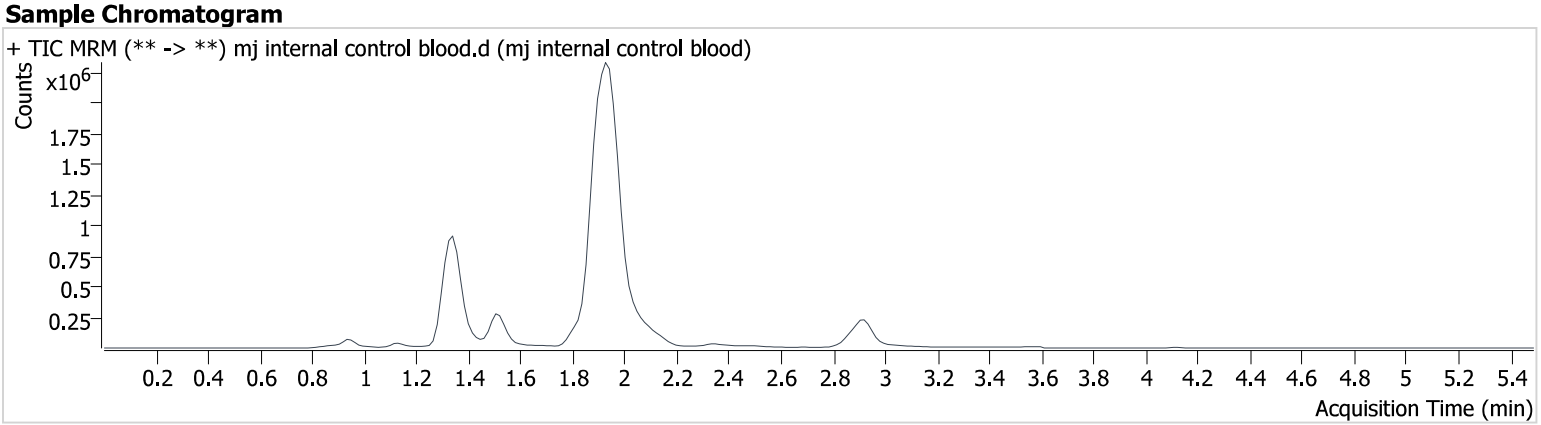
c2023-0____-_

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 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**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 7:46:13 PM		
Sample Info.			

Sample Chromatogram



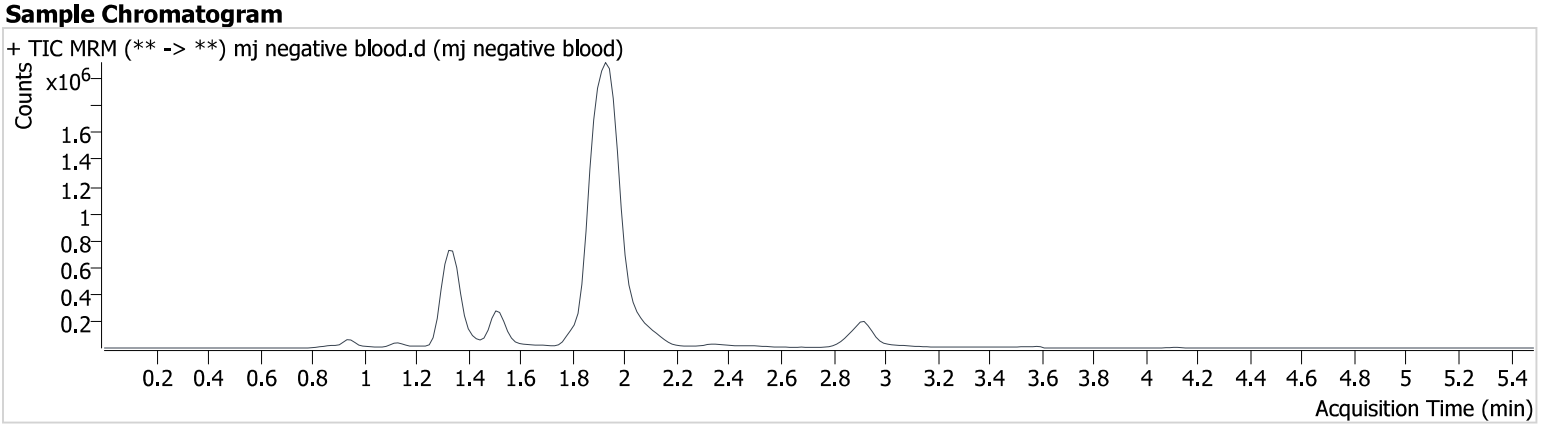
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.334	44174	∞	866.95	∞	2502609	4.200 ng/ml
THC	2.926	161591	5669533949 5357.3	26.67	∞	1204901	4.682 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 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**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 7:52:47 PM		
Sample Info.			

