

Worklist: 6118

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>	
M2022-3908	4	BCK	AM 27 Blood THC Quant by LC-QQQ	
M2022-3912	3	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-2664	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-2760	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-2787	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-2825	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-2887	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-2892	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
* P2022-2894	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-2909	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-2931	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-2950	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-2953	1	BCK	AM 27 Blood THC Quant by LC-QQQ	

*Sample was not ran with this batch due to insufficient sample

SC

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

Extraction Date: 10/05/2022

Analyst: Sarah Collins

Plate lot#: IDP-108-3-220802

Retest Date: 02/02/2023

Mobile phase A: 0.1% Formic Acid in LCMS Water

Mobile phase B: 0.1% Formic acid in Acetonitrile

Blank Blood Lot: Lampire 22B52015-1

Blank Urine Lot: N/A

Column: UCT Selectra DA 100 x 2.1mm 3um

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: add 1.5mL urine to blank plate, add 250µl 1N KOH. Shake and incubate at 40 degrees for 15 minutes. Using a calibrated pipette, add **1000µl blood and urine (if applicable) (calibrated pipette)** into the appropriate wells of analytical (standards) plate. **Pipette ID:** 3382167
- 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 4. Pipette **500µL 0.1% formic acid in water blood sample, 500 µL saturated phosphate buffer in urine** in wells of analytical plate.
- 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 6. Transfer **700-800µL of blood+acid or urine+acid** mixture to corresponding wells of SLE+ plate. Amount transferred: 800 uL
- 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)**
- 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.
- 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.
- 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: Curve Range: THC-OH 3-100 (calibrator 1 dropped due to ratio), Carboxy-THC 10-250 (calibrator 1 dropped due to ratio)

SC

	1	2	3	4	5	6
A	IS + Cal. 1	IS + QC_1 urine		p2022-2909-1	p2022-2825-1	IS + QC_1 blood start
B	IS + Cal. 2			p2022-2953-1	p2022-2787-1	IS + Cal. 7
C	IS + Cal. 3			p2022-2950-1	p2022-2760-1	IS + Cal. 6
D	IS + Cal. 4			p2022-2931-1	p2022-2664-1	IS + Cal. 5
E	IS + Cal. 5			p2022-2909-1*	m2022-3912-3	IS + Cal. 4
F	IS + Cal. 6				m2022-3908-4	IS + Cal. 3
G	IS + Cal. 7			p2022-2892-1	negative blood	IS + Cal. 2
H	IS + QC_1 blood			p2022-2887-1	IS + QC_1 blood end	IS + Cal. 1

All wells to contain 100 µl of residual DMSO

SC

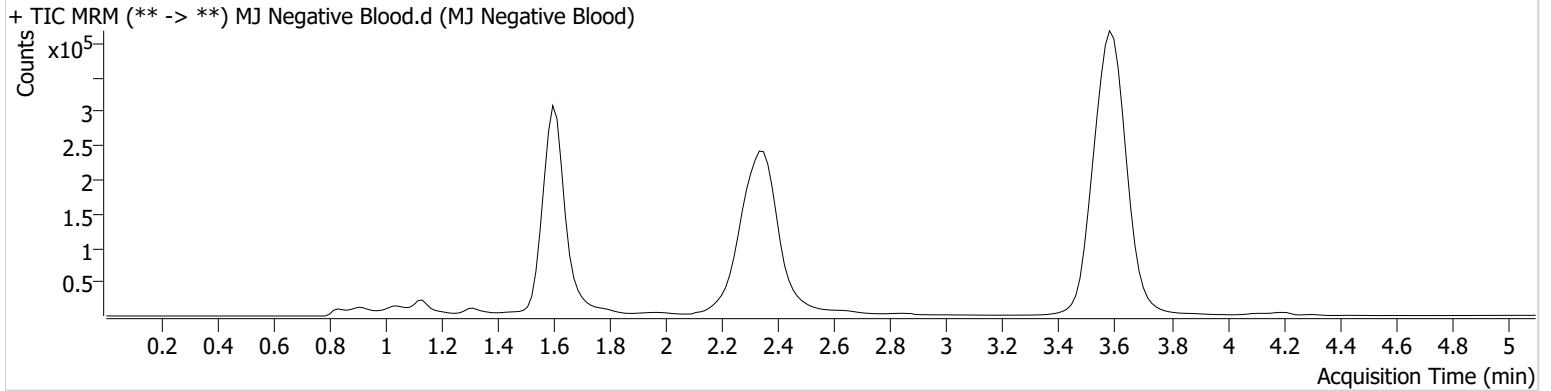


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/6/2022 1:15:18 PM

Instrument	Falco (069901)	Data File	MJ Negative Blood.d
Type	Sample	Sample	MJ Negative Blood
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P5-G5	Comment	
Injection Volume	10		
Acq. Date-Time	10/5/2022 2:00:25 PM		
Sample Info.			

