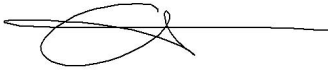





REVIEWED
By Sarah Collins at 2:20 pm, Mar 02, 2022



3/2/2022

Worklist: 5652

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>	
C2022-0283	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
C2022-0284	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
C2022-0346	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 2/25/22
Plate lot#: 211018

Analyst: Anne Nord
Plate re-test: 4/18/22

Mobile phase A: 0.1% Formic Acid in LCMS Water MTBE
Mobile phase B: 0.1% Formic acid in Acetonitrile Hexane
LCMS Methanol

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:

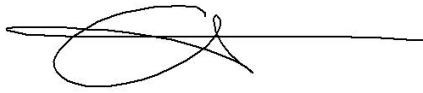
- 1. Remove standards, plate, controls, and samples from cold storage. Allow to reach room temperature.
- 2. Urine hydrolysis: add 1.5 ml urine to blank plate, add 250 ul 1N KOH mix and incubate at 40 degrees for 15 minutes.
Pipette 1000µL blood (calibrated pipette) Pipette ID: k52558g in wells of analytical (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 3-100*

THC results between 1 and 3 will be reported as < 3 ng.



	1	2	3	4	5	6
a	cal 1	Internal control (urine)				
b	cal 2	negative blood				
c	cal 3	284-1				
d	cal 4	346-1				
e	Cal 5	negative urine				
f	cal 6	283-1				
g	cal 7					
h	Internal control (blood)					

A



IDAHO STATE POLICE

MEMORANDUM

DATE: 3/3/2022

TO: Toxicology Discipline/ Jason Crowe

FROM: Celena Shrum- Toxicology Discipline lead

SUBJECT: Use of internal control in lieu of external control

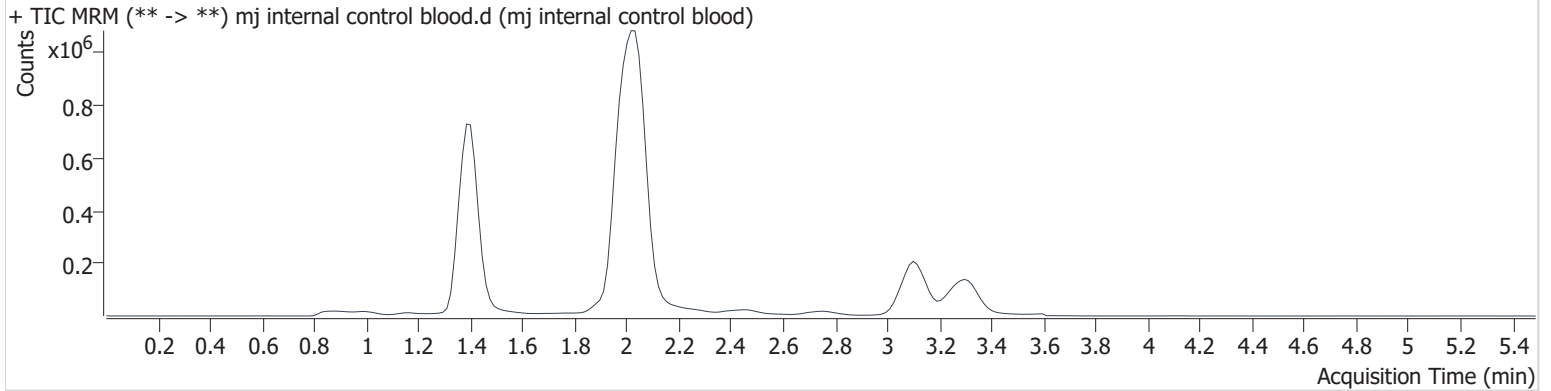
Toxicology Analytical Methods #25, 26, and 27 specify that if a run contains urine samples, a negative control and **external** urine control must also be included in the run. The purpose of this control is to demonstrate that the extraction worked as intended and to ensure that the results and concentrations obtained are accurate. It was decided in October 2021 that extra QC's would be included on the analytical plates so that they could be used as an internal control for runs with urine cases instead of continuing with including an external control. An internal control serves the same purpose as an external control but is prepared and placed on the analytical plate rather than being prepared in-house and placed on the plate at the time of testing. Utilizing internal controls versus external increases the efficacy of the controls used by ensuring consistent spiking and preparation, eliminating evaporation of compounds, etc. There is no quality issue with any of the cases, since an additional urine control was used that served the same purpose as the external control, but it was a violation of the wording specified in the method.

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 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	2/25/2022 1:01:46 PM		
Sample Info.			

