TS
10/19/2023

Worklist: 6535

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION	
M2023-4038		COBCK	AM 27 Blood THC Quant by LC-QQQ	
M2023-4209	5	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-2690		BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-2706		BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-2971		BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-2977		BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-3022		BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-3037		BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-3053		BCK	AM 27 Blood THC Quant by LC-QQQ	
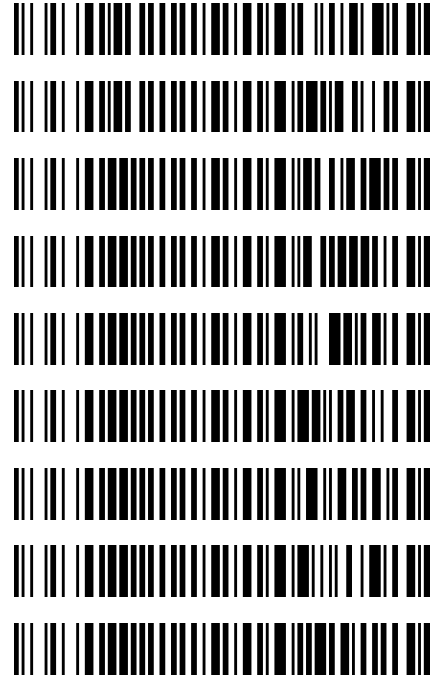
Worklist: 6529

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION	
P2023-2558	3	BCK	AM 27 Blood THC Quant by LC-QQQ	||||||||||||||||
P2023-2564	1	BCK	AM 27 Blood THC Quant by LC-QQQ	||||||||||||||||
P2023-2567	1	BLOOD	AM 27 Blood THC Quant by LC-QQQ	||||||||||||||||
P2023-2740	1	BCK	AM 27 Blood THC Quant by LC-QQQ	||||||||||||||||
P2023-2784	1	BCK	AM 27 Blood THC Quant by LC-QQQ	||||||||||||||||
P2023-2891	1	BCK	AM 27 Blood THC Quant by LC-QQQ	||||||||||||||||
P2023-3057	1	BCK	AM 27 Blood THC Quant by LC-QQQ	||||||||||||||||
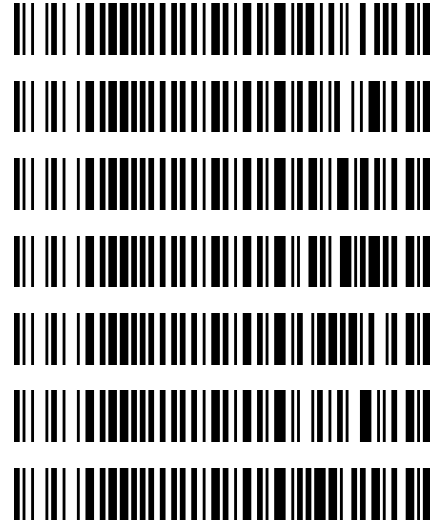
Samples will be re-extracted and ran at a later date.

TS

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

Extraction Date: 10/18/2023   Analyst: Celena Shrum
Plate lot#: 230627   Plate Retest Date: 12/27/2023
Mobile phase A: 0.1% Formic Acid in LCMS Water   **Mobile phase B:** 0.1% Formic acid in Acetonitrile
Blank Blood Lot: Lampire 23E52981   **Column**: Phenomenex Phenyl Hexyl (4.6x50mm, 2.6um)
LCMS-QQQ ID: 069901

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 (if applicable): add 1.5mL urine to blank plate, add 250μl 1N KOH. Shake and incubate at 40 degrees for 15 minutes.
☒ 3. Using a calibrated pipette, add **1000μl blood or 1000μl hydrolyzed urine** into the appropriate wells of the analytical (standards) plate. **Pipette ID: #42**
☒ 4. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
☒ 5. Add **500μL of 0.1% formic acid in water to blood samples or 500μl of saturated phosphate buffer to urine samples** to the appropriate wells of the analytical plate.
☒ 6. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
☒ 7. Transfer **800μL of blood+acid mixture or urine+acid** to corresponding wells of SLE+ plate.
☒ 8. 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: 067104
☒ 9. Wait 5 minutes.
☒ 10. Add **2.25mL MTBE. (*Add in 3 increments of 750uL*)**
☒ 11. Wait 5 minutes.
☒ 12. Apply positive pressure for approx. 15 seconds. *(10-15 PSI- Selector to the left).*
☒ 13. Add **2.25mL Hexane. (*Add in 3 increments of 750uL*)**
☒ 14. Wait 5 minutes.
☒ 15. Apply positive pressure for approx. 15 seconds. *(10-15 PSI- Selector to the left).*
☒ 16. Remove plate containing eluate. Place on SPE Dry and evaporate to dryness at approx. 35°C. *SPE Dry ID: 067103*
☒ 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 and OH-THC 3ng/mL (quantitative), Carboxy-THC: 5ng/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. THC concentrations of 1-3ng/mL will be reported qualitatively.
☒ 5. Did all QCs pass for each analyte? (if not, 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: The run stopped acquiring about half-way through. The samples that failed to acquire as well as the end QC were reconstituted, and the run was restarted the next day (10/19/23). The end QC had missed being reconstituted and did not inject, so it was reconstituted and reinjected on 10/20/23.   Curve Limits: THC 3-100- Calibrator 1 dropped due to accuracy and ratio.

Tamara Salazar included case samples in this run. Celena Shrum acted as the primary analyst and performed steps 4-17. I, Tamara Salazar, approve of all steps utilized in this method. TS

Due to poor chromatography in some compounds, the samples included in worklist 6529 will be re-extracted and ran at a later date.