Sample Chromatogram



SC

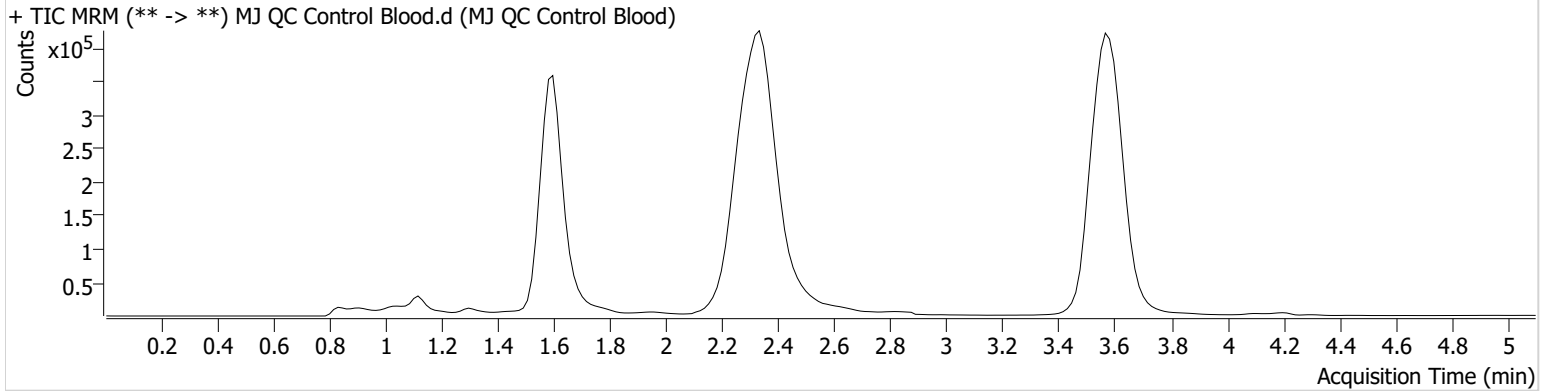


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/6/2022 1:15:18 PM

Instrument	Falco (069901)	Data File	MJ QC Control Blood.d
Type	QC	Sample	MJ QC Control Blood
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P5-A6	Comment	
Injection Volume	10		
Acq. Date-Time	10/5/2022 1:45:14 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	120934	∞	13.0	262.81	1464038	4.7682 ng/ml
THC-COOH	1.625	116394	∞	44.4	∞	255130	14.7687 ng/ml
THC	3.586	147338	632.28	30.5	∞	3188181	4.9986 ng/ml

SC

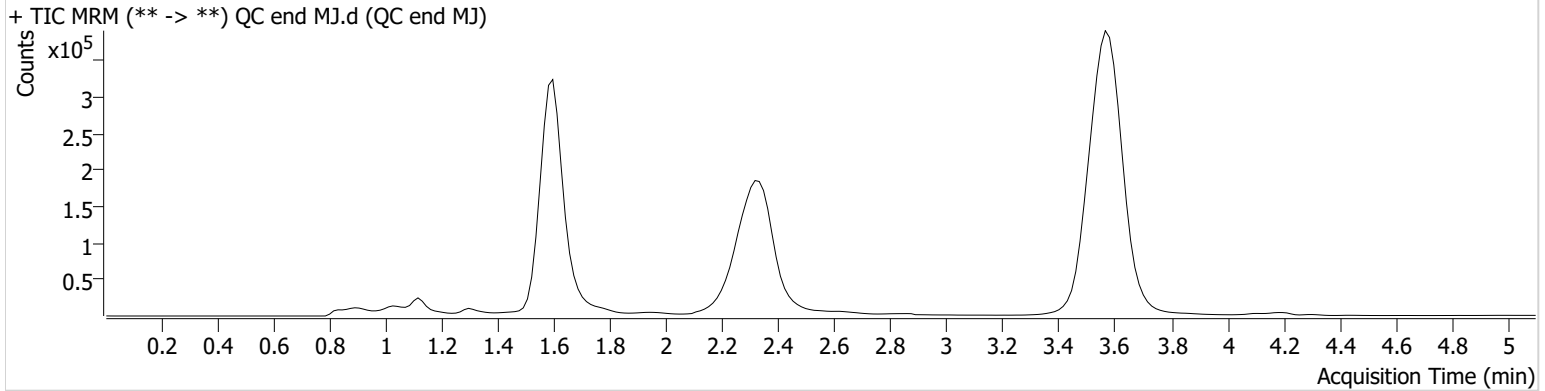


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/6/2022 1:15:18 PM

Instrument	Falco (069901)	Data File	QC end MJ.d
Type	QC	Sample	QC end MJ
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P5-H5	Comment	
Injection Volume	10		
Acq. Date-Time	10/5/2022 5:18:21 PM		