Sample Chromatogram



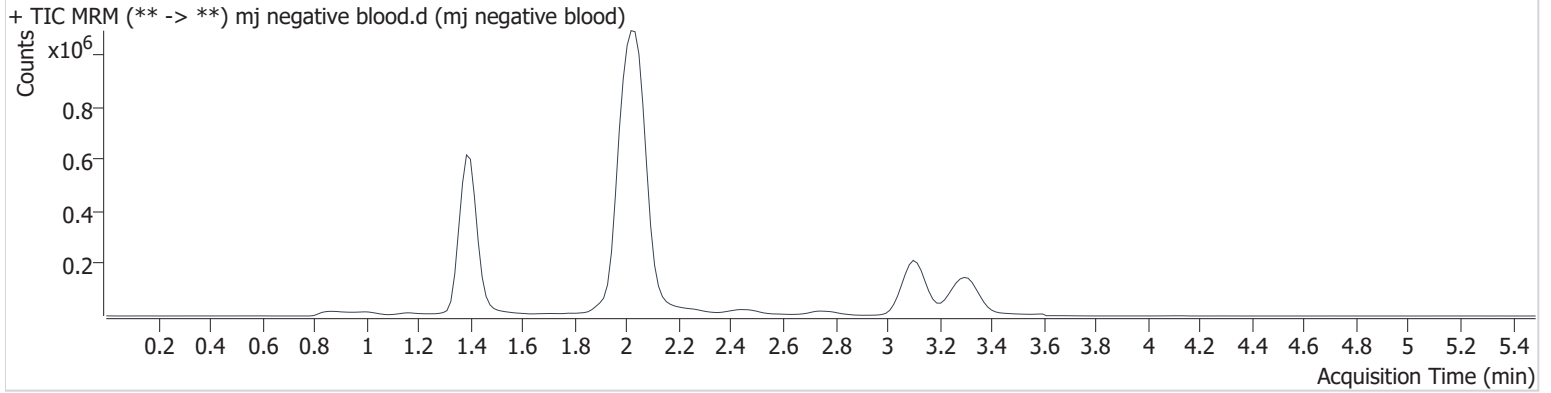
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	35311	1526.8	884.6	445.8	2229617	4.943 ng/ml
THC-COOH	1.416	142154	296.8	35.5	593.6	704904	14.536 ng/ml
THC	3.137	86313	3075.0	25.9	266.7	754057	4.466 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 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	2/25/2022 1:08:31 PM		
Sample Info.			

Sample Chromatogram



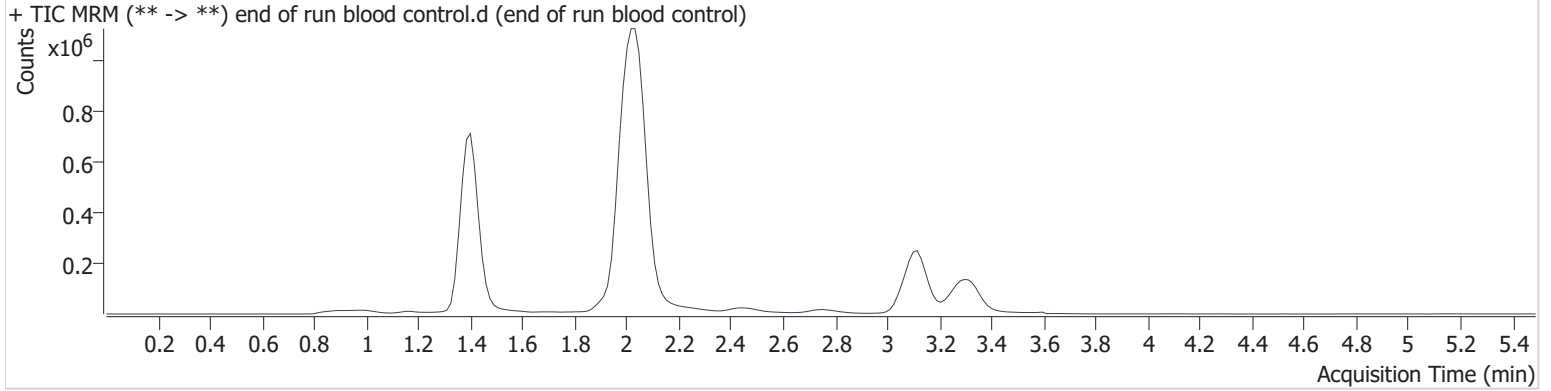
AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 AM

Instrument	69679	Data File	end of run blood control.d
Type	QC	Sample	end of run blood control
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-H1	Comment	
Injection Volume	10		
Acq. Date-Time	2/25/2022 2:28:39 PM		

Sample Info.

Sample Chromatogram



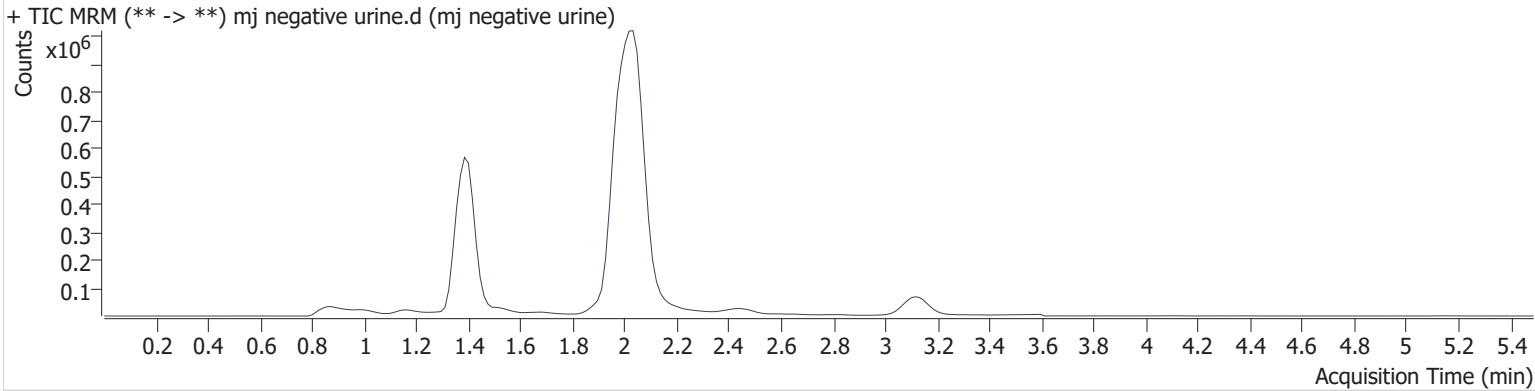
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	34697	349.5	744.5	∞	1993322	5.397 ng/ml
THC-COOH	1.431	115868	297.5	42.4	447.0	717949	11.725 ng/ml
THC	3.137	103652	∞	27.8	519.3	919575	4.404 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 AM

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-E2	Comment	
Injection Volume	10		
Acq. Date-Time	2/25/2022 1:48:34 PM		
Sample Info.			