	1	2	3	4	5	6
a				P2023-2784-1 TS	P2023-2977-1	QC 1
b				P2023-2740-1 TS	P2023-2971-1	cal 100 ng
c				P2023-2567-1 TS	P2023-2706-1	cal 50 ng
d				P2023-2564-1 TS	P2023-2690-1	cal 25 ng
e				P2023-2558-3 TS	M2023-4209-5	cal 10ng
f			P2023-3022-1	P2023-3053-1	M2023-4038-1	cal 5 ng
g			P2023-3057-1 TS	P2023-3037-1	NEG Blood	cal 3 ng
h			P2023-2891-1 TS	P2023-3022-1*	QC 2	cal 1ng

*Moved during SLE portion of the extraction

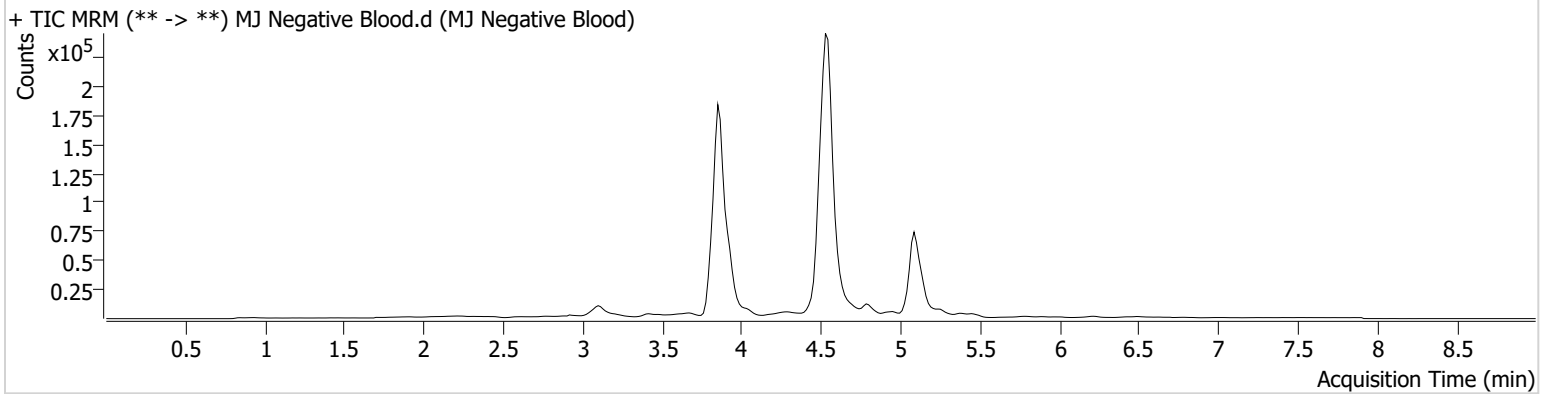# AM #27 Cannabinoids Quant. Results

Batch results   D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Calibration Last Update   10/20/2023 10:03:02 AM

Instrument	Falco (069901)	**Data File**	MJ Negative Blood.d
Type	Sample	**Sample**	MJ Negative Blood
Acq. Method	AM 27 Agilent Method.m	**Operator**	Celena Shrum
Sample Position	P1-G5	**Comment**	Only drugs and concentrations listed on the laboratory
Injection Volume	10		report itself are appropriate to be used for
Acq. Date-Time	10/18/2023 6:09:47 PM		interpretation purposes. Any drugs or values included
Sample Info.			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

+ TIC MRM (** -> **) MJ Negative Blood.d (MJ Negative Blood)
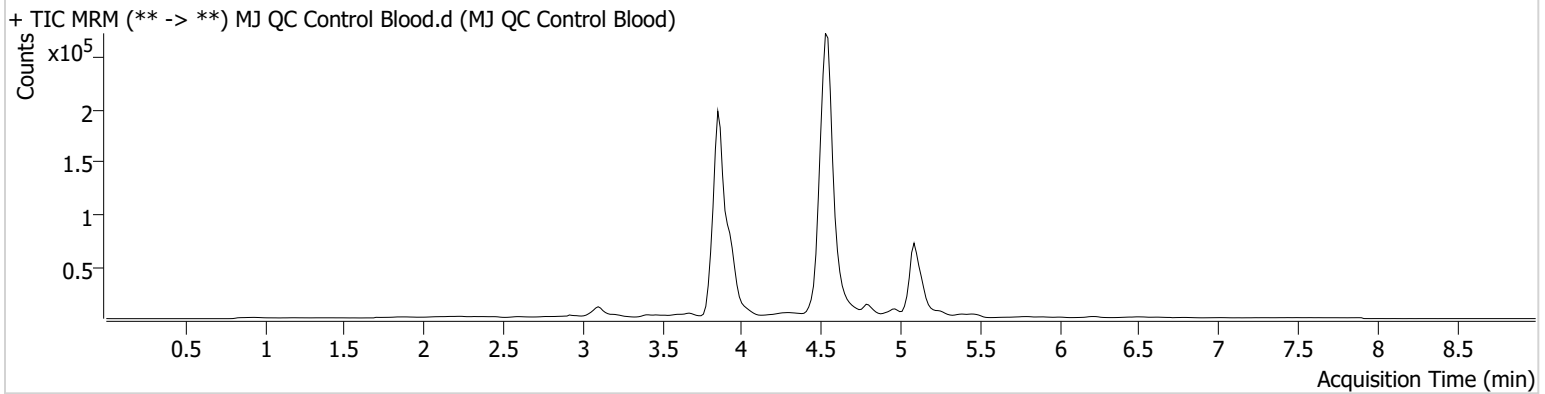
AM #27 Cannabinoids Quant. Results

Batch results   D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Calibration Last Update   10/20/2023 10:03:02 AM

Instrument	Falco (069901)	**Data File**	MJ QC Control Blood.d
Type	QC	**Sample**	MJ QC Control Blood
Acq. Method	AM 27 Agilent Method.m	**Operator**	Celena Shrum
Sample Position	P1-A6	**Comment**	*Only drugs and concentrations listed on the laboratory*
Injection Volume	10		*report itself are appropriate to be used for*
Acq. Date-Time	10/18/2023 5:43:33 PM		*interpretation purposes. Any drugs or values included*
Sample Info.			*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

+ TIC MRM (** -> **) MJ QC Control Blood.d (MJ QC Control Blood)