Sample Chromatogram



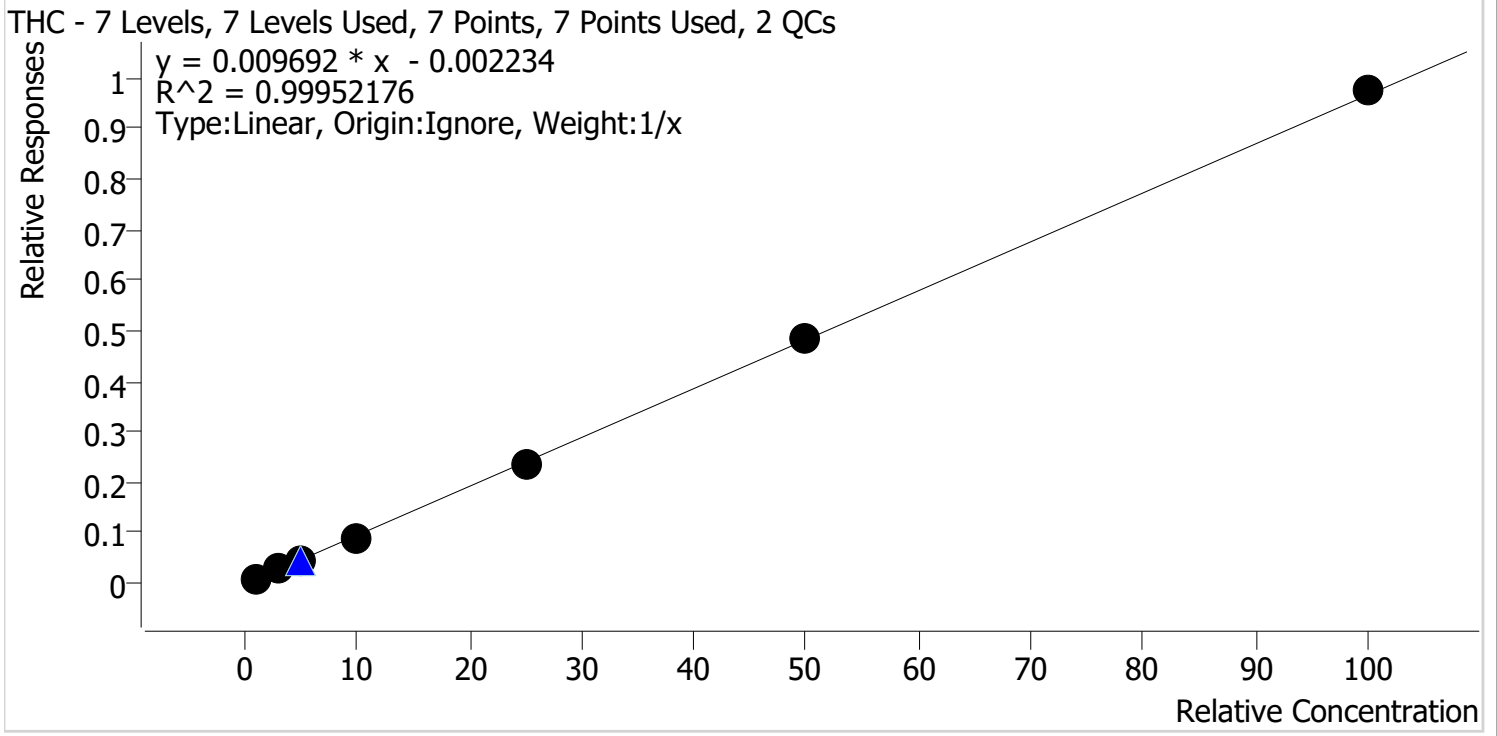
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	110263	∞	13.0	328.06	1347101	4.7212 ng/ml
THC-COOH	1.625	93216	∞	52.2	717.32	238898	12.0671 ng/ml
THC	3.586	138687	1239.10	29.4	57.01	3102969	4.8420 ng/ml

SC



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update 10/6/2022 1:15 PM
Analyst Name ISP\datastor
Analyte THC **Internal Standard** THC-D3