Sample Chromatogram



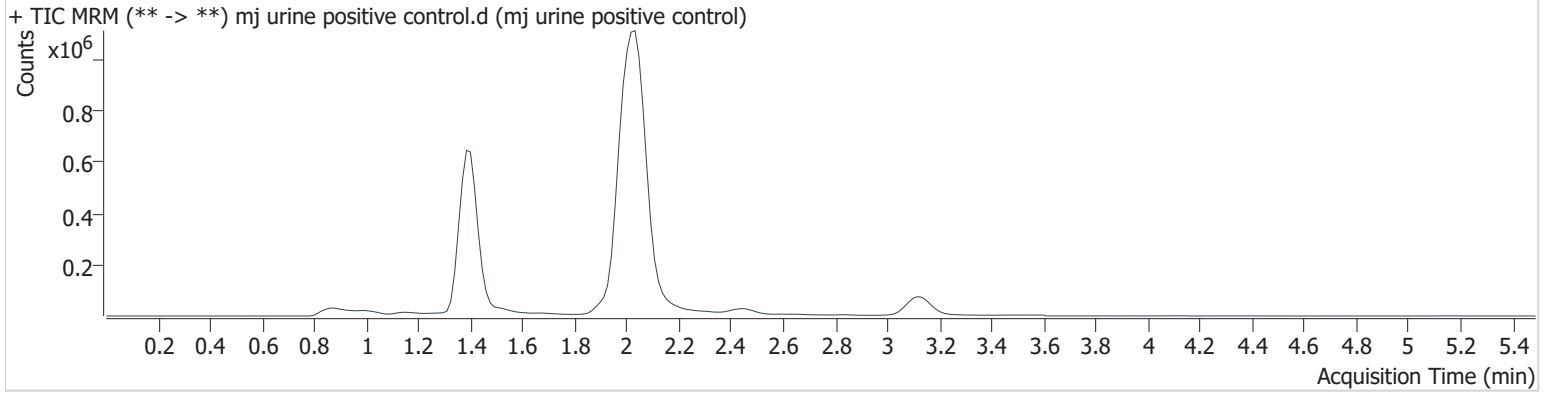
AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 AM

Instrument	69679	Data File	mj urine positive control.d
Type	QC	Sample	mj urine positive control
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-A2	Comment	
Injection Volume	10		
Acq. Date-Time	2/25/2022 2:21:55 PM		

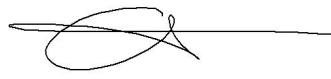
Sample Info.

Sample Chromatogram



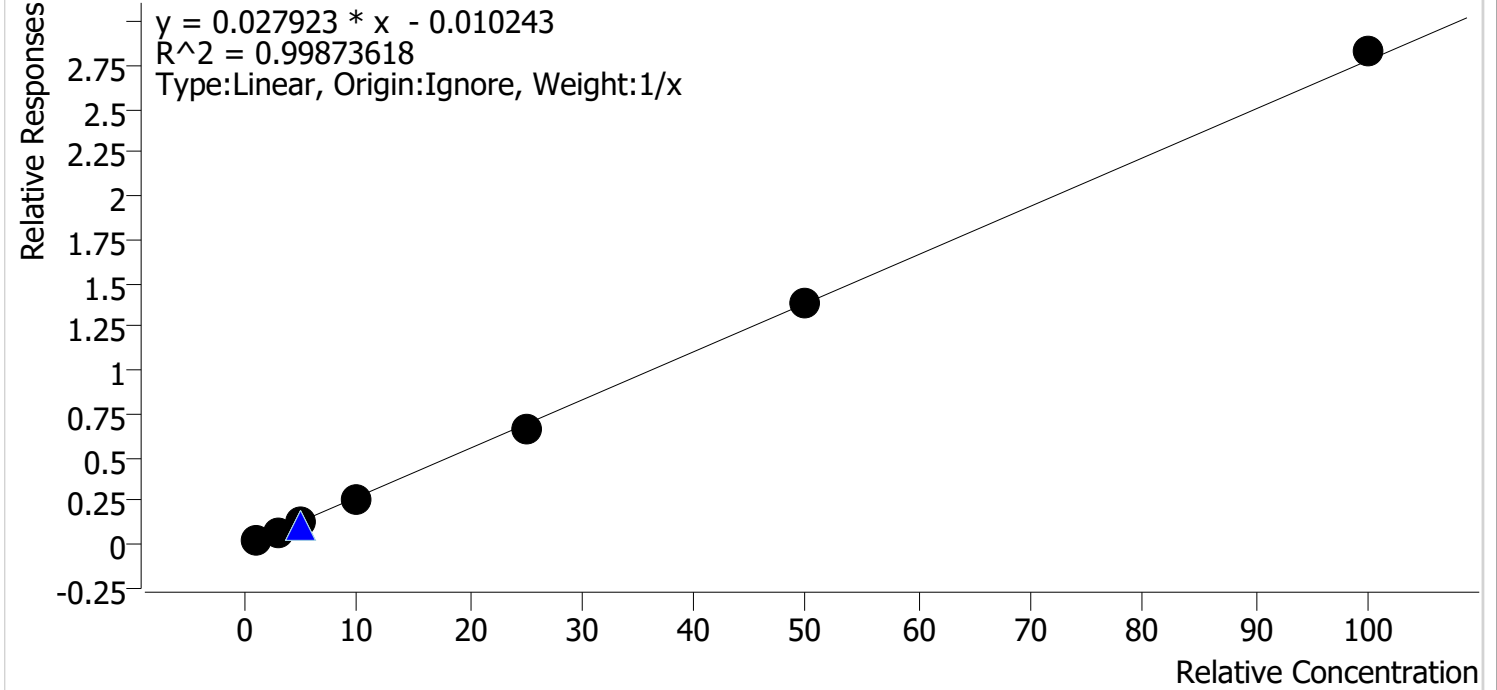
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	36762	435.3	763.3	∞	2089794	5.451 ng/ml
THC-COOH	1.431	78013	369.0	39.6	∞	519649	10.938 ng/ml
THC	3.137	49423	∞	27.2	255.1	425747	4.524 ng/ml