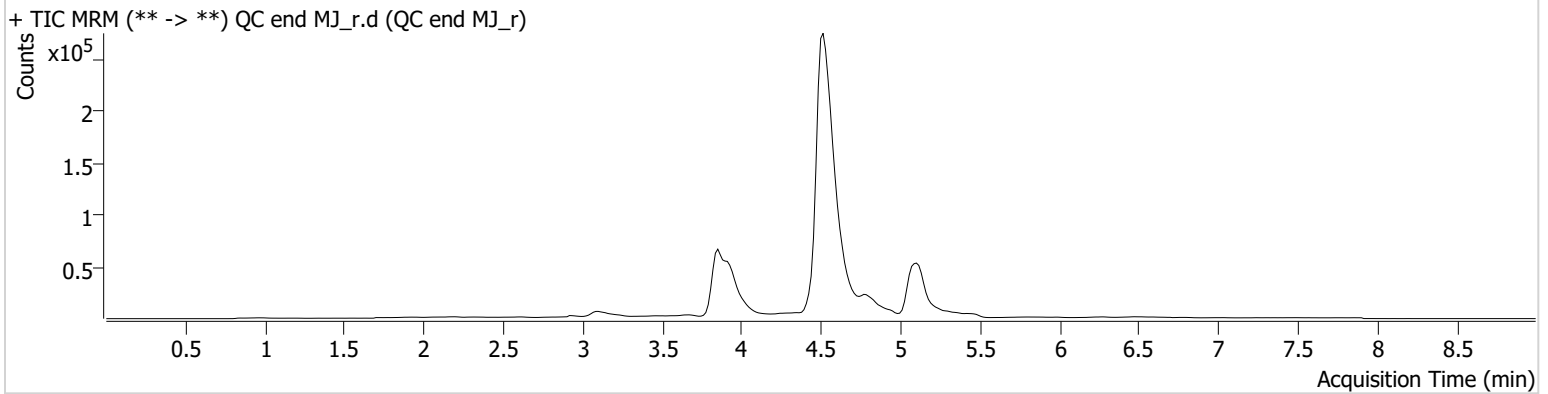| Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.090	11289	43.36	32.0	∞	271386	4.4886 ng/ml
THC-COOH	3.939	14527	272.23	245.6	675.65	163919	14.3398 ng/ml
THC-OH	3.865	51857	∞	13.5	72.89	861140	4.3919 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Calibration Last Update 10/20/2023 10:03:02 AM

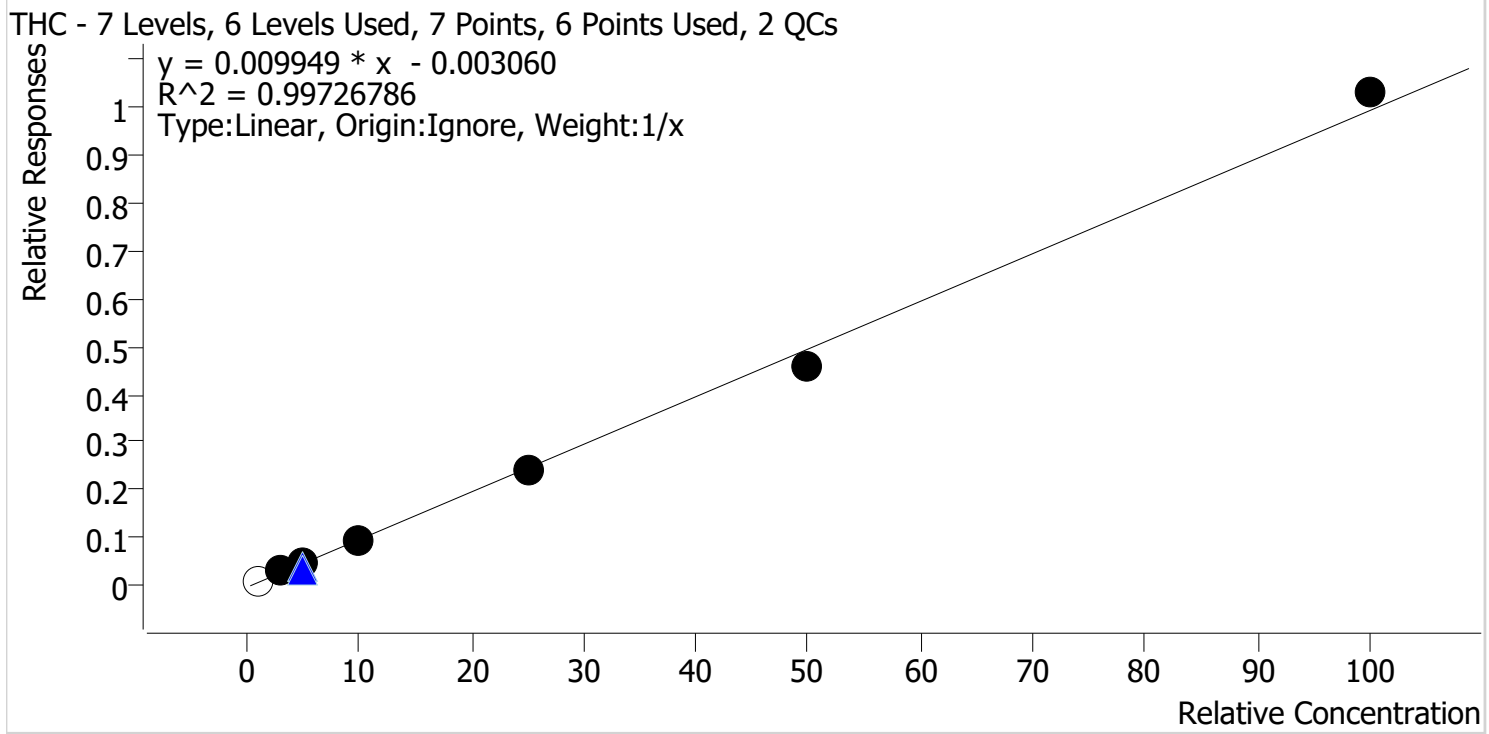|			
Instrument	Falco (069901)	**Data File**	QC end MJ_r.d
Type	QC	**Sample**	QC end MJ_r
Acq. Method	AM 27 Agilent Method.m	**Operator**	Celena Shrum
Sample Position	P1-H5	**Comment**	Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. 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	10/20/2023 8:44:32 AM		
Sample Info.			