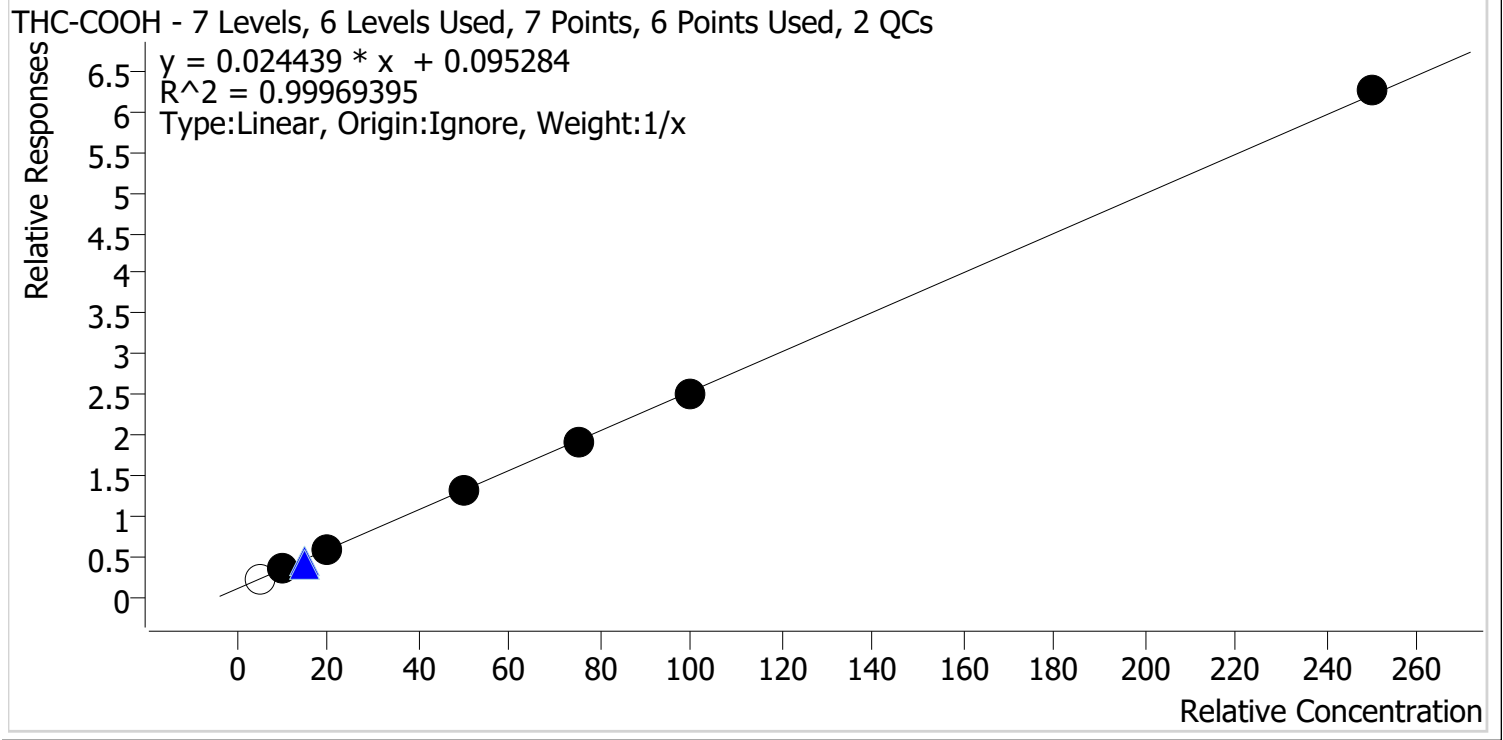
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	✓	1.0	1.1	112.5
Cal 2 MJ	2	✓	3.0	2.9	97.8
Cal 3 MJ	3	✓	5.0	4.7	94.7
Cal 4 MJ	4	✓	10.0	9.6	96.1
Cal 5 MJ	5	✓	25.0	24.3	97.1
Cal 6 MJ	6	✓	50.0	50.5	100.9
Cal 7 MJ	7	✓	100.0	100.9	100.9

SC



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update 10/6/2022 1:15 PM
Analyst Name ISP\datastor
Analyte THC-COOH **Internal Standard** THC-COOH-D9