Compound Calibration Report



Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Last Cal. Update 2/28/2022 9:23 AM
Analyst Name ISP\datastor
Analyte THC **Internal Standard** THC-d3

THC - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 3 QCs



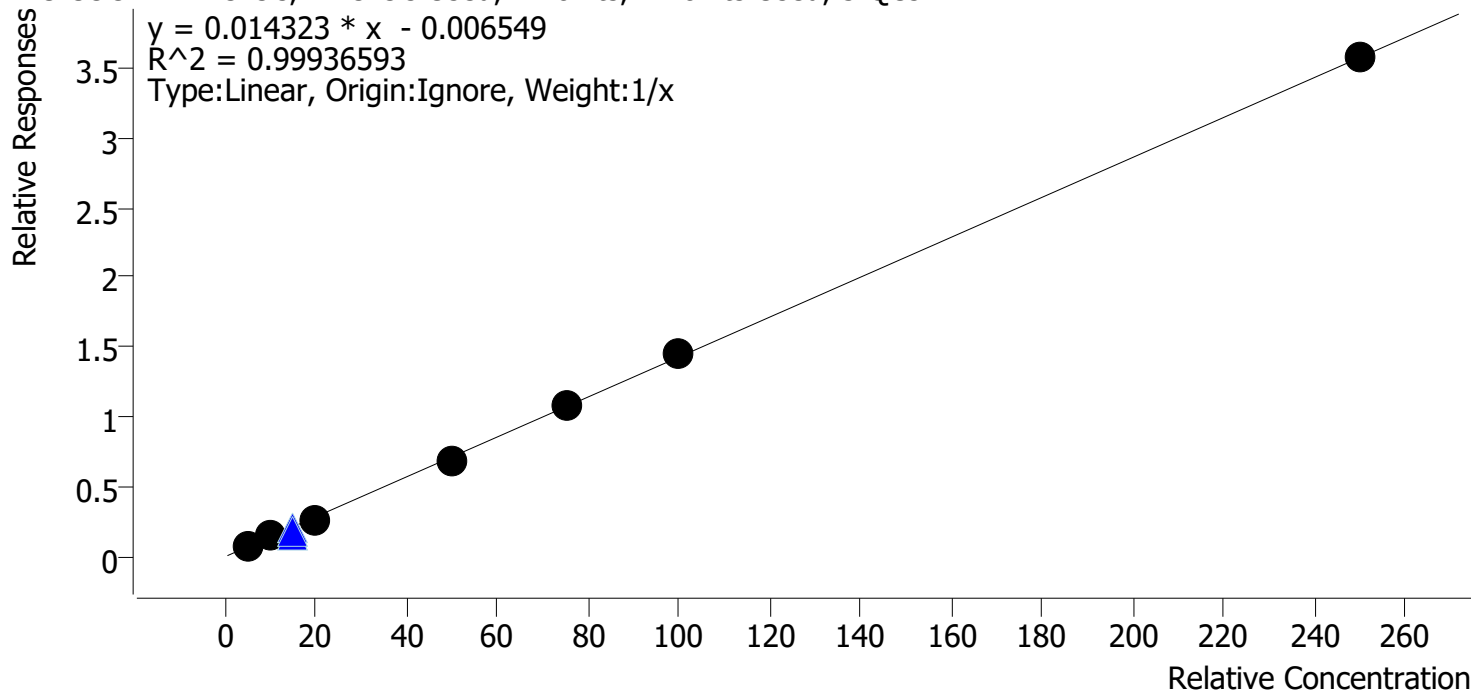
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✓	1.0	1.2	121.5
mj cal2	2	✓	3.0	2.9	95.9
mj cal 3	3	✓	5.0	4.6	92.1
mj cal 4	4	✓	10.0	9.1	91.2
mj cal 5	5	✓	25.0	24.4	97.4
mj cal 6	6	✓	50.0	50.0	100.0
mj cal 7	7	✓	100.0	101.8	101.8

Compound Calibration Report



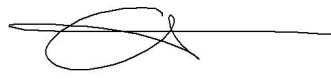
Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Last Cal. Update 2/28/2022 9:23 AM
Analyst Name ISP\datastor
Analyte THC-COOH **Internal Standard** THC-COOH-d9