Sample Chromatogram

+ TIC MRM (** -> **) QC end MJ_r.d (QC end MJ_r)

Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.090	10624	∞	29.1	16.91	317024	3.6758 ng/ml
THC-COOH	3.954	7963	194.04	251.6	∞	83159	15.4394 ng/ml
THC-OH	3.865	27863	∞	12.7	23.87	413341	4.9174 ng/ml

TS

AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results   D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Last Cal. Update   10/20/2023 10:03 AM
Analyst Name   ISP\Datastor
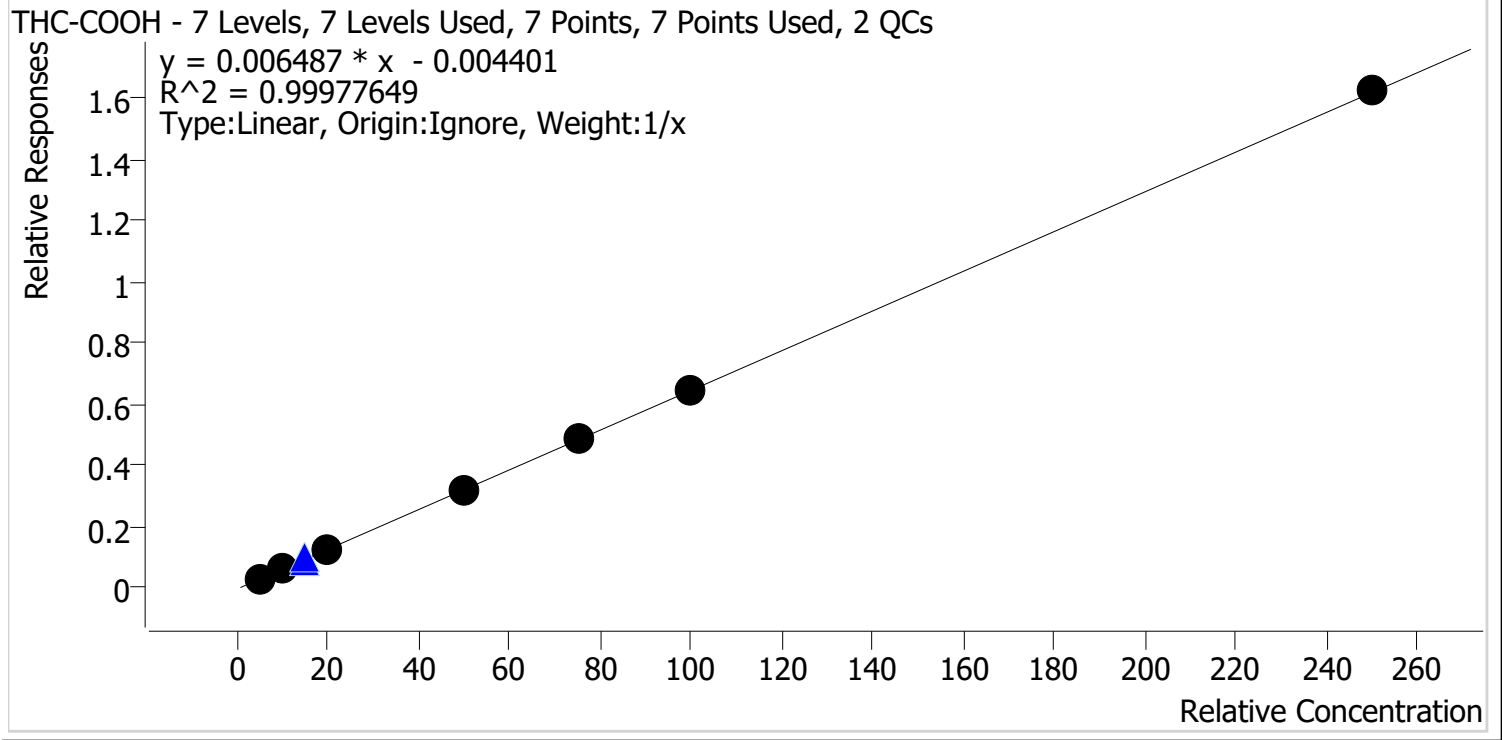**Analyte**   THC   **Internal Standard**   THC-D3

THC - 7 Levels, 6 Levels Used, 7 Points, 6 Points Used, 2 QCs
$y = 0.009949 * x - 0.003060$
$R^2 = 0.99726786$
Type:Linear, Origin:Ignore, Weight:1/x

Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	✗	1.0	1.4	138.7
Cal 2 MJ	2	✔	3.0	3.2	107.6
Cal 3 MJ	3	✔	5.0	5.0	99.1
Cal 4 MJ	4	✔	10.0	9.8	97.6
Cal 5 MJ	5	✔	25.0	24.6	98.5
Cal 6 MJ	6	✔	50.0	46.7	93.4
Cal 7 MJ	7	✔	100.0	103.7	103.7

AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Last Cal. Update 10/20/2023 10:03 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, 2 QCs

$y = 0.006487 * x - 0.004401$
$R^2 = 0.99977649$
Type:Linear, Origin:Ignore, Weight:1/x