Sample Chromatogram

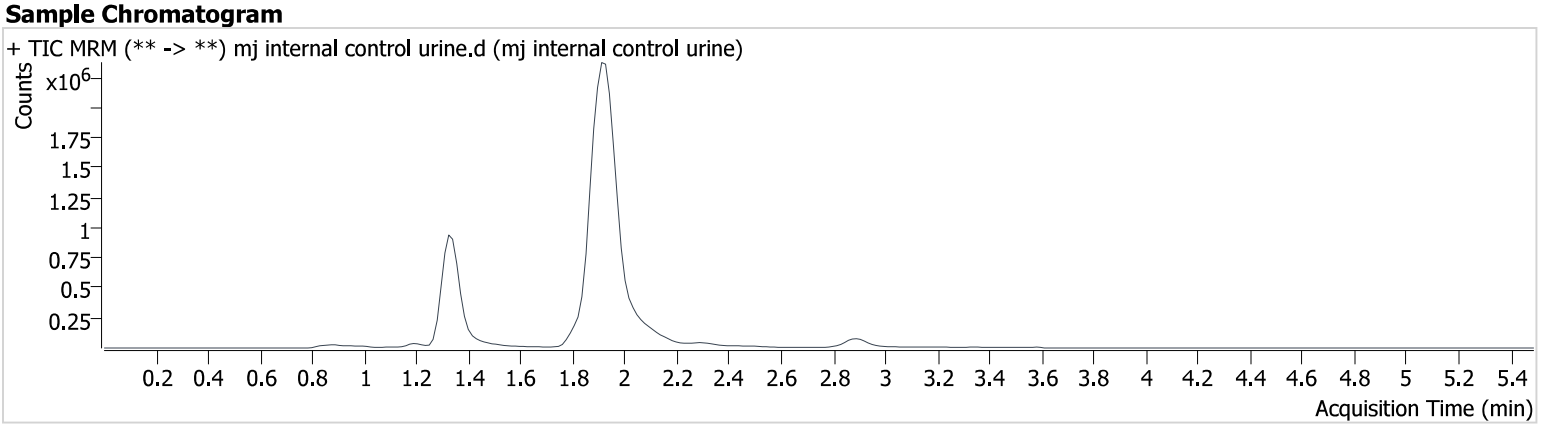


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 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**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 11:23:51 PM		
Sample Info.			

Sample Chromatogram



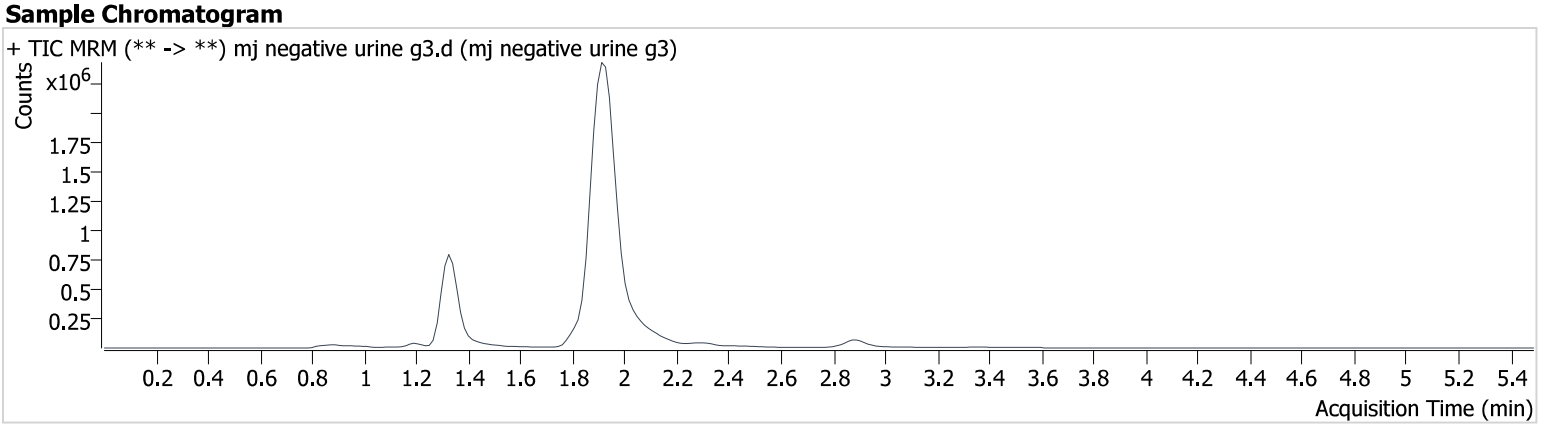
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.334	49945	∞	883.73	∞	2718091	4.356 ng/ml
THC	2.896	57376	∞	22.40	∞	421630	4.744 ng/ml

AM #27 Cannabinoids

Batch results	D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update	6/7/2023 11:47:04 AM

Instrument	69679	**Data File**	mj negative urine g3.d
Type	Sample	**Sample**	mj negative urine g3
Acq. Method	AM 27 THC quant.m	**Operator**	Anne Nord
Sample Position	P3-G3	**Comment**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 10:37:41 PM		
Sample Info.			