THC-COOH - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 3 QCs



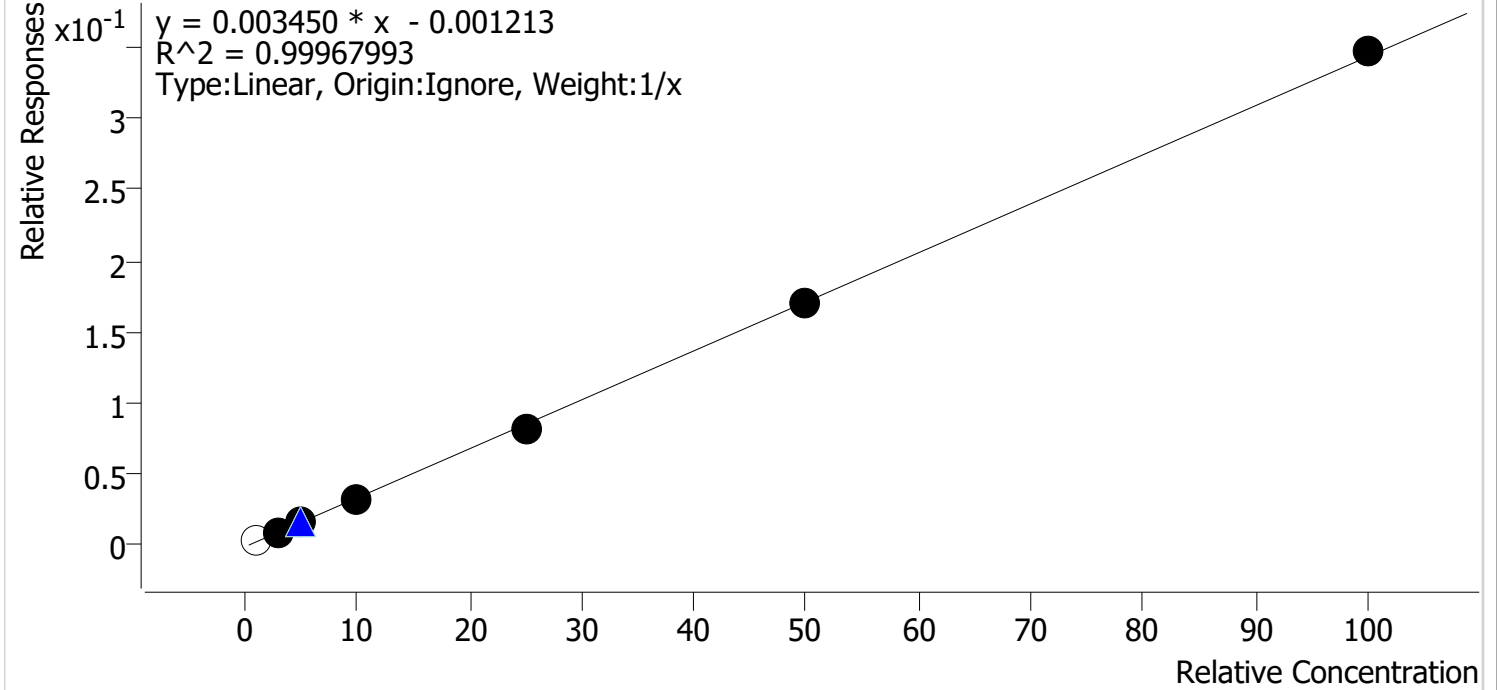
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	✓	5.0	5.4	108.8
mj cal2	2	✓	10.0	9.9	98.7
mj cal 3	3	✓	20.0	18.5	92.5
mj cal 4	4	✓	50.0	48.6	97.2
mj cal 5	5	✓	75.0	75.5	100.6
mj cal 6	6	✓	100.0	102.2	102.2
mj cal 7	7	✓	250.0	249.9	100.0

Compound Calibration Report



Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Last Cal. Update 2/28/2022 9:23 AM
Analyst Name ISP\datastor
Analyte THC-OH **Internal Standard** THC-OH-d3

THC-OH - 7 Levels, 6 Levels Used, 7 Points, 6 Points Used, 3 QCs



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal 1	1	x	1.0	1.5	146.1
mj cal2	2	✓	3.0	3.2	105.7
mj cal 3	3	✓	5.0	4.9	97.8
mj cal 4	4	✓	10.0	9.9	98.8
mj cal 5	5	✓	25.0	24.3	97.1
mj cal 6	6	✓	50.0	49.8	99.5
mj cal 7	7	✓	100.0	101.0	101.0