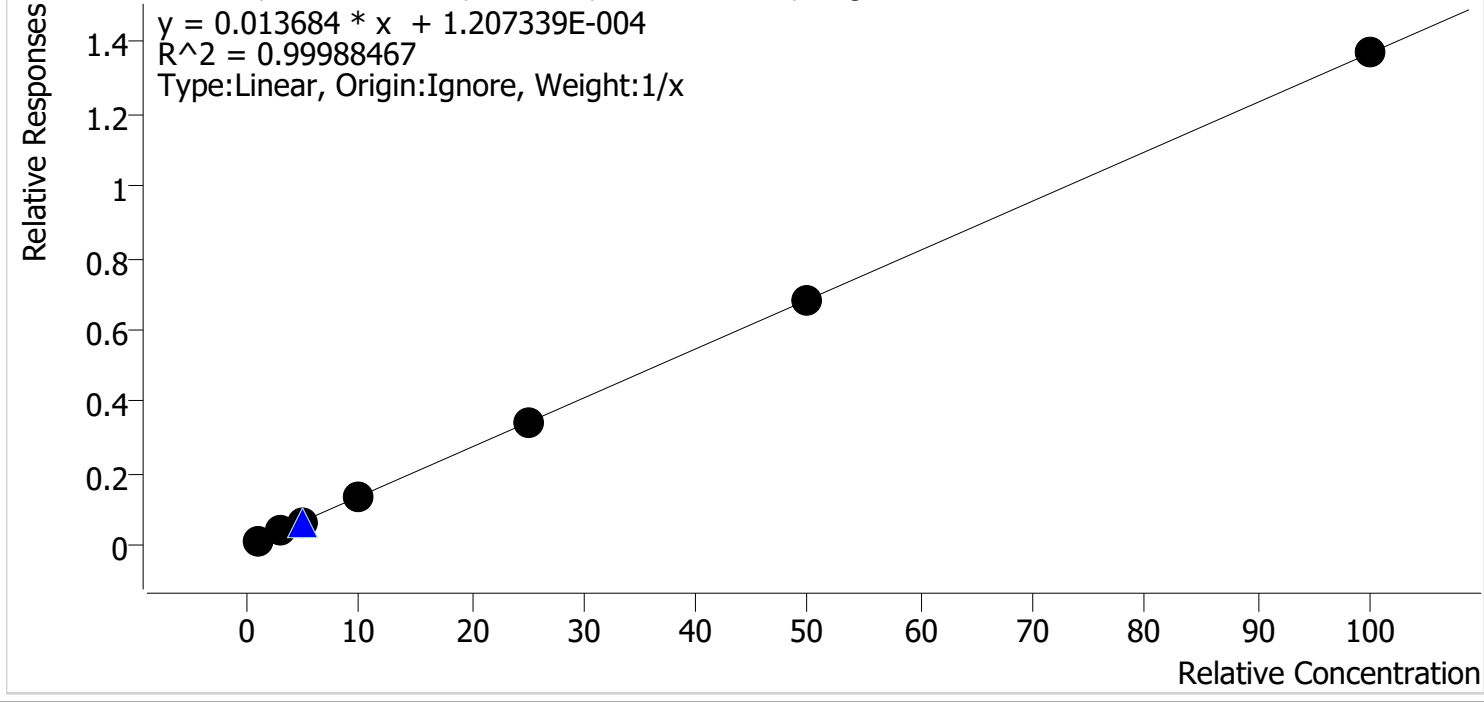| Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	✔	5.0	5.1	102.7
Cal 2 MJ	2	✔	10.0	10.1	101.1
Cal 3 MJ	3	✔	20.0	19.4	97.2
Cal 4 MJ	4	✔	50.0	48.8	97.6
Cal 5 MJ	5	✔	75.0	76.4	101.9
Cal 6 MJ	6	✔	100.0	99.2	99.2
Cal 7 MJ	7	✔	250.0	250.9	100.4

TS   cg

AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results   D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Last Cal. Update   10/20/2023 10:03 AM
Analyst Name   ISP\Datastor
Analyte   THC-OH   **Internal Standard**   THC-OH-D3

THC-OH - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 2 QCs

$y = 0.013684 * x + 1.207339E\text{-}004$
$R^2 = 0.99988467$
Type:Linear, Origin:Ignore, Weight:1/x

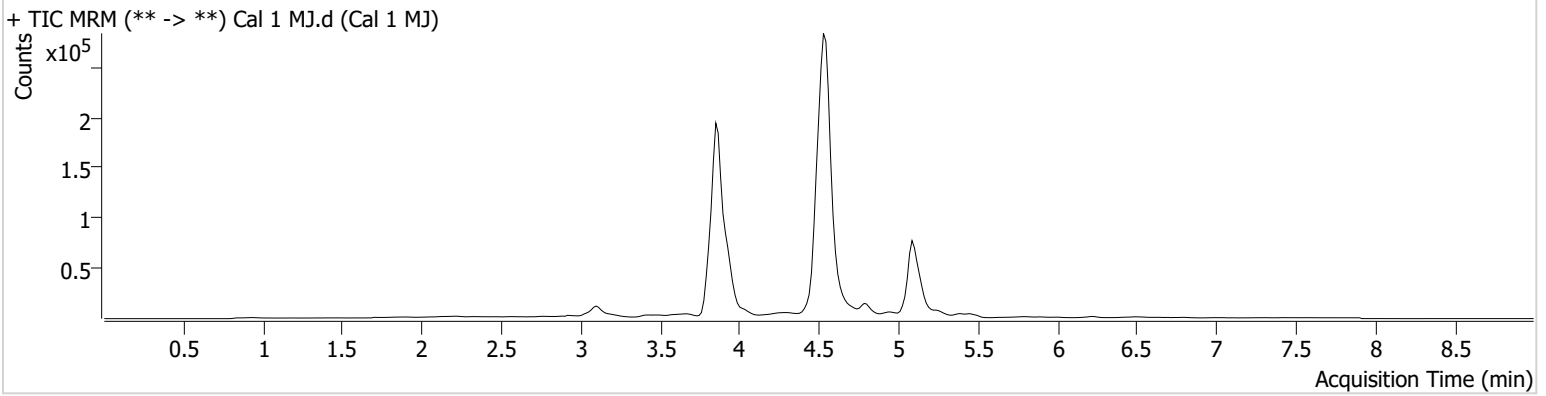| Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	✔	1.0	1.0	102.5
Cal 2 MJ	2	✔	3.0	3.1	101.7
Cal 3 MJ	3	✔	5.0	4.8	95.3
Cal 4 MJ	4	✔	10.0	10.0	99.5
Cal 5 MJ	5	✔	25.0	25.3	101.4
Cal 6 MJ	6	✔	50.0	49.8	99.5
Cal 7 MJ	7	✔	100.0	100.1	100.1

Generated at 10:04 AM on 10/20/2023

AM #27 Cannabinoids Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Calibration Last Update 10/20/2023 10:03:02 AM

Instrument	Falco (069901)	**Data File**	Cal 1 MJ.d
Type	Cal	**Sample**	Cal 1 MJ
Acq. Method	AM 27 Agilent Method.m	**Operator**	Celena Shrum
Sample Position	P1-H6	**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	10/18/2023 3:58:27 PM		
Sample Info.			