Sample Chromatogram

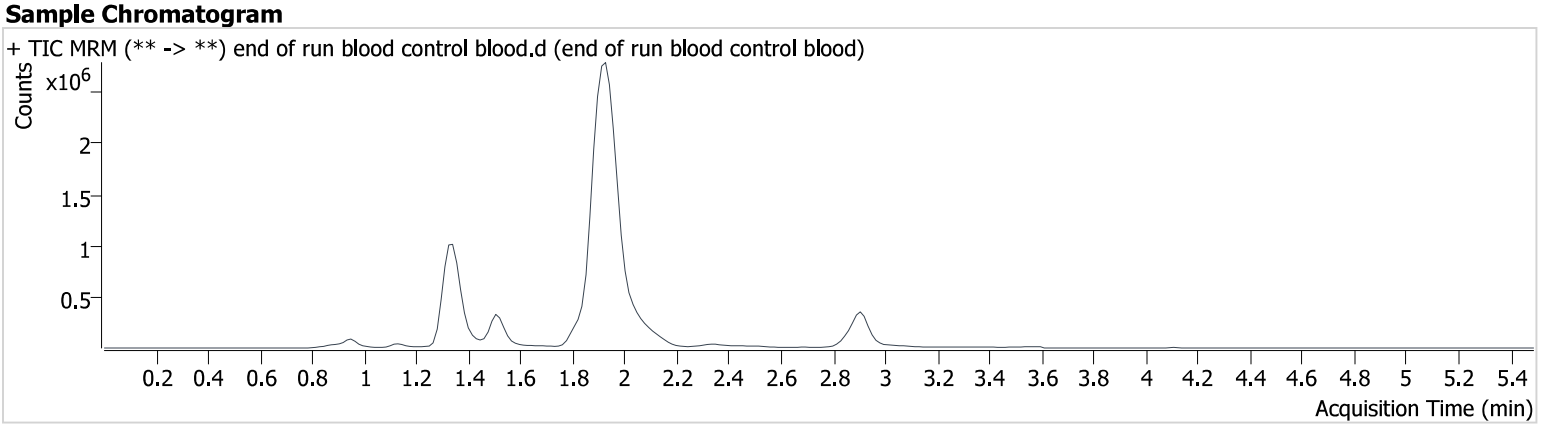


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 AM

Instrument	69679	**Data File**	end of run blood control blood.d
Type	Sample	**Sample**	end of run blood control blood
Acq. Method	AM 27 THC quant.m	**Operator**	Anne Nord
Sample Position	P3-H1	**Comment**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 11:30:27 PM		
Sample Info.			

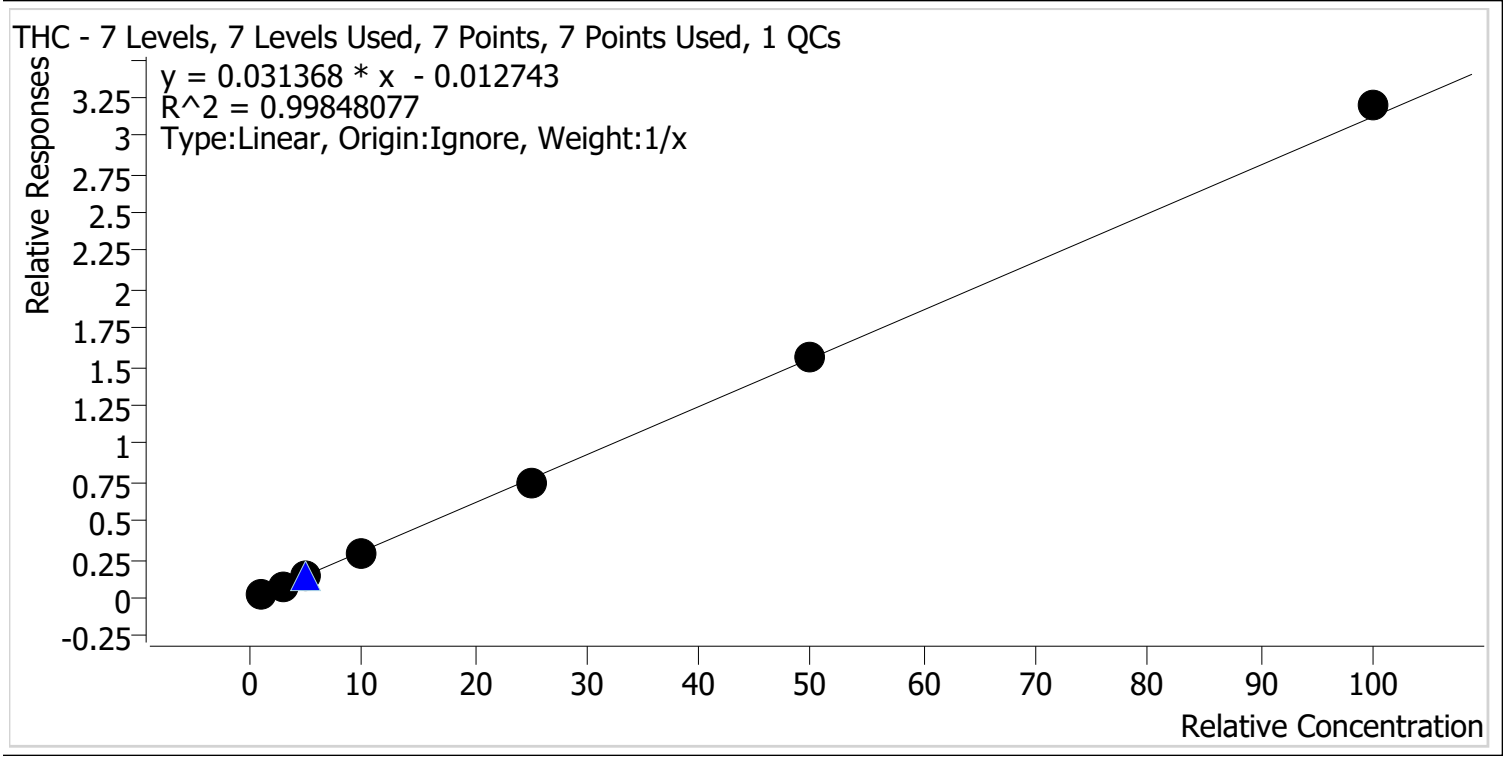
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.334	48314	1716.1	867.45	∞	2743659	4.191 ng/ml
THC	2.911	231247	∞	21.52	354.2	1616622	4.966 ng/ml