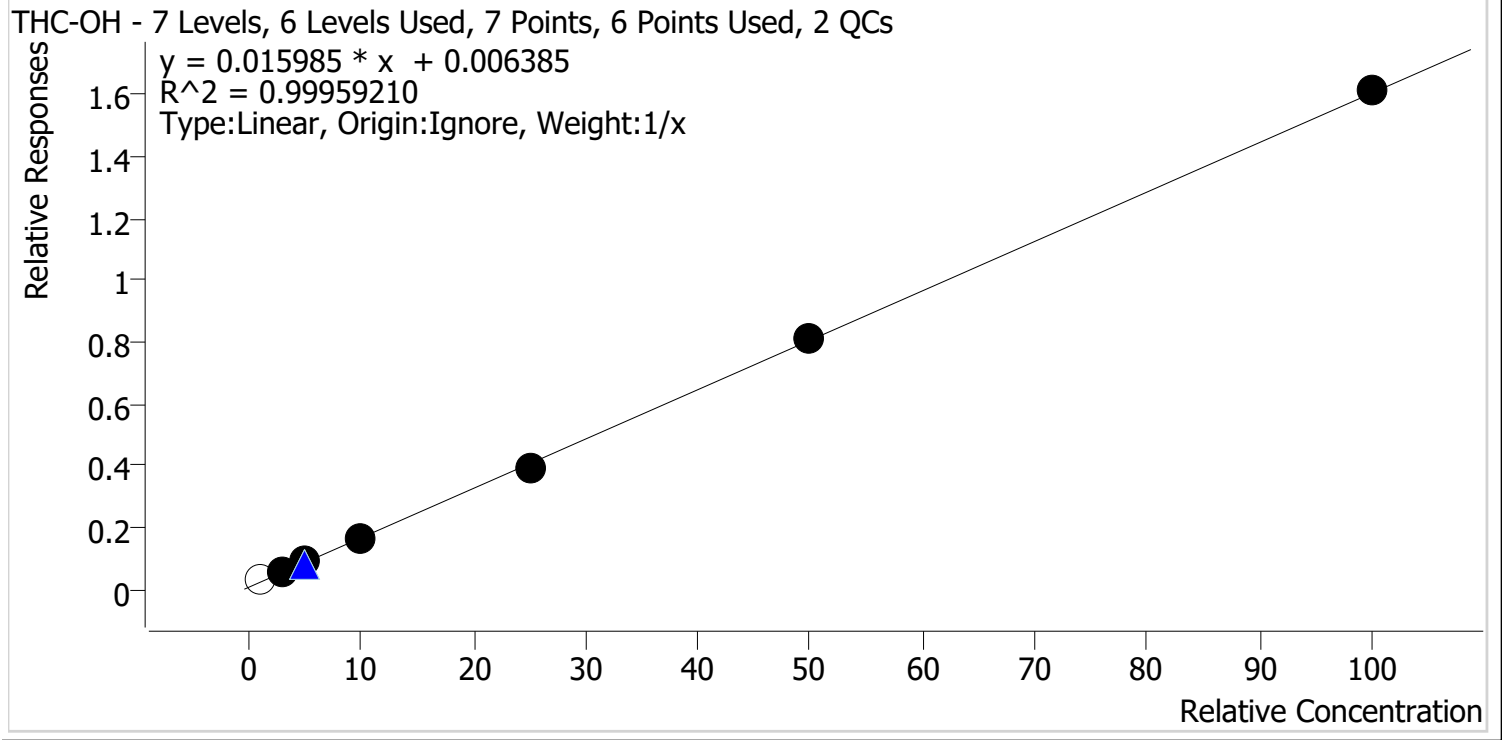
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	x	5.0	5.1	101.0
Cal 2 MJ	2	✓	10.0	10.6	105.6
Cal 3 MJ	3	✓	20.0	19.2	96.1
Cal 4 MJ	4	✓	50.0	49.6	99.3
Cal 5 MJ	5	✓	75.0	74.3	99.0
Cal 6 MJ	6	✓	100.0	99.1	99.1
Cal 7 MJ	7	✓	250.0	252.2	100.9

SC



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update 10/6/2022 1:15 PM
Analyst Name ISP\datastor
Analyte THC-OH **Internal Standard** THC-OH-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	x	1.0	1.7	172.9
Cal 2 MJ	2	✓	3.0	3.1	104.4
Cal 3 MJ	3	✓	5.0	5.1	101.2
Cal 4 MJ	4	✓	10.0	9.6	96.1
Cal 5 MJ	5	✓	25.0	24.2	96.7
Cal 6 MJ	6	✓	50.0	50.5	101.1
Cal 7 MJ	7	✓	100.0	100.5	100.5