Sample Chromatogram

+ TIC MRM (** -> **) Cal 1 MJ.d (Cal 1 MJ)

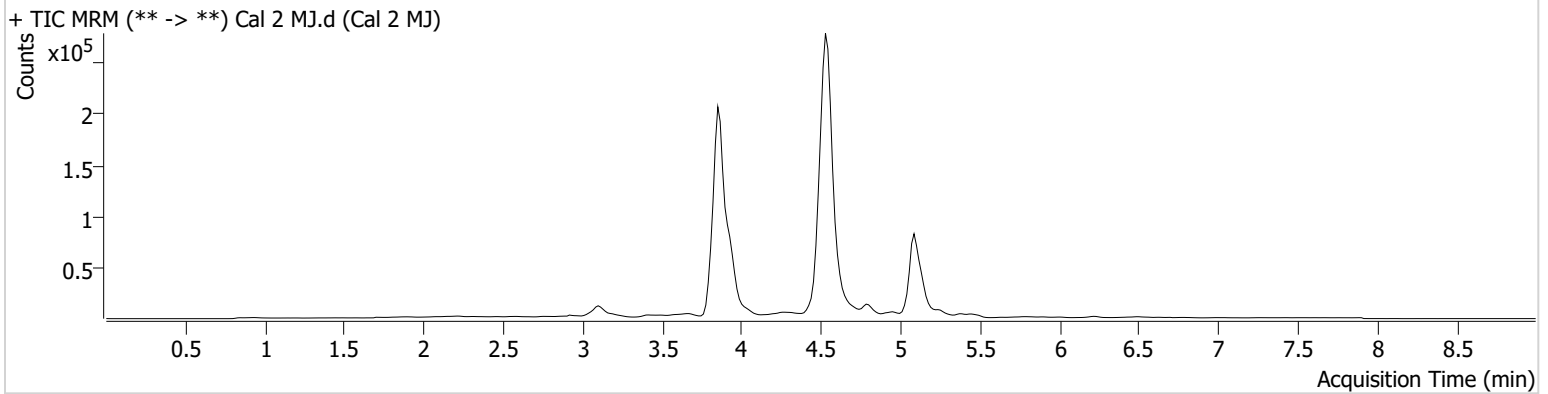| Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.090	3358	25.60	56.0 **High**	∞	312854	1.3865 ng/ml
THC-COOH	3.939	4886	129.63	252.6	∞	168986	5.1353 ng/ml
THC-OH	3.865	12740	∞	12.5	15.34	900971	1.0246 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results　　　　　D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Calibration Last Update　10/20/2023 10:03:02 AM

Instrument	Falco (069901)	**Data File**	Cal 2 MJ.d
Type	Cal	**Sample**	Cal 2 MJ
Acq. Method	AM 27 Agilent Method.m	**Operator**	Celena Shrum
Sample Position	P1-G6	**Comment**	Only drugs and concentrations listed on the laboratory
Injection Volume	10		report itself are appropriate to be used for
Acq. Date-Time	10/18/2023 4:11:43 PM		interpretation purposes. Any drugs or values included
Sample Info.			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

+ TIC MRM (** -> **) Cal 2 MJ.d (Cal 2 MJ)

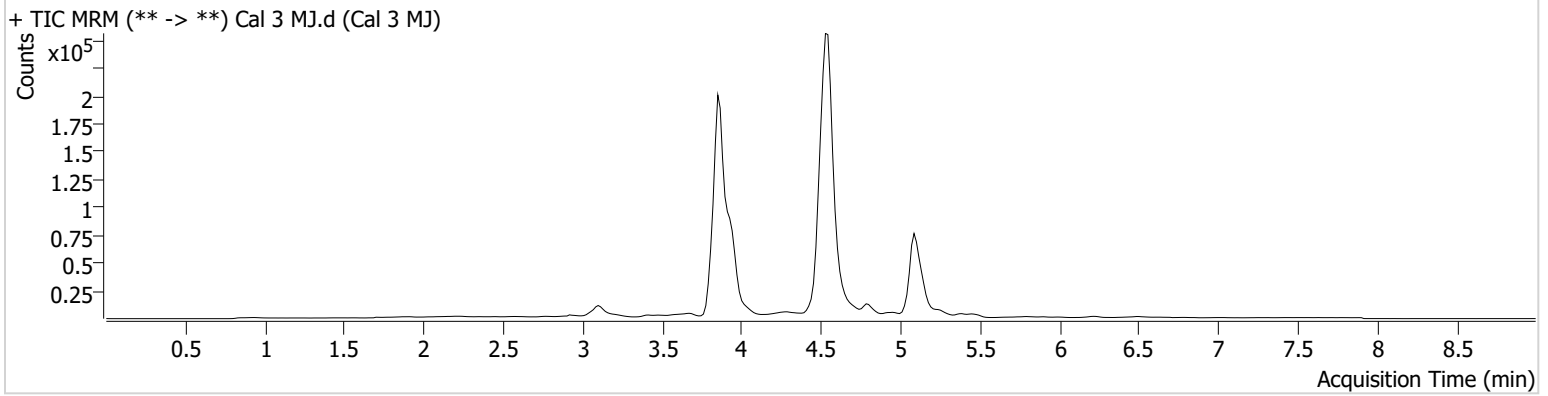| Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.090	9393	∞	31.4	24.66	323107	3.2294 ng/ml
THC-COOH	3.939	10473	191.90	241.1	193.95	171118	10.1136 ng/ml
THC-OH	3.865	37843	∞	14.3	∞	903533	3.0520 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results   D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Calibration Last Update   10/20/2023 10:03:02 AM

Instrument	Falco (069901)	**Data File**	Cal 3 MJ.d
Type	Cal	**Sample**	Cal 3 MJ
Acq. Method	AM 27 Agilent Method.m	**Operator**	Celena Shrum
Sample Position	P1-F6	**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	10/18/2023 4:24:49 PM		
Sample Info.			