Compound Calibration Report

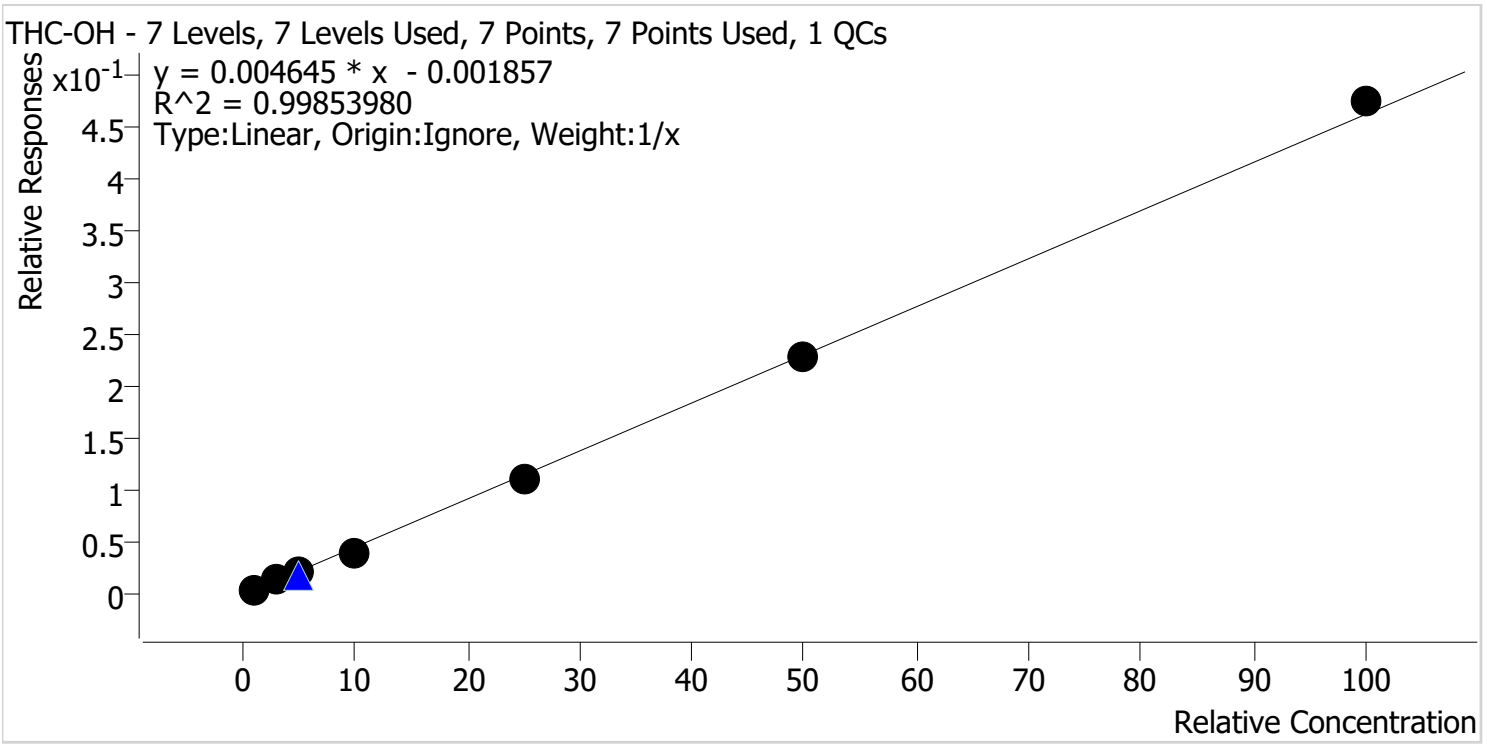
Batch results	D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Last Cal. Update	6/7/2023 11:47 AM
Analyst Name	ISP\datastor
Analyte	THC
Internal Standard	THC-d3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✔	1.0	1.2	121.4
mj cal 2	2	✔	3.0	2.9	96.8
mj cal 3	3	✔	5.0	4.6	92.7
mj cal 4	4	✔	10.0	9.1	91.5
mj cal 5	5	✔	25.0	23.9	95.5
mj cal 6	6	✔	50.0	49.9	99.8
mj cal 7	7	✔	100.0	102.3	102.3

