5/26/2022

REVIEWED
By Britany Wylie at 3:02 pm, May 31, 2022

Worklist: 5933

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION	
C2022-0949	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2022-0995	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2022-1038	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2022-1040	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2022-1061	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2022-1075	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2022-1122	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 5/25/22 Analyst: Anne Nord
Plate lot#: 220309 Plate re-test: 9/9/2022
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: 22B52020 **Urine Blank:** 21522 **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 blood (calibrated pipette) 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
☒ 4. Case sample response for THC 1ng/ml, OH-THC 3ng/mL (quantitative blood), Carboxy-THC: 5 ng/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? (if not is it describe in comments section)
☒ 6 Enter QCs into control charting.
☒ 7 Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS: *THC-OH curve range 3-100*
Entered wrong case number for P3-A3, labeled as C2022-0949-1 it is actually C2022-0995-1
Entered wrong case number for one of the samples run at P3-B3 labeled as C2022-0995-1 it is actually C2022-1038-1
P3-A2 labeled as internal blood control it is actually internal urine control.

Idaho State Police
Forensic Services

Request for Departure from an Analytical Method or Quality Standard

Deviation Number (assigned by QM): **TOX-22-02**

Date of Request:
03/02/2022

Requestor/Discipline:
Celena Shrum/Toxicology

Analytical Method/Quality Standard, Revision #:
Toxicology AM #25, AM #26, and AM #27, Revision 13

Temporary or Permanent Deviation:
Permanent

Scope of Deviation (record specific information, e.g. affected programs, evidence types, expected end date; etc):

Deviation will remain in place until the change is made in the next method revision.

Deviation Request (Describe detailed instructions of the changes being made; include reference to specific section number(s) in the method manual):

Toxicology AM #25 3.3.1.1 Internal standards are prepared by the ToxBox plate manufacturer and contained on the 96 well plate. If the run contains urine samples, a positive external urine
control must also be run.

Toxicology AM #26 3.3.2 A negative control will be run with each extraction. If the run contains urine samples, a negative urine control and external positive urine control must also be included.

Toxicology AM #27 3.3.2 A negative control will be run with each extraction. If the run contains urine samples, a negative urine control and positive external urine control will also be included in the run.

The deviation is to include the option of using an internal urine control in lieu of an external urine control.

Technical Justification for Analytical Method Deviations:

Internal controls serve the same purpose as external controls but also helps to avoid the possible issues that can occur with using external controls (incorrect spiking, incorrect preparation, evaporation of compounds, etc.). If these errors occur, runs need to be repeated and this wastes time, sample, and supplies.

Technical Review

☒ Departure approved
Comments:

☐ Departure Not Approved
Comments:

Approver: Rachel Cutler Date: 3/2/22
Title: Lab Manager

Quality Review

Quality Approver: Jason Crowe
Title: Quality Manager
Date: 3/2/2022

	1	2	3	4	5	6
a	cal 1	Internal urine	0995-1			
b	cal 2	negative blood	1038-1			
c	cal 3	1040-1				
d	cal 4	1061-1				
e	Cal 5	1075-1				
f	cal 6	1122-1				
g	cal 7	negative urine				
h	Internal control (blood)	0949-1				

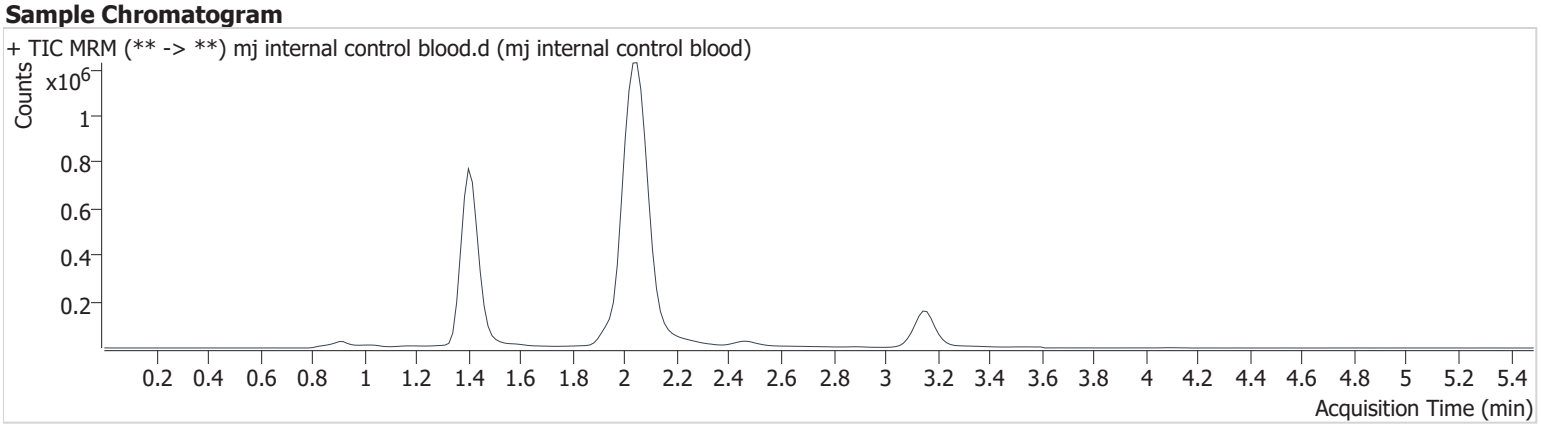
c2022-____-_

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

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	5/25/2022 12:40:47 PM		
Sample Info.			