SC

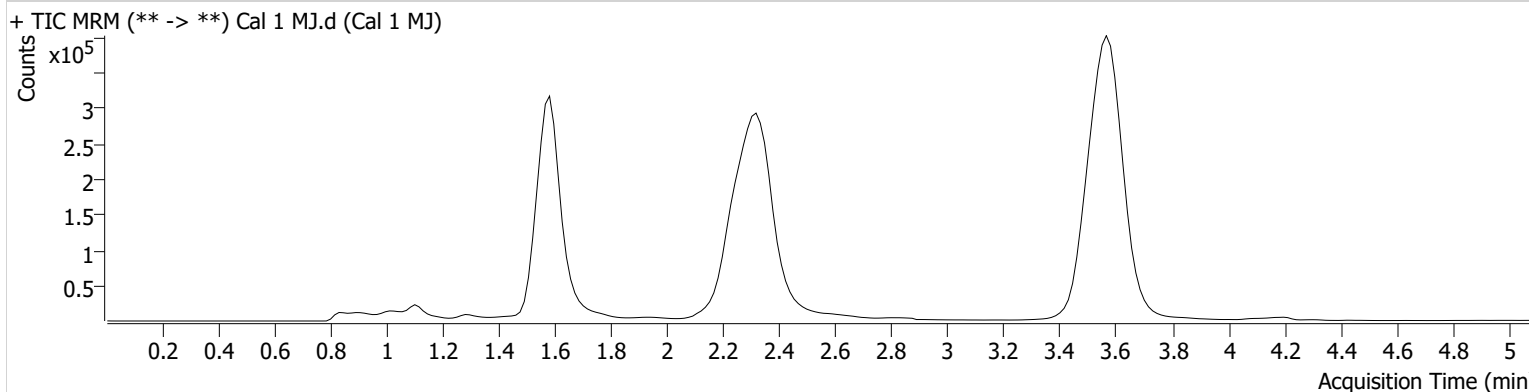


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/6/2022 1:15:18 PM

Instrument	Falco (069901)	Data File	Cal 1 MJ.d
Type	Cal	Sample	Cal 1 MJ
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P5-H6	Comment	
Injection Volume	10		
Acq. Date-Time	10/5/2022 12:44:13 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.648	51083	∞	7.2 Low	32.51	1501706	1.7286 ng/ml Low
THC-COOH	1.610	57039	∞	29.8 Low	149.73	260769	5.0514 ng/ml
THC	3.586	30465	77.11	34.9	∞	3512586	1.1253 ng/ml

SC

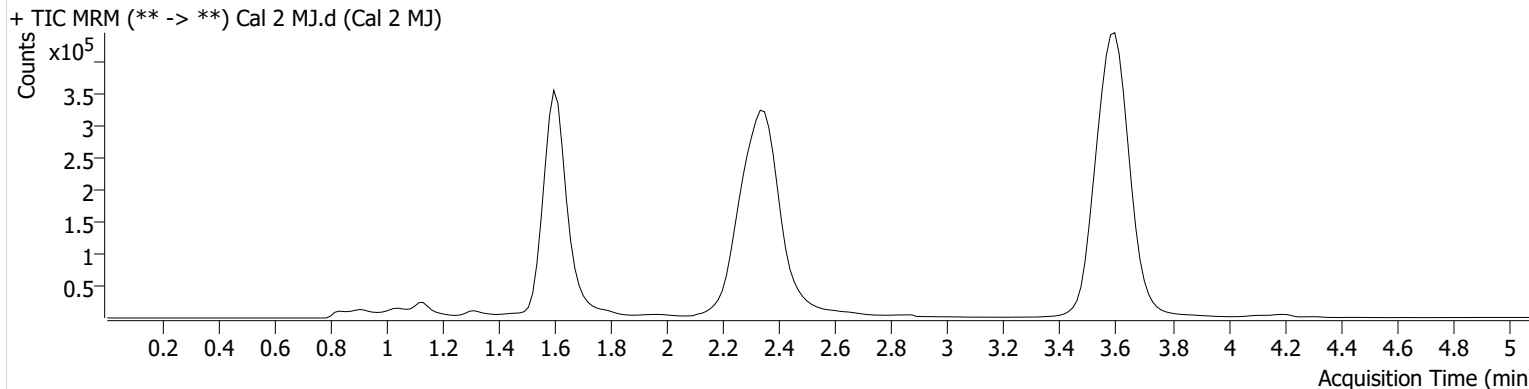


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/6/2022 1:15:18 PM

Instrument	Falco (069901)	Data File	Cal 2 MJ.d
Type	Cal	Sample	Cal 2 MJ
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P5-G6	Comment	
Injection Volume	10		
Acq. Date-Time	10/5/2022 12:51:59 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.618	86825	19.40	11.3	249.71	1537669	3.1330 ng/ml
THC-COOH	1.640	93888	∞	44.4	232.09	265686	10.5609 ng/ml
THC	3.616	94379	828.60	30.7	39.31	3603092	2.9331 ng/ml