Compound Calibration Report

Batch results	D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Last Cal. Update	6/7/2023 11:47 AM
Analyst Name	ISP\datastor
Analyte	THC-OH
Internal Standard	THC-OH-d3



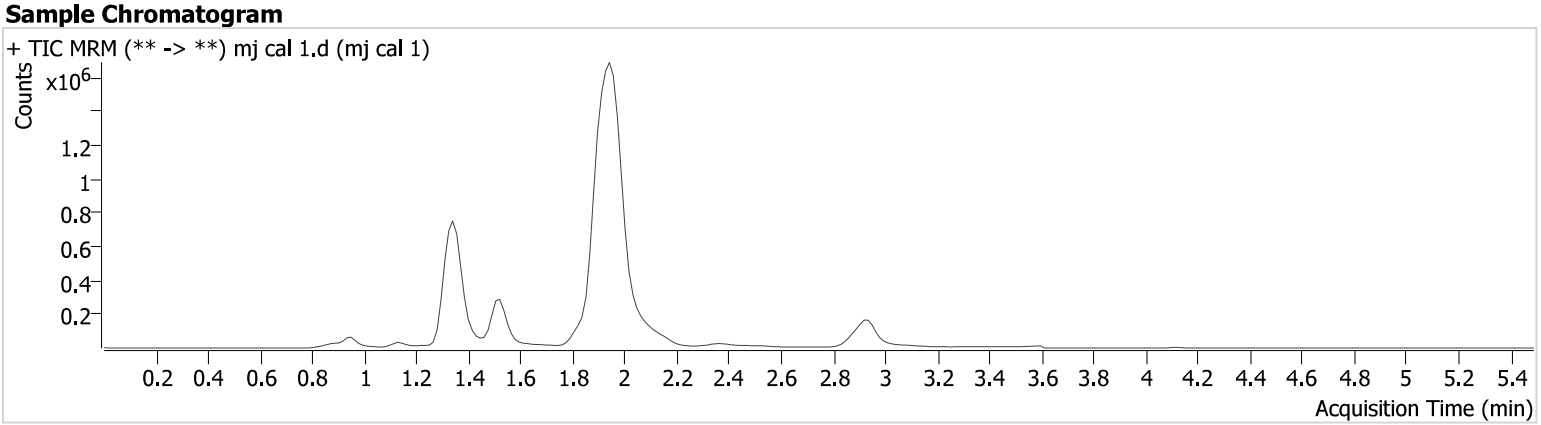
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✔	1.0	1.1	114.7
mj cal 2	2	✔	3.0	3.0	101.1
mj cal 3	3	✔	5.0	4.8	96.6
mj cal 4	4	✔	10.0	9.1	90.8
mj cal 5	5	✔	25.0	23.8	95.3
mj cal 6	6	✔	50.0	49.5	99.0
mj cal 7	7	✔	100.0	102.6	102.6

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 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**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 7:00:04 PM		
Sample Info.			