Sample Chromatogram



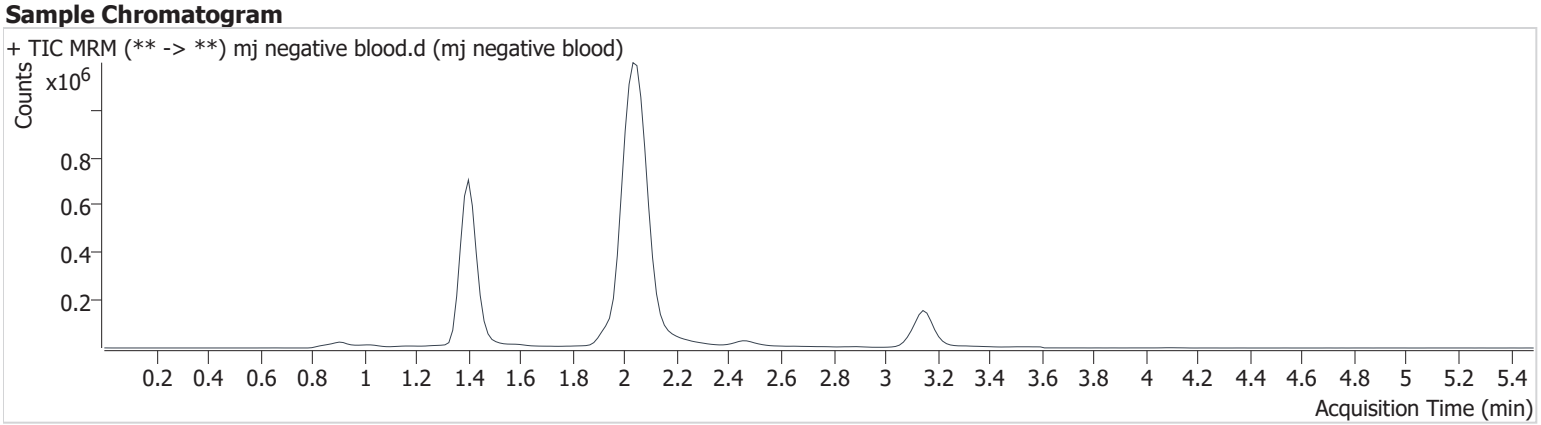
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.406	256428	∞	12.7	∞	2273851	4.797 ng/ml
THC-COOH	1.433	44084	∞	272.7	∞	645920	14.123 ng/ml
THC	3.167	86912	∞	23.0	∞	805074	4.544 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

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	5/25/2022 12:47:32 PM		
Sample Info.			

Sample Chromatogram

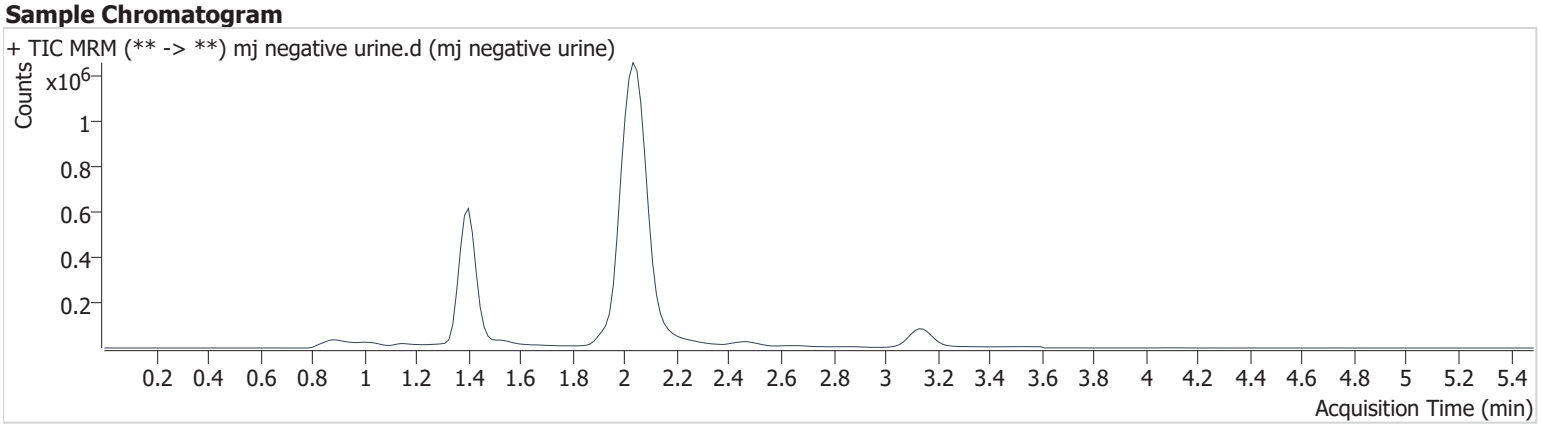


AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

Instrument	69679	**Data File**	mj negative urine.d
Type	Sample	**Sample**	mj negative urine
Acq. Method	AM 27 THC quant.m	**Operator**	Anne Nord
Sample Position	P3-G2	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/25/2022 2:01:03 PM		
Sample Info.			