Sample Chromatogram

+ TIC MRM (** -> **) Cal 3 MJ.d (Cal 3 MJ)

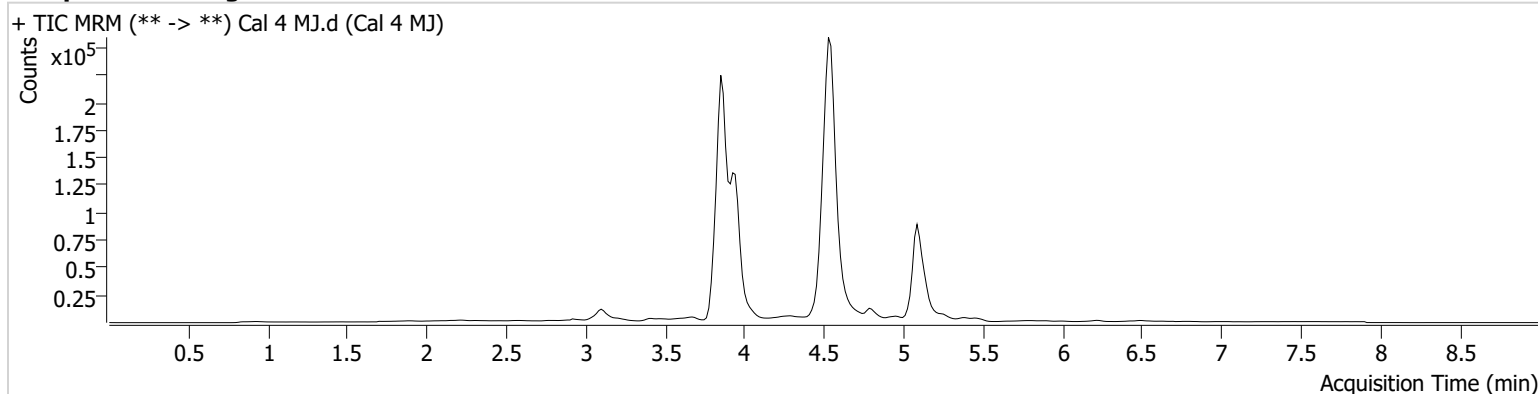| Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.090	14484	∞	27.7	∞	313087	4.9572 ng/ml
THC-COOH	3.939	20074	∞	244.1	1055.45	165022	19.4311 ng/ml
THC-OH	3.865	55497	∞	14.5	118.62	849974	4.7627 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results   D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Calibration Last Update   10/20/2023 10:03:02 AM

Instrument	Falco (069901)	**Data File**	Cal 4 MJ.d
Type	Cal	**Sample**	Cal 4 MJ
Acq. Method	AM 27 Agilent Method.m	**Operator**	Celena Shrum
Sample Position	P1-E6	**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	10/18/2023 4:37:56 PM		
Sample Info.			

Sample Chromatogram

+ TIC MRM (** -> **) Cal 4 MJ.d (Cal 4 MJ)

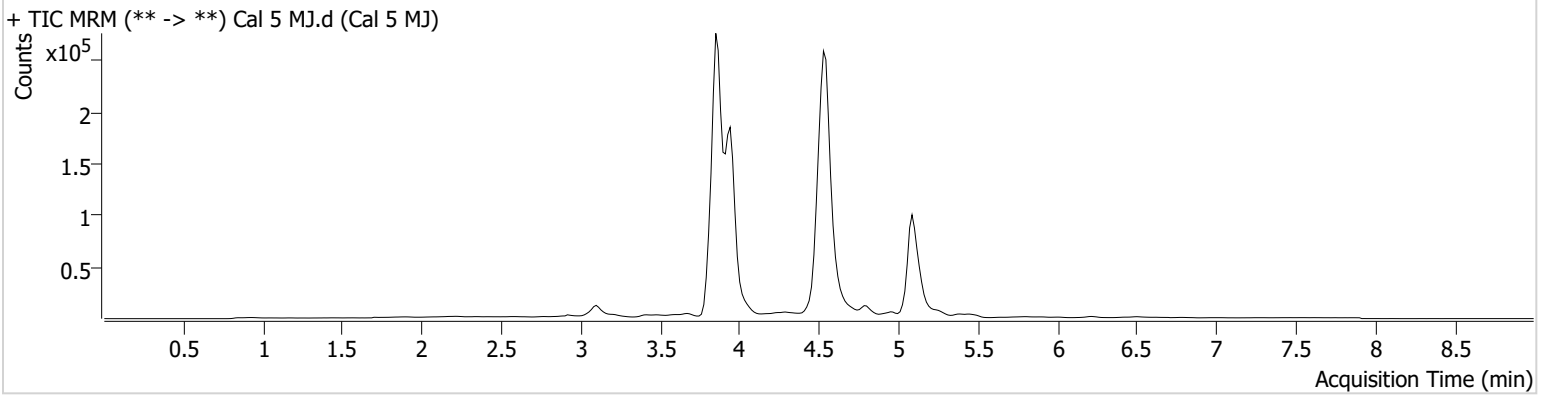| Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.090	30119	∞	28.4	∞	320337	9.7579 ng/ml
THC-COOH	3.939	52489	2009.52	244.7	∞	168213	48.7819 ng/ml
THC-OH	3.865	120692	∞	15.2	∞	885247	9.9545 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results   D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Calibration Last Update   10/20/2023 10:03:02 AM

Instrument	Falco (069901)	**Data File**	Cal 5 MJ.d
Type	Cal	**Sample**	Cal 5 MJ
Acq. Method	AM 27 Agilent Method.m	**Operator**	Celena Shrum
Sample Position	P1-D6	**Comment**	Only drugs and concentrations listed on the laboratory
Injection Volume	10		report itself are appropriate to be used for
Acq. Date-Time	10/18/2023 4:51:04 PM		interpretation purposes. Any drugs or values included
Sample Info.			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

+ TIC MRM (** -> **) Cal 5 MJ.d (Cal 5 MJ)