SC

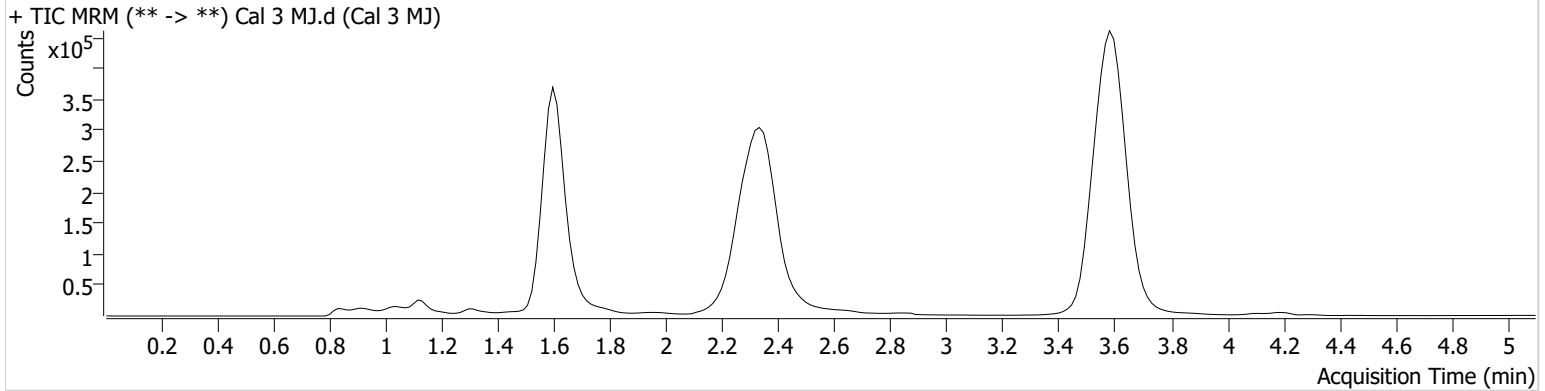


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/6/2022 1:15:18 PM

Instrument	Falco (069901)	Data File	Cal 3 MJ.d
Type	Cal	Sample	Cal 3 MJ
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P5-F6	Comment	
Injection Volume	10		
Acq. Date-Time	10/5/2022 12:59:36 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	127053	∞	11.9	394.18	1455562	5.0613 ng/ml
THC-COOH	1.640	146002	∞	49.1	765.94	258392	19.2218 ng/ml
THC	3.601	155391	1252.41	29.3	82.75	3558922	4.7354 ng/ml

SC

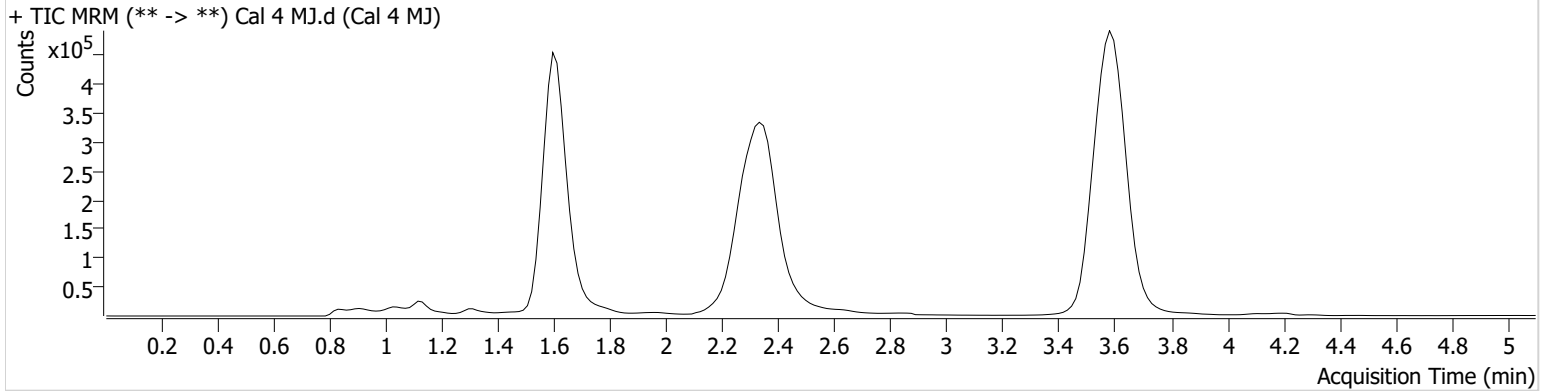


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/6/2022 1:15:18 PM

Instrument	Falco (069901)	Data File	Cal 4 MJ.d
Type	Cal	Sample	Cal 4 MJ
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P5-E6	Comment	
Injection Volume	10		
Acq. Date-Time	10/5/2022 1:07:12 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	242716	∞	13.4	∞	1517089	9.6094 ng/ml
THC-COOH	1.640	347602	310.86	54.2	2817.71	265700	49.6327 ng/ml
THC	3.601	323281	2326.77	28.2	377.30	3555327	9.6122 ng/ml