Sample Chromatogram



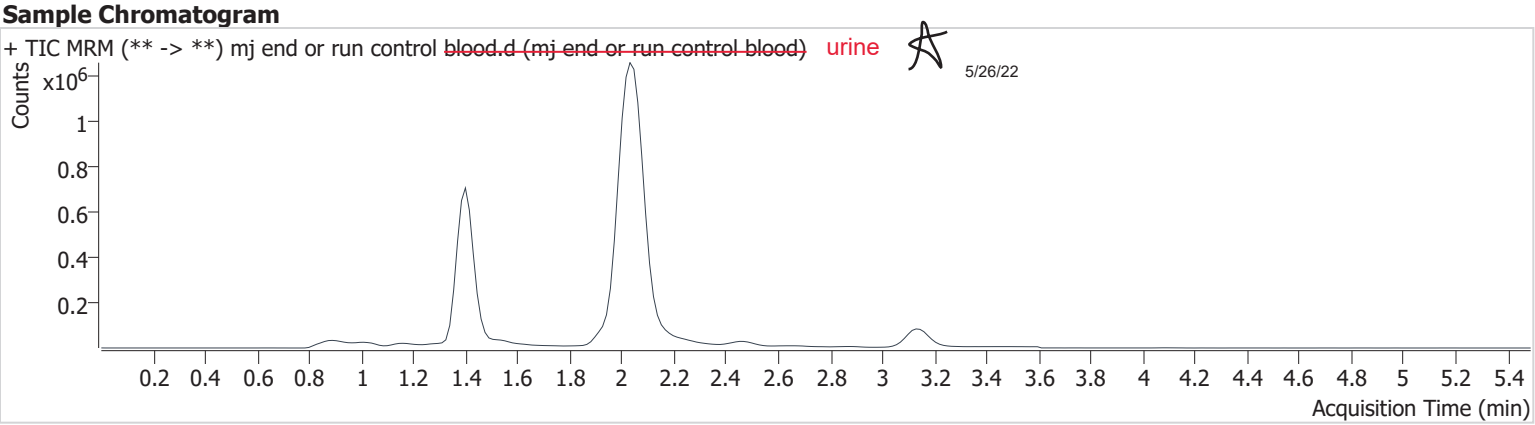
AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

Instrument 69679
Type Sample
Acq. Method AM 27 THC quant.m
Sample Position P3-A2
Injection Volume 10
Acq. Date-Time 5/25/2022 3:07:23 PM
Sample Info.

Data File mj end or run control blood.d
Sample mj end or run control ~~blood~~ urine 5/26/22
Operator Anne Nord
Comment

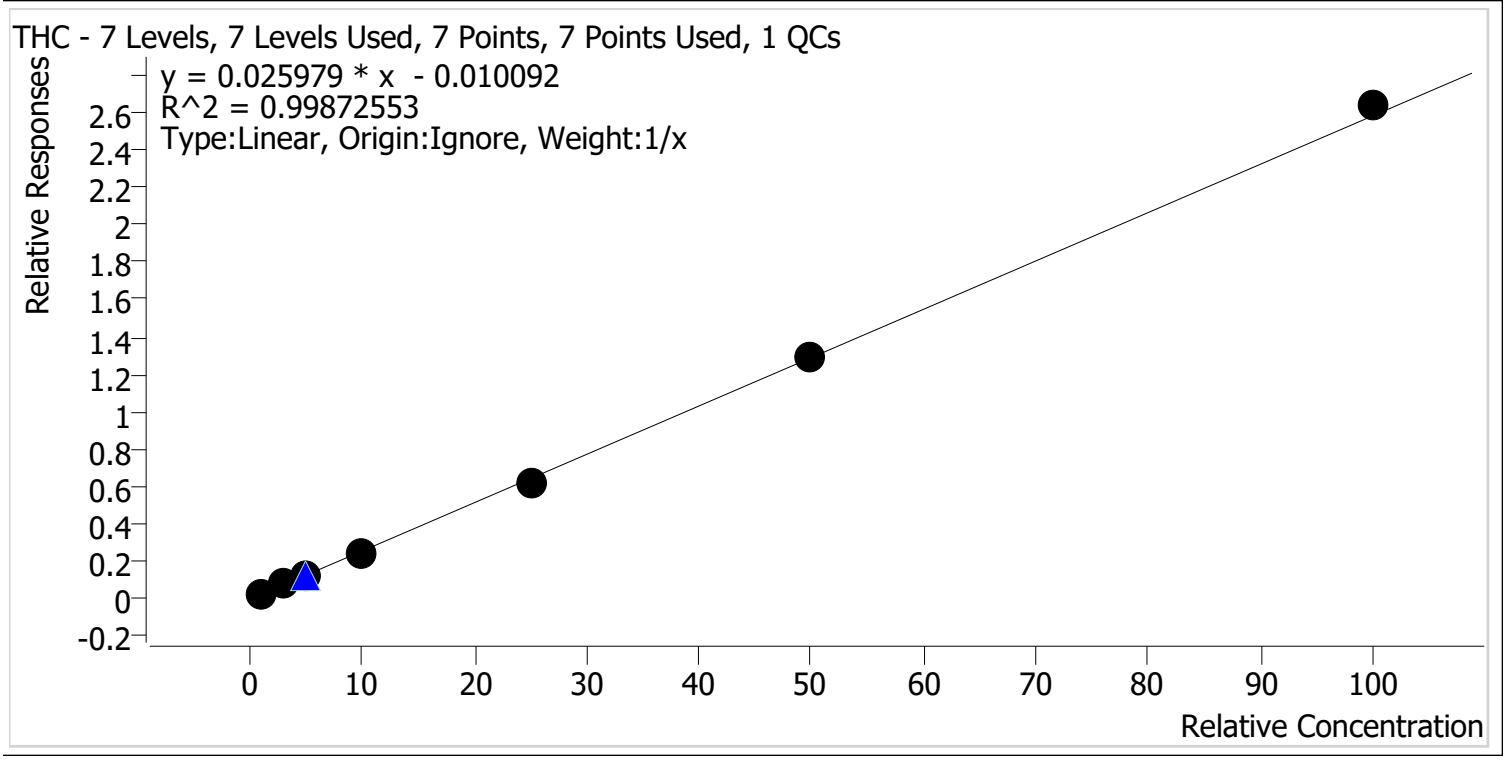
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.406	257603	∞	12.6	∞	2313977	4.745 ng/ml
THC-COOH	1.433	28083	∞	224.7	∞	404774	14.332 ng/ml
THC	3.152	52126	∞	27.1	∞	458019	4.769 ng/ml