Sample Chromatogram



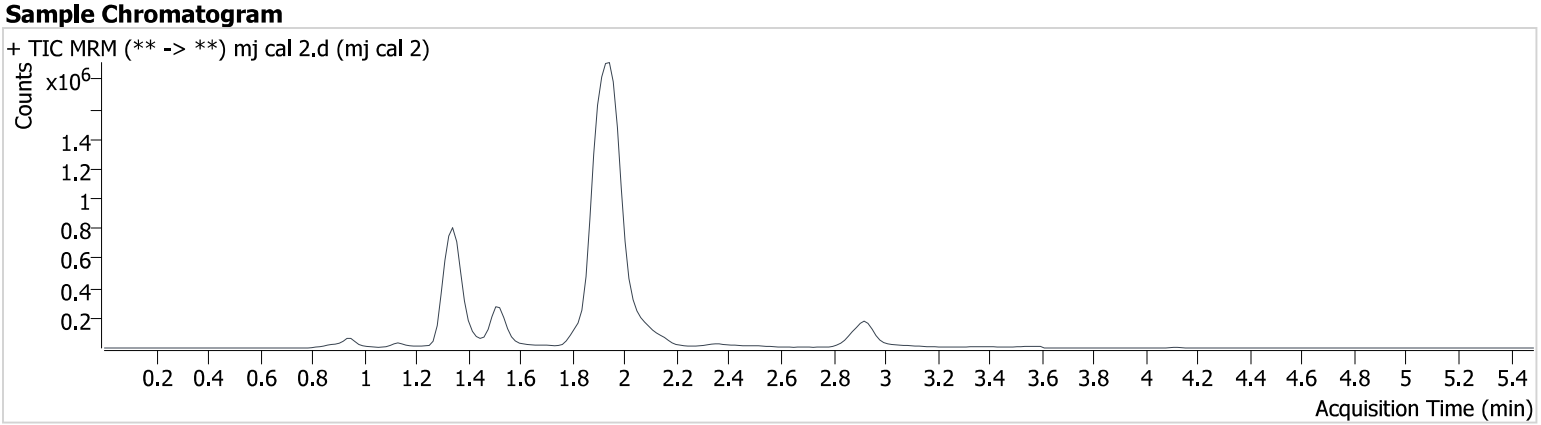
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.349	8567	∞	851.17	∞	2466954	1.147 ng/ml	Low
THC	2.941	24796	413.2	20.60	41.5	978843	1.214 ng/ml	

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 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**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 7:06:48 PM		
Sample Info.			

Sample Chromatogram



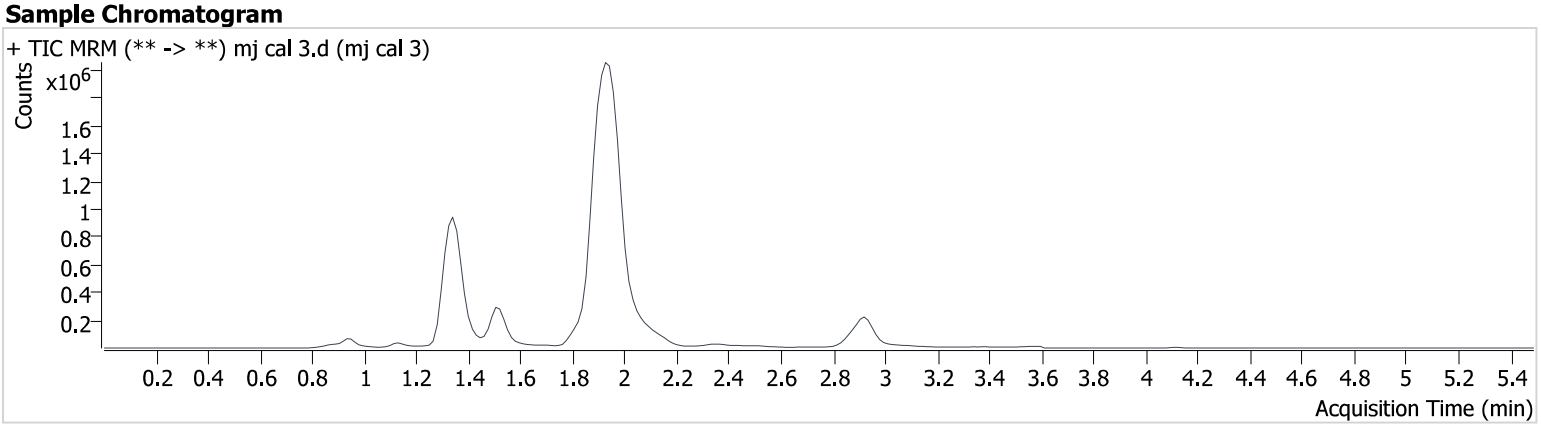
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.334	29928	∞	877.15	∞	2448113	3.032 ng/ml
THC	2.926	78953	2355.1	24.90	∞	1008028	2.903 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 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**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 7:13:22 PM		
Sample Info.			