dropped cal 1 due to ratio being out of range

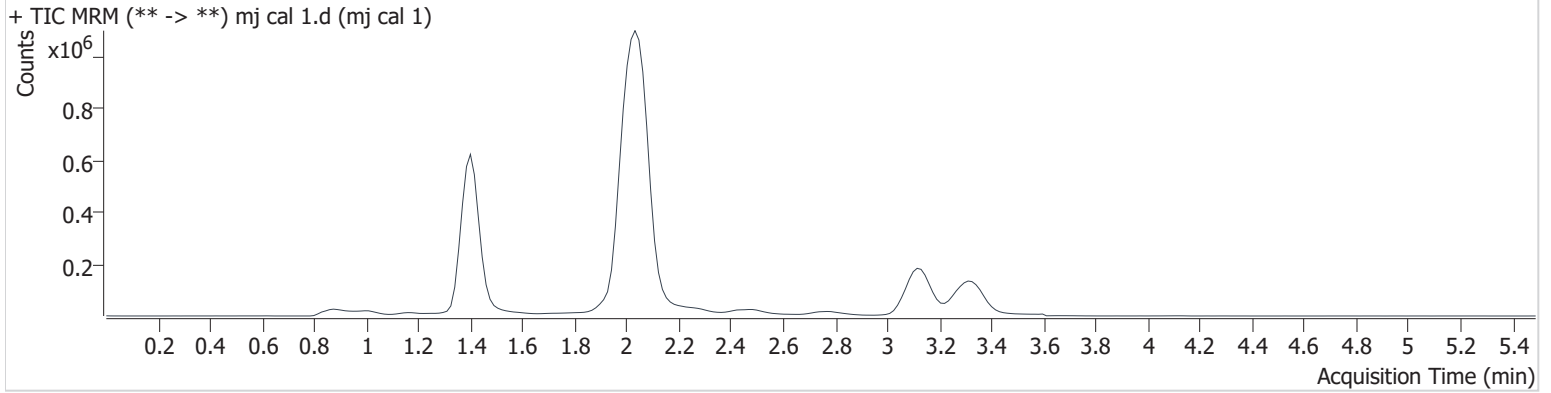
AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 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	2/25/2022 12:08:12 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.409	8163	988.8	508.3 Low	∞	2133117	1.461 ng/ml Low
THC-COOH	1.431	45204	78.9	29.7	28.8	633364	5.440 ng/ml
THC	3.137	16637	∞	28.0	41.3	702579	1.215 ng/ml

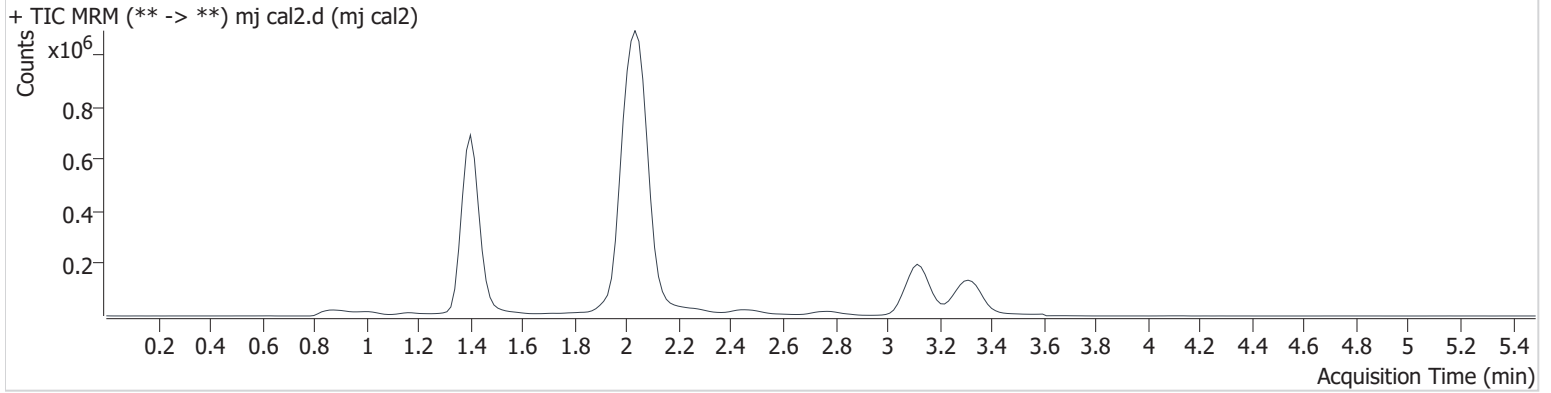
AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 AM

Instrument	69679	Data File	mj cal2.d
Type	Cal	Sample	mj cal2
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-B1	Comment	
Injection Volume	10		
Acq. Date-Time	2/25/2022 12:14:57 PM		

Sample Info.

Sample Chromatogram



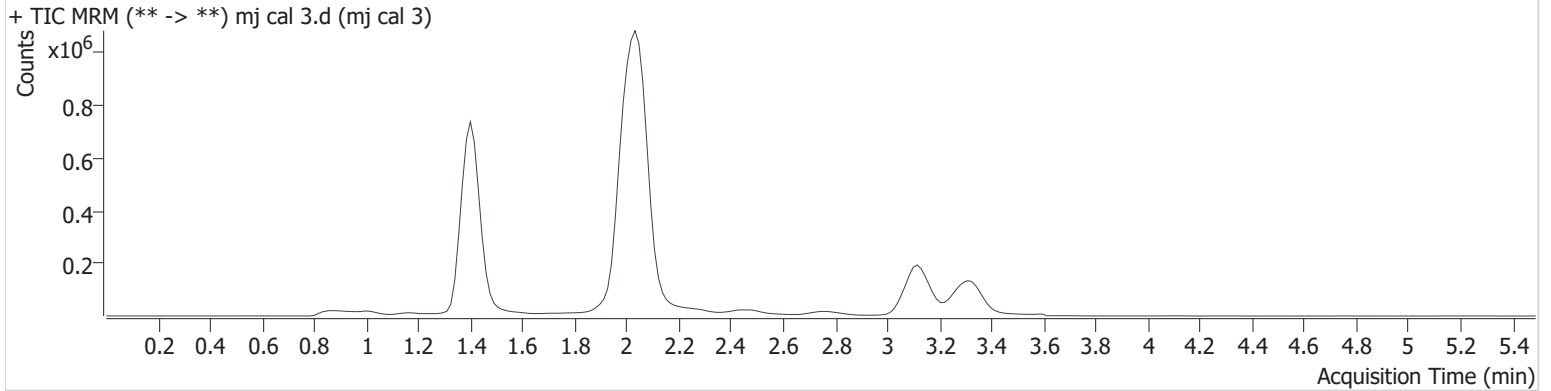
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.409	20922	∞	737.6	285.3	2150644	3.172 ng/ml
THC-COOH	1.431	85166	194.5	33.9	60.8	631506	9.873 ng/ml
THC	3.137	49713	∞	27.8	120.5	709102	2.878 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 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	2/25/2022 12:21:39 PM		
Sample Info.			