Compound Calibration Report

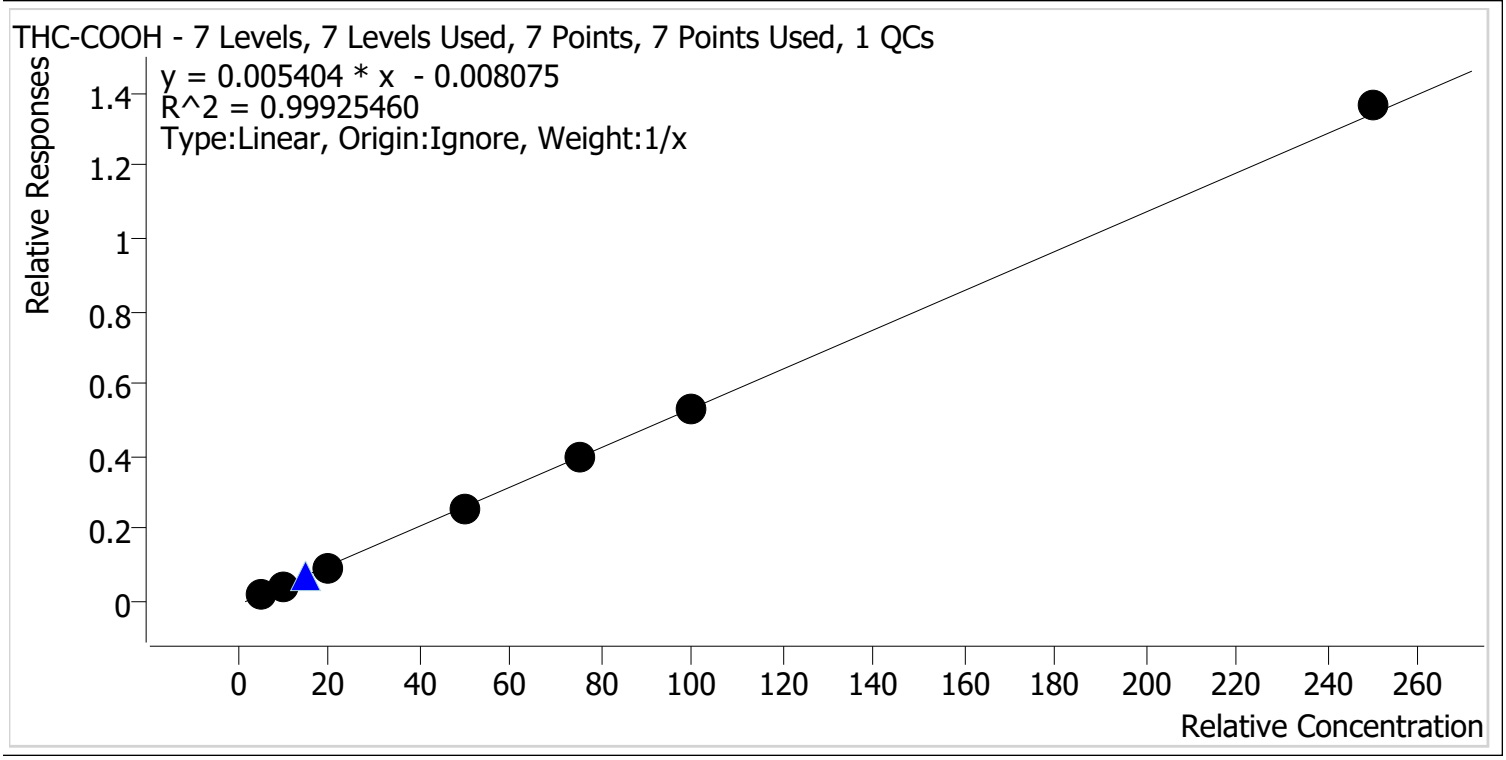
Batch results	D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Last Cal. Update	5/26/2022 3:45 PM
Analyst Name	ISP\datastor
Analyte	THC
Internal Standard	THC-d3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✔	1.0	1.2	117.7
mj cal 2	2	✔	3.0	3.0	99.9
mj cal 3	3	✔	5.0	4.7	93.0
mj cal 4	4	✔	10.0	9.1	91.4
mj cal 5	5	✔	25.0	24.0	96.1
mj cal 6	6	✔	50.0	49.9	99.9
mj cal 7	7	✔	100.0	102.1	102.1