Sample Chromatogram



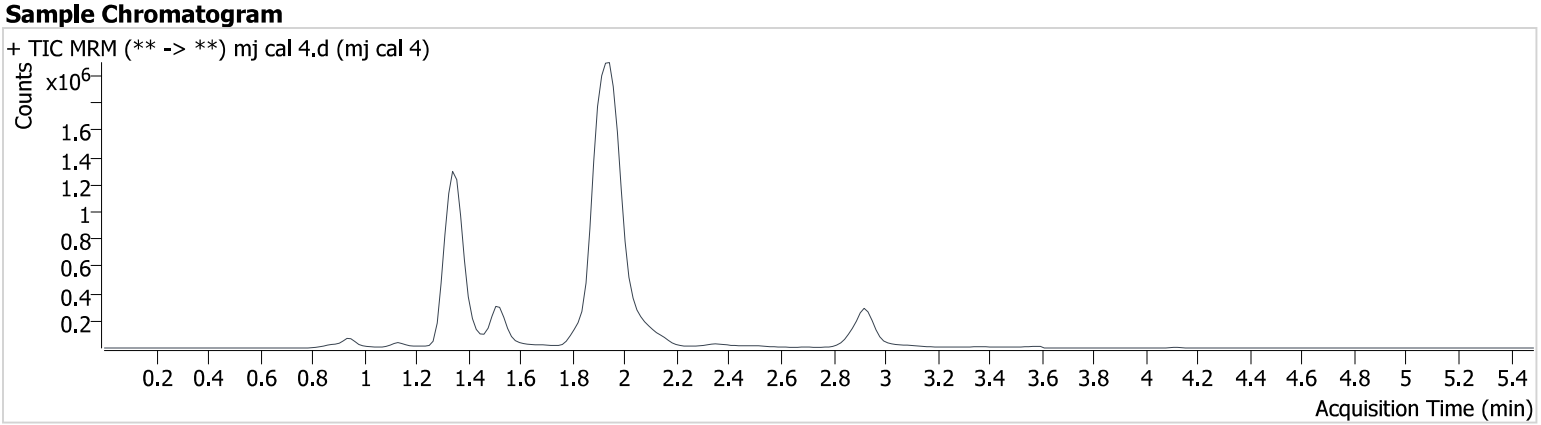
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.334	52456	789.1	870.03	∞	2550160	4.828 ng/ml
THC	2.926	152585	1078120757 86735.0	23.10	∞	1150200	4.635 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 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**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 7:19:56 PM		
Sample Info.			

Sample Chromatogram



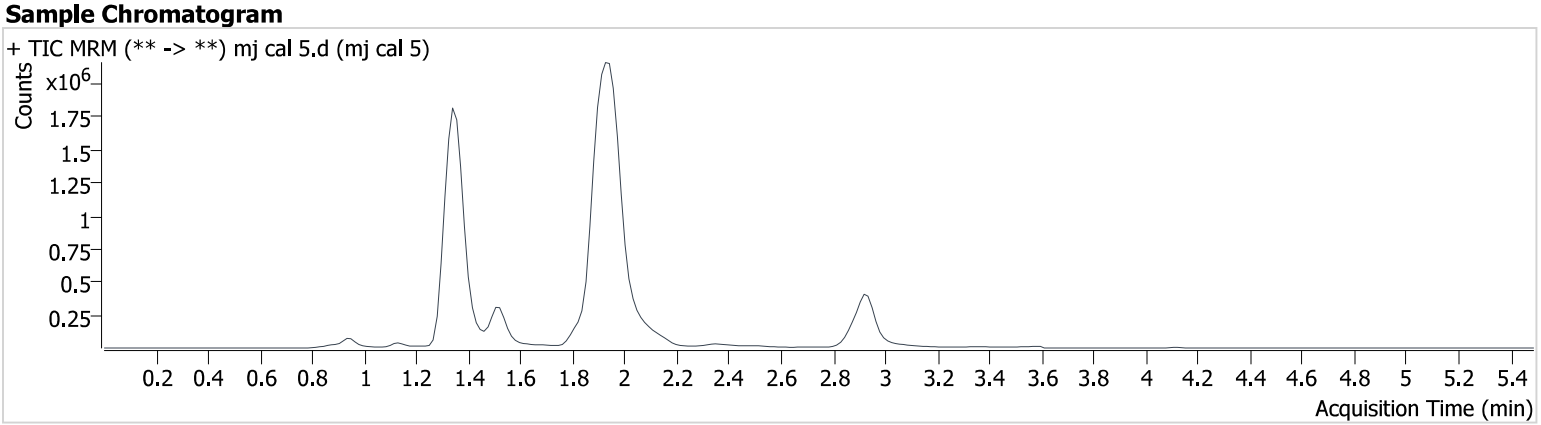
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.334	107537	∞	920.51	∞	2667482	9.079 ng/ml
THC	2.926	345724	∞	23.95	1542.1	1260781	9.148 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 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**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 7:26:31 PM		
Sample Info.			