Sample Chromatogram



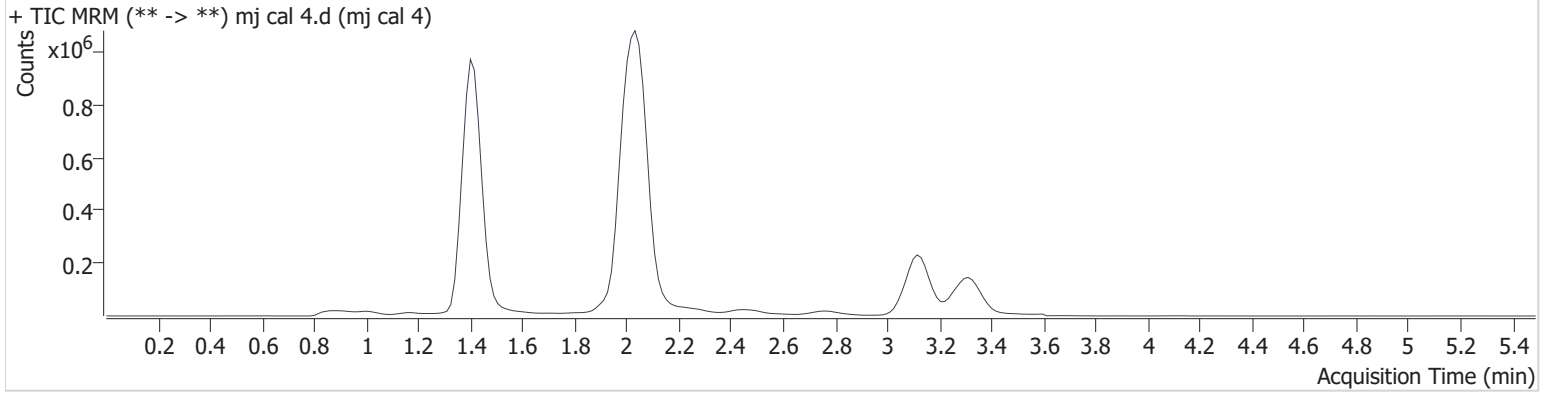
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.409	33887	1029.7	837.2	∞	2165340	4.888 ng/ml
THC-COOH	1.431	166274	687.6	35.9	4888.1	643635	18.493 ng/ml
THC	3.137	80819	3536177249 5376.8	24.5	340.7	682861	4.606 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 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	2/25/2022 12:28:21 PM		
Sample Info.			

Sample Chromatogram



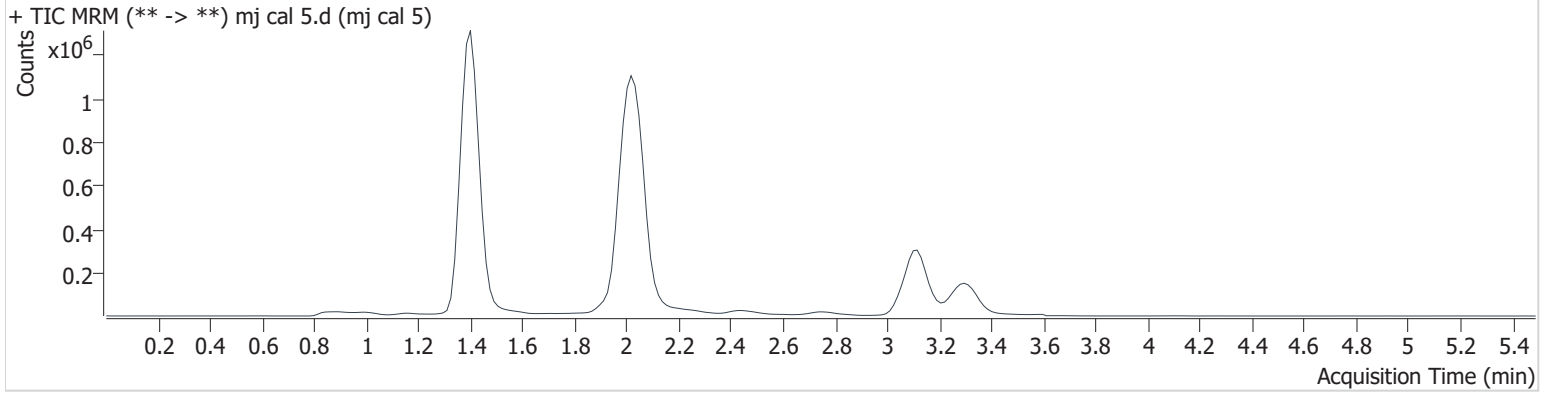
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	73216	∞	849.7	∞	2226544	9.884 ng/ml
THC-COOH	1.431	441840	516.8	38.1	1525.8	640790	48.597 ng/ml
THC	3.137	184631	∞	24.3	∞	755080	9.124 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 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	2/25/2022 12:35:04 PM		
Sample Info.			