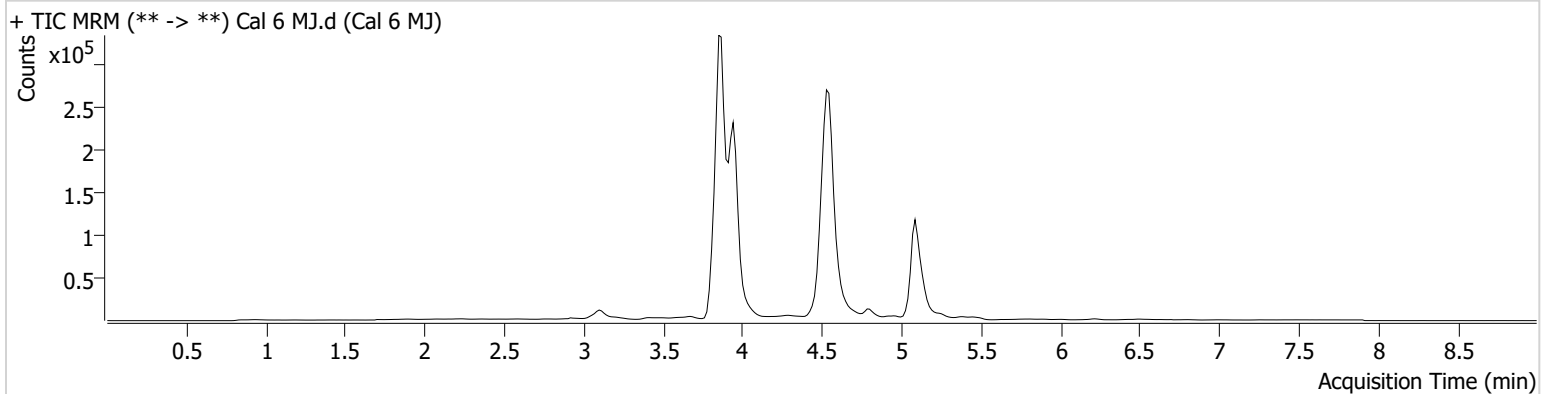| Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.090	76134	508.89	26.3	39.17	314625	24.6293 ng/ml
THC-COOH	3.939	83443	∞	242.1	2175.72	169894	76.3922 ng/ml
THC-OH	3.865	314485	∞	14.5	869.33	906343	25.3482 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results   D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Calibration Last Update   10/20/2023 10:03:02 AM

Instrument	Falco (069901)	**Data File**	Cal 6 MJ.d
Type	Cal	**Sample**	Cal 6 MJ
Acq. Method	AM 27 Agilent Method.m	**Operator**	Celena Shrum
Sample Position	P1-C6	**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	10/18/2023 5:04:12 PM		
Sample Info.			

Sample Chromatogram

+ TIC MRM (** -> **) Cal 6 MJ.d (Cal 6 MJ)

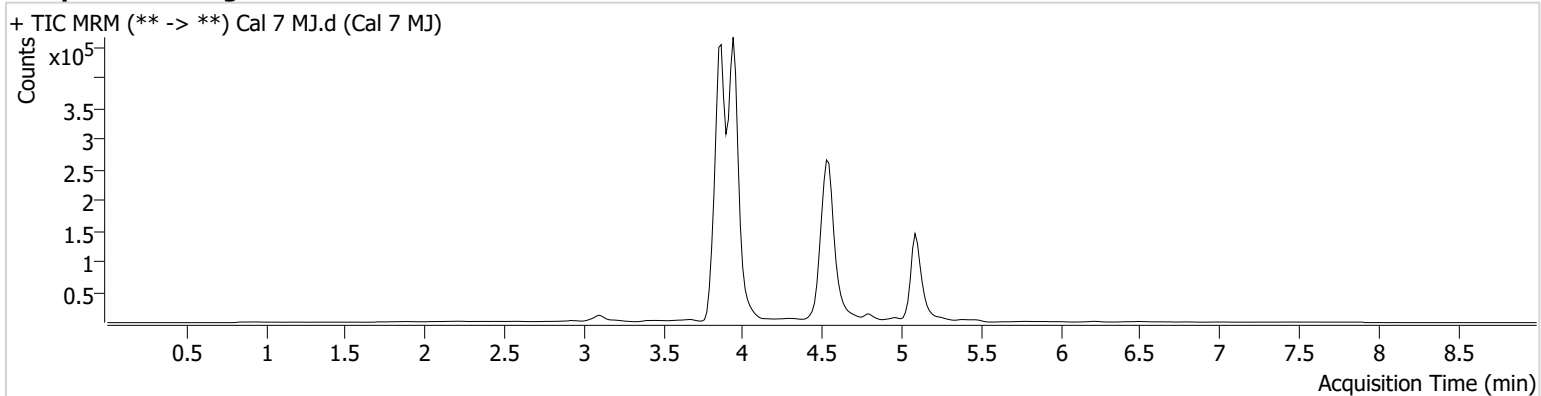| Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.090	139050	∞	26.5	∞	301336	46.6874 ng/ml
THC-COOH	3.939	101200	2477.49	247.4	1868.43	158335	99.2079 ng/ml
THC-OH	3.865	589195	∞	14.5	529.84	865136	49.7610 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results   D:\MassHunter\Data\2023\AM 27 28\101823 AM 27 28 CS TS\QuantResults\AM 27.batch.bin
Calibration Last Update   10/20/2023 10:03:02 AM

Instrument	Falco (069901)	**Data File**	Cal 7 MJ.d
Type	Cal	**Sample**	Cal 7 MJ
Acq. Method	AM 27 Agilent Method.m	**Operator**	Celena Shrum
Sample Position	P1-B6	**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	10/18/2023 5:17:18 PM		
Sample Info.			

Sample Chromatogram

+ TIC MRM (** -> **) Cal 7 MJ.d (Cal 7 MJ)

Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.090	265815	∞	24.8	∞	258307	103.7388 ng/ml
THC-COOH	3.939	248874	5442.22	240.6	9041.00	153303	250.9380 ng/ml
THC-OH	3.865	1137037	∞	14.7	∞	830054	100.0970 ng/ml