Compound Calibration Report

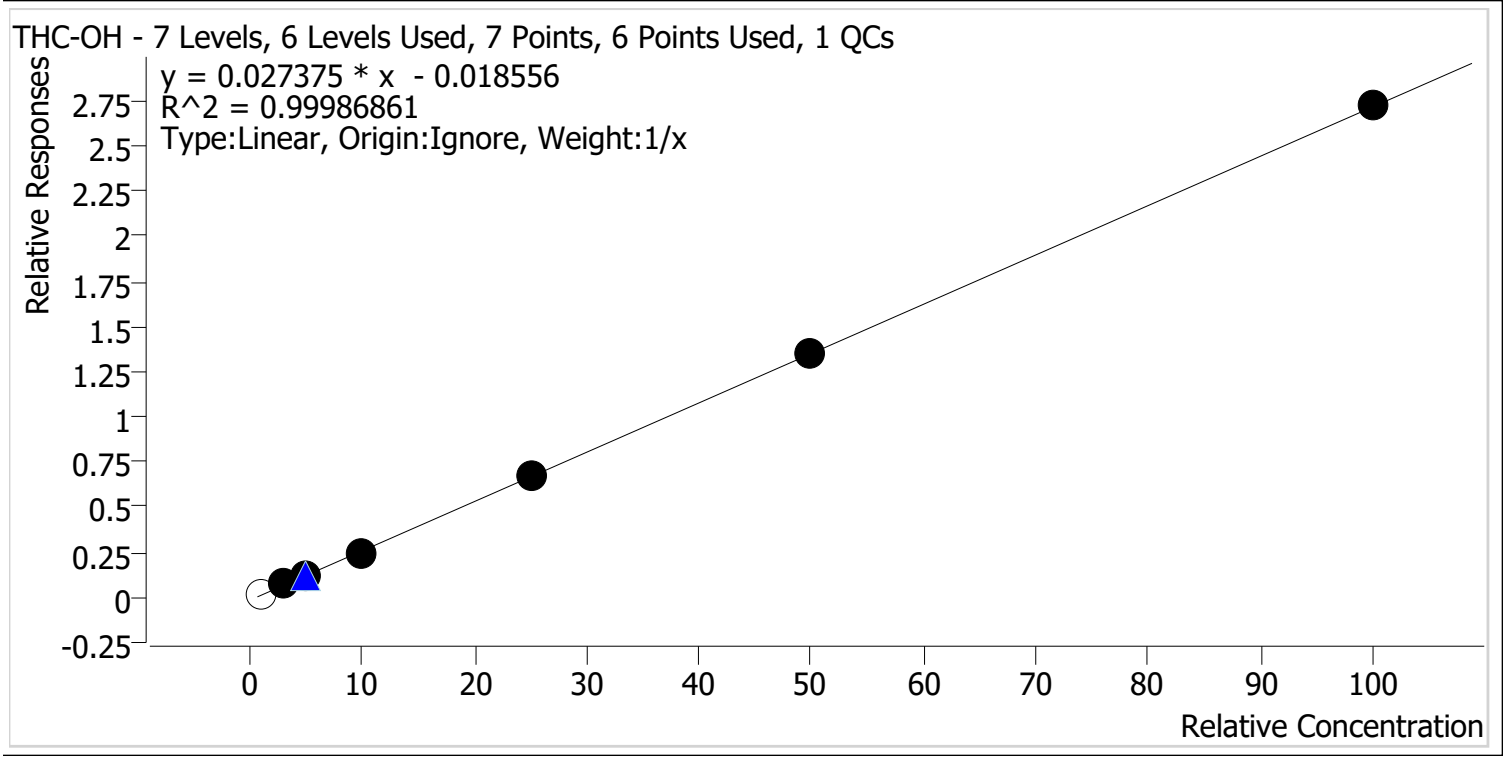
Batch results	D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Last Cal. Update	5/26/2022 3:45 PM
Analyst Name	ISP\datastor
Analyte	THC-COOH
Internal Standard	THC-COOH-d9



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✔	5.0	5.6	111.5
mj cal 2	2	✔	10.0	9.8	97.8
mj cal 3	3	✔	20.0	18.9	94.4
mj cal 4	4	✔	50.0	48.1	96.3
mj cal 5	5	✔	75.0	74.6	99.5
mj cal 6	6	✔	100.0	98.8	98.8
mj cal 7	7	✔	250.0	254.2	101.7

Compound Calibration Report

Batch results	D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Last Cal. Update	5/26/2022 3:45 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	1.3	132.4
mj cal 2	2	✔	3.0	3.1	103.4
mj cal 3	3	✔	5.0	5.0	100.1
mj cal 4	4	✔	10.0	9.6	96.2
mj cal 5	5	✔	25.0	25.0	99.8
mj cal 6	6	✔	50.0	50.1	100.2
mj cal 7	7	✔	100.0	100.2	100.2