Sample Chromatogram



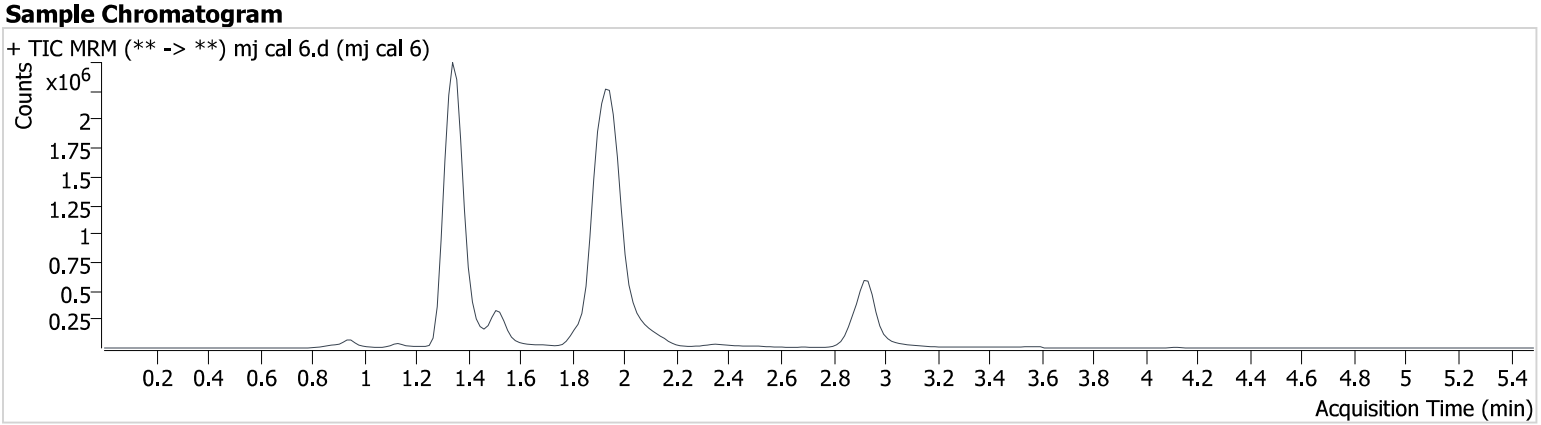
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.334	287139	∞	891.51	∞	2638660	23.827 ng/ml
THC	2.926	928001	∞	23.99	∞	1260216	23.882 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 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**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 7:33:05 PM		
Sample Info.			

Sample Chromatogram



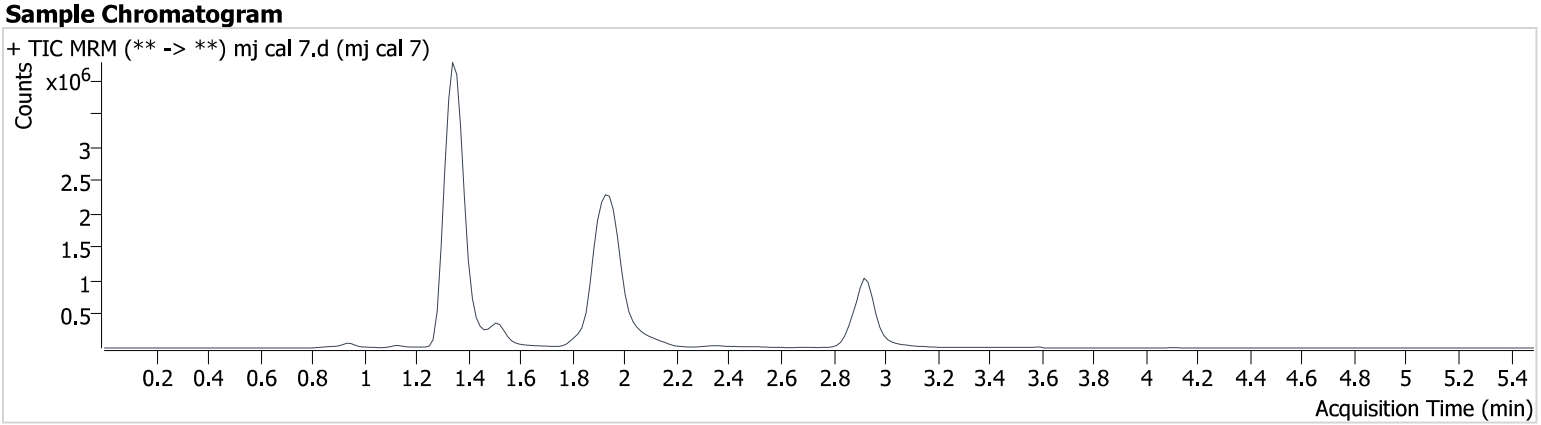
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.334	592768	∞	869.59	∞	2600315	49.476 ng/ml
THC	2.941	1915528	∞	24.01	∞	1233536	49.911 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2023\am 27-28\060623\QuantResults\cann.batch.bin
Calibration Last Update 6/7/2023 11:47:04 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**	
Injection Volume	10		
Acq. Date-Time	6/6/2023 7:39:39 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.334	1158355	10975.8	850.71	∞	2439770	102.612 ng/ml
THC	2.926	3974124	∞	24.18	5996209098667740.0	1243300	102.307 ng/ml