Sample Chromatogram



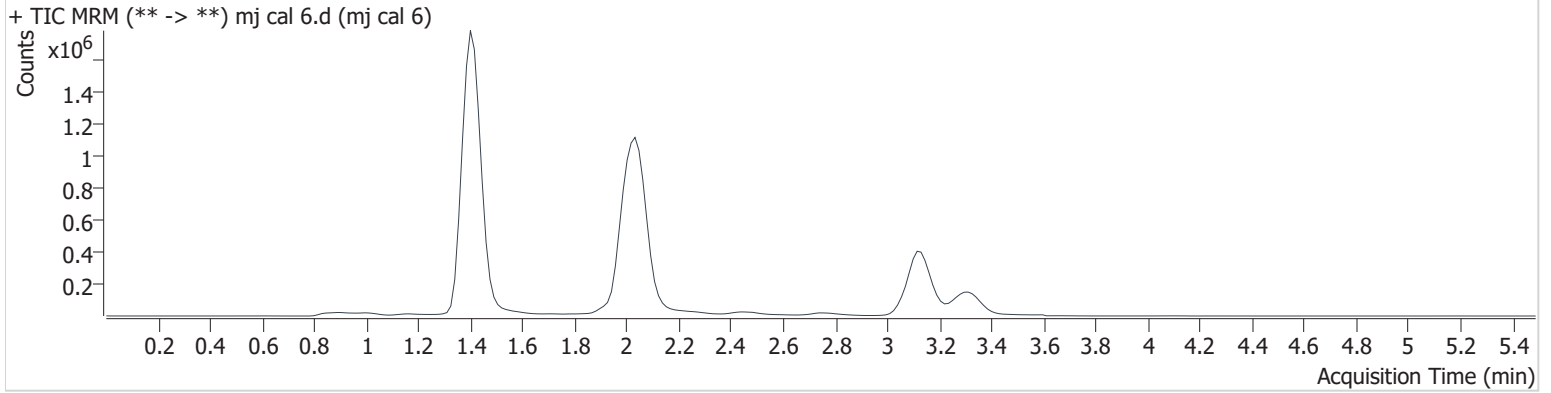
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	174133	∞	858.4	∞	2109359	24.282 ng/ml
THC-COOH	1.416	681294	2061.3	37.3	5535.1	633994	75.481 ng/ml
THC	3.122	516760	∞	23.5	789.7	771528	24.354 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 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	2/25/2022 12:41:46 PM		
Sample Info.			

Sample Chromatogram



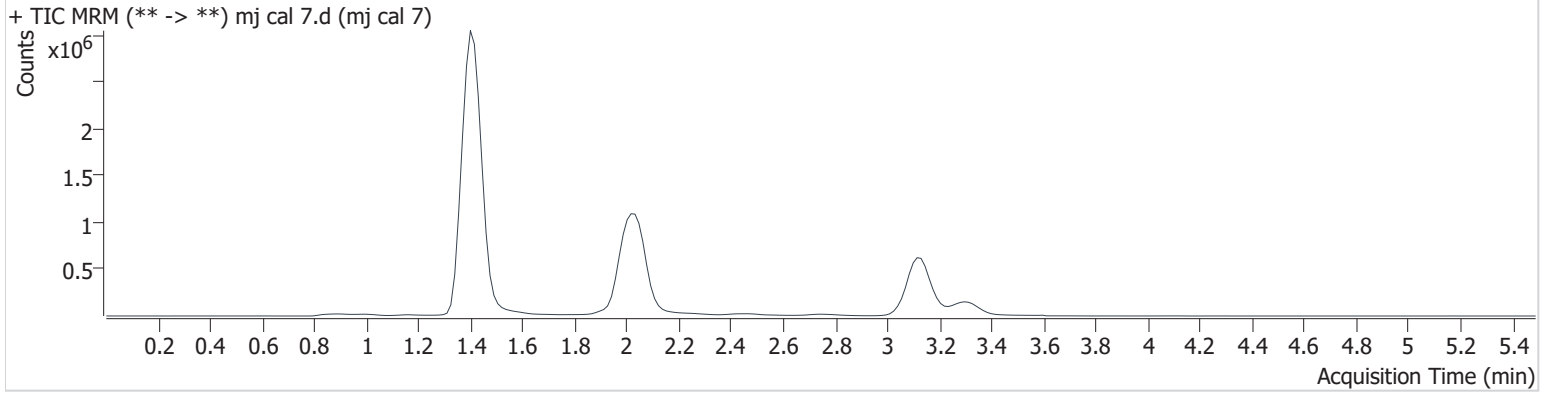
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	358909	∞	845.1	∞	2105201	49.772 ng/ml
THC-COOH	1.431	888358	4632.9	37.5	768.5	609515	102.212 ng/ml
THC	3.137	1062300	6085292078 484380.0	23.2	6730.7	766547	49.998 ng/ml

AM #27 Cannabinoids

Batch results D:\MassHunter\Data\2022\am 27-28\022522\QuantResults\cann.batch.bin
Calibration Last Update 2/28/2022 9:23:51 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	2/25/2022 12:48:28 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.394	719850	∞	841.2	∞	2073180	101.003 ng/ml
THC-COOH	1.431	2108026	3585.8	38.3	2135.6	589996	249.904 ng/ml
THC	3.137	2237056	∞	23.9	20890	789643	101.826 ng/ml
					21508		
					22600.		
					0		