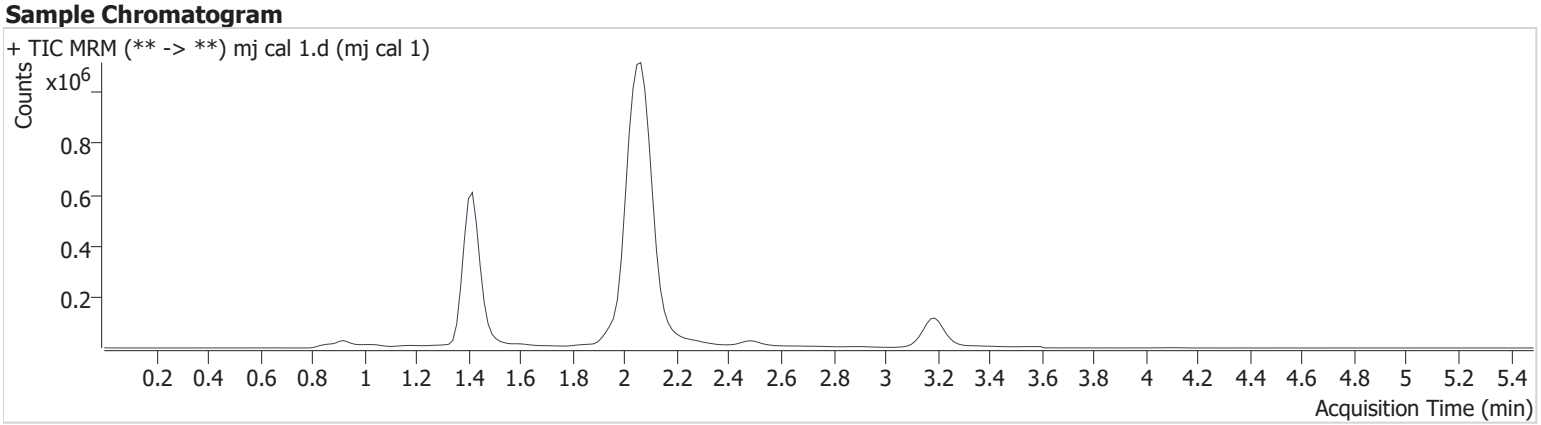
dropped cal 1 ratio out of range

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

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	5/25/2022 11:47:10 AM		
Sample Info.			

Sample Chromatogram



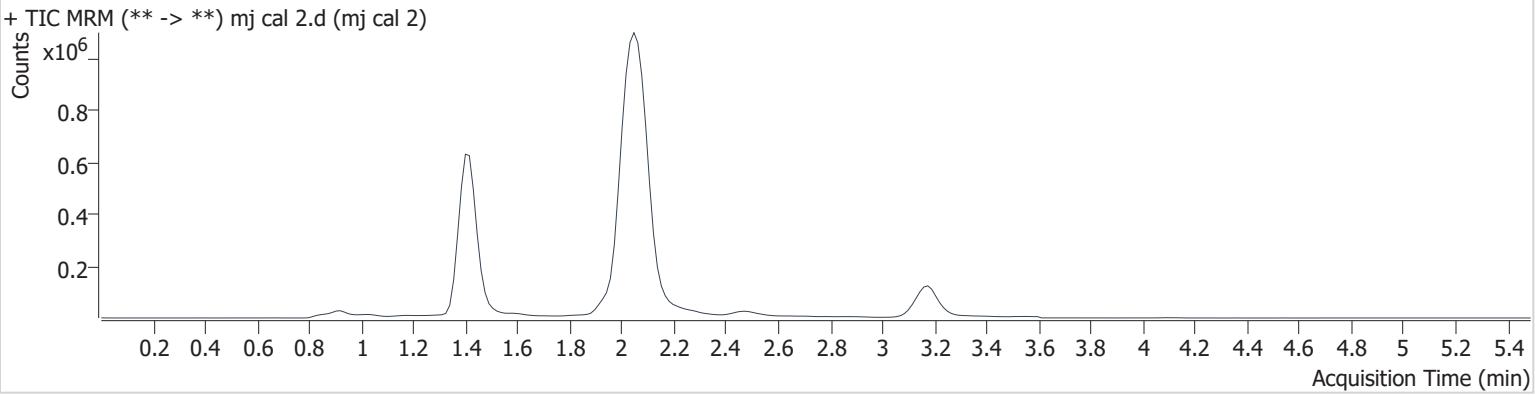
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.421	36963	∞	18.4 **High**	∞	2090689	1.324 ng/ml	**Low**
THC-COOH	1.448	11631	441.9	260.3	56.0	527175	5.576 ng/ml	
THC	3.197	13592	541.0	24.5	31.1	663812	1.177 ng/ml	

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

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	5/25/2022 11:53:54 AM		
Sample Info.			

Sample Chromatogram



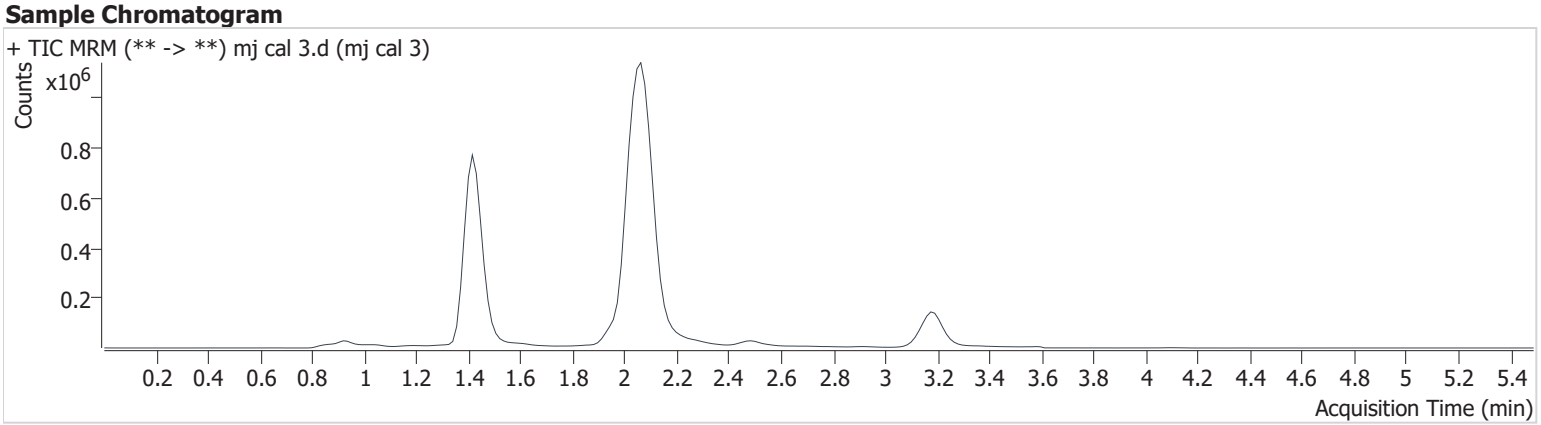
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.406	140798	576.9	13.3	∞	2120443	3.103 ng/ml
THC-COOH	1.433	24688	1057.0	265.5	130.1	551464	9.778 ng/ml
THC	3.182	46503	∞	23.9	621.4	686557	2.996 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

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	5/25/2022 12:00:36 PM		
Sample Info.			

Sample Chromatogram



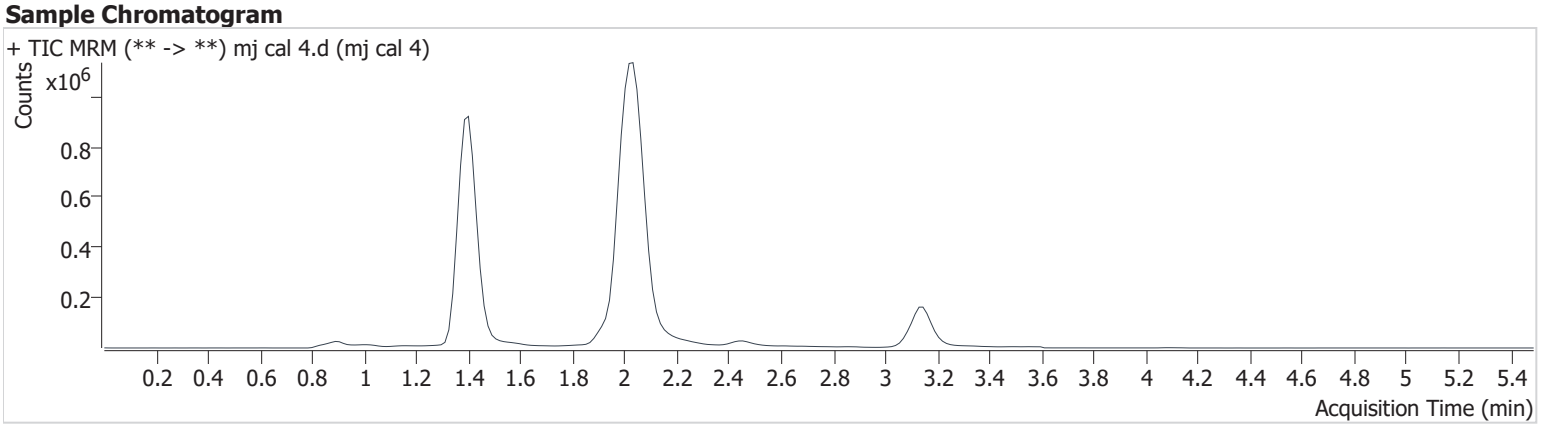
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.421	275144	∞	12.1	∞	2322582	5.005 ng/ml
THC-COOH	1.448	57058	249.0	263.9	∞	607109	18.884 ng/ml
THC	3.197	83862	∞	26.3	170.4	757186	4.652 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

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	5/25/2022 12:07:18 PM		
Sample Info.			

Sample Chromatogram



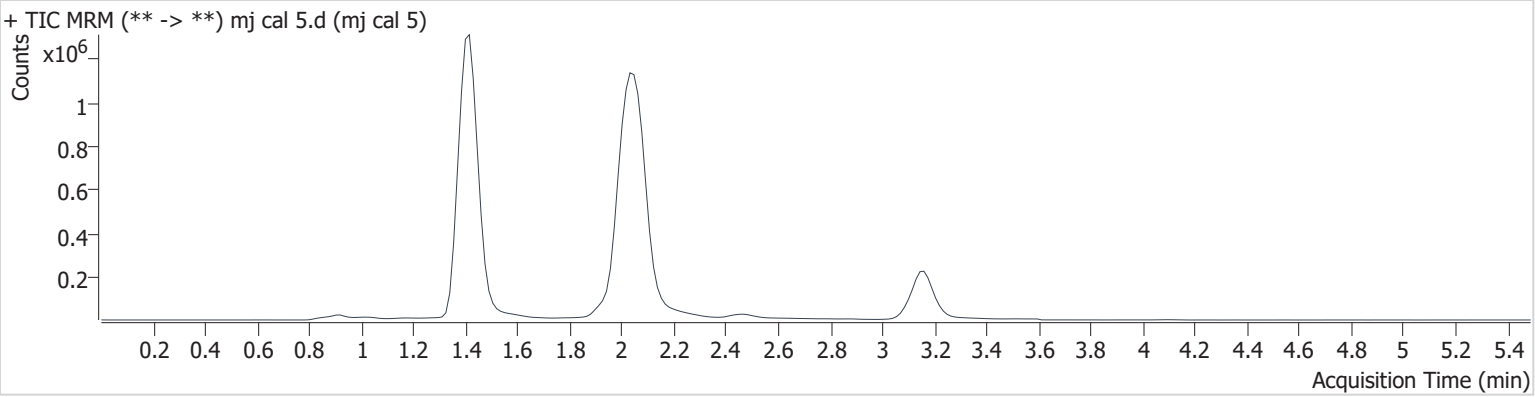
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.391	545473	∞	11.8	∞	2228018	9.621 ng/ml
THC-COOH	1.418	149890	3940.8	267.9	∞	594559	48.142 ng/ml
THC	3.152	168689	∞	24.0	∞	741799	9.142 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

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	5/25/2022 12:14:02 PM		
Sample Info.			

Sample Chromatogram



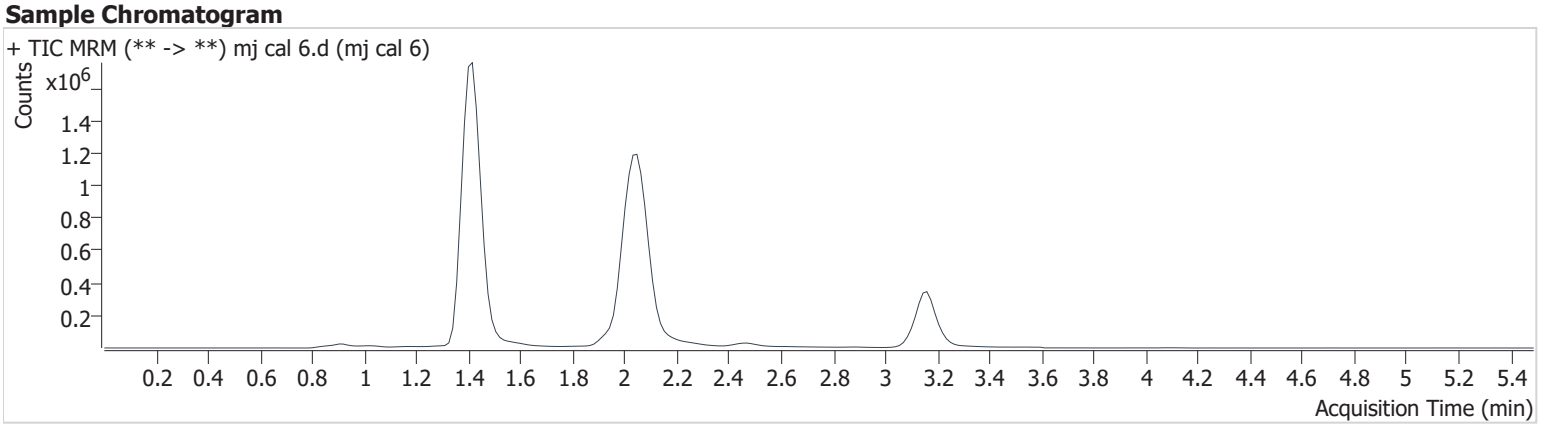
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.406	1613596	∞	12.1	∞	2427504	24.960 ng/ml
THC-COOH	1.433	251516	∞	263.3	∞	636351	74.629 ng/ml
THC	3.167	482879	∞	23.5	∞	786462	24.022 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

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	5/25/2022 12:20:45 PM		
Sample Info.			

Sample Chromatogram



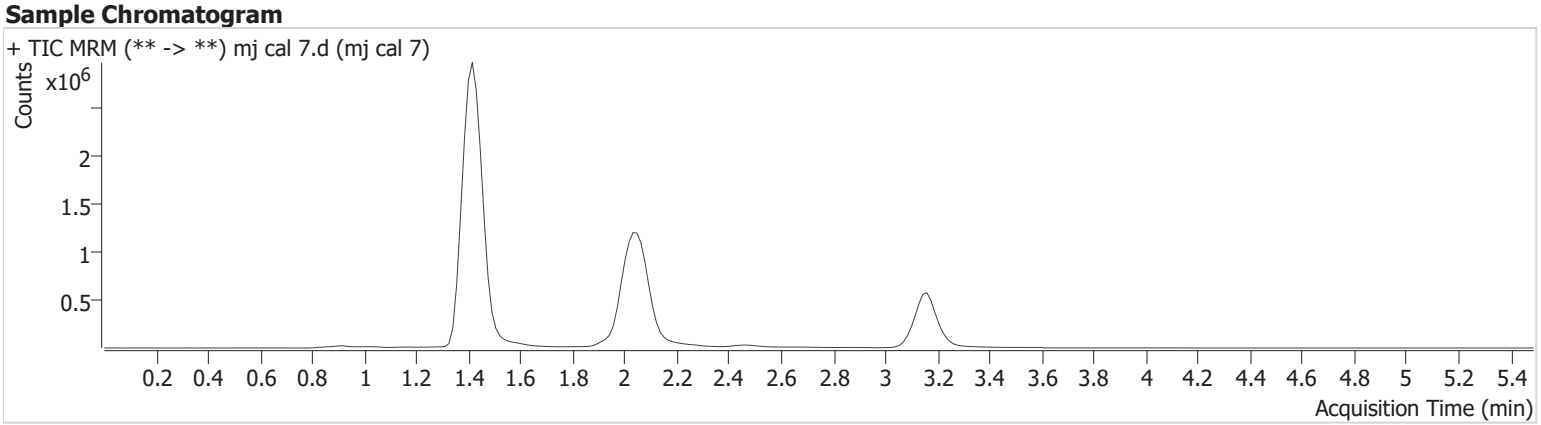
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.406	3119681	∞	12.2	∞	2306739	50.082 ng/ml
THC-COOH	1.433	324025	1525.3	263.1	∞	616079	98.813 ng/ml
THC	3.167	1002065	∞	23.5	∞	778597	49.929 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\052522\QuantResults\cann.batch.bin
Calibration Last Update 5/26/2022 3:45:58 PM

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	5/25/2022 12:27:27 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.406	6202039	∞	12.6	∞	2275834	100.228 ng/ml
THC-COOH	1.433	782484	∞	260.2	∞	572996	254.178 ng/ml
THC	3.167	2174073	∞	24.0	∞	822905	102.083 ng/ml