SC

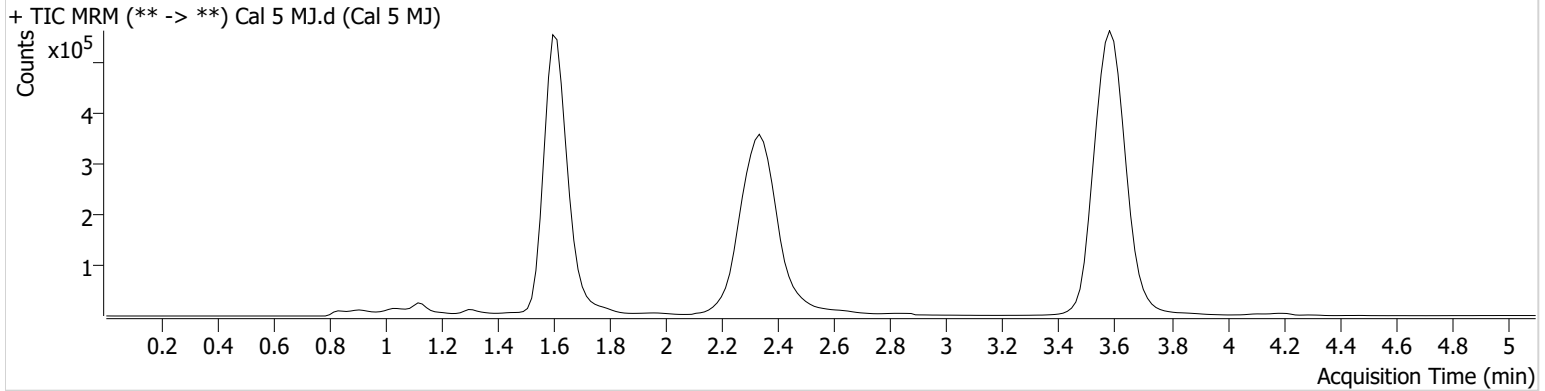


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/6/2022 1:15:18 PM

Instrument	Falco (069901)	Data File	Cal 5 MJ.d
Type	Cal	Sample	Cal 5 MJ
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P5-D6	Comment	
Injection Volume	10		
Acq. Date-Time	10/5/2022 1:14:48 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	569297	∞	13.4	∞	1449668	24.1683 ng/ml
THC-COOH	1.625	485471	1226.76	56.2	2238.31	254175	74.2549 ng/ml
THC	3.601	791347	7135.14	28.3	1120.63	3396066	24.2726 ng/ml

SC

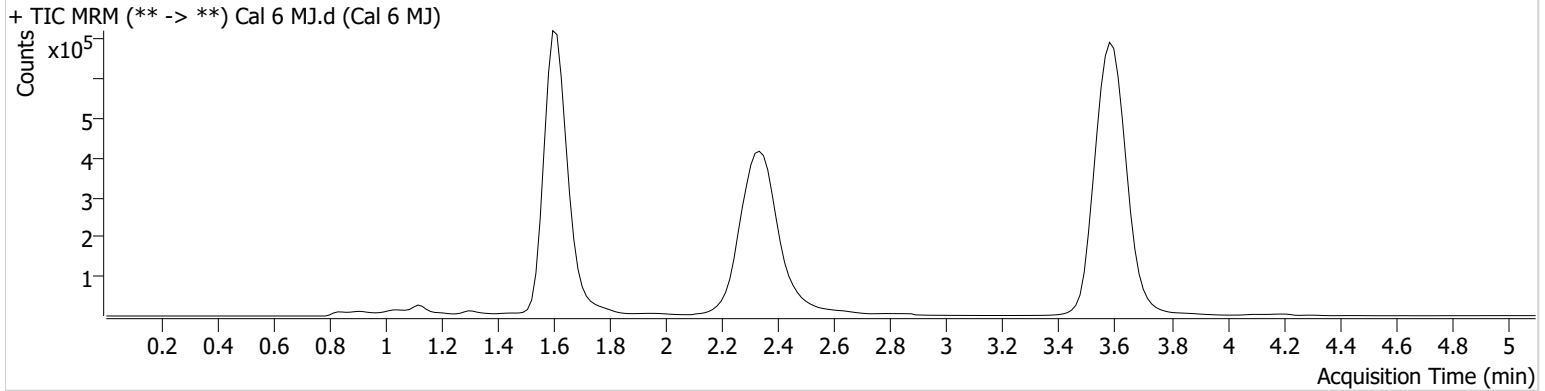


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/6/2022 1:15:18 PM

Instrument	Falco (069901)	Data File	Cal 6 MJ.d
Type	Cal	Sample	Cal 6 MJ
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P5-C6	Comment	
Injection Volume	10		
Acq. Date-Time	10/5/2022 1:22:24 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	1191613	∞	13.1	3436.99	1463255	50.5465 ng/ml
THC-COOH	1.625	643175	∞	57.0	3110.63	255440	99.1303 ng/ml
THC	3.601	1641942	∞	27.8	∞	3373008	50.4558 ng/ml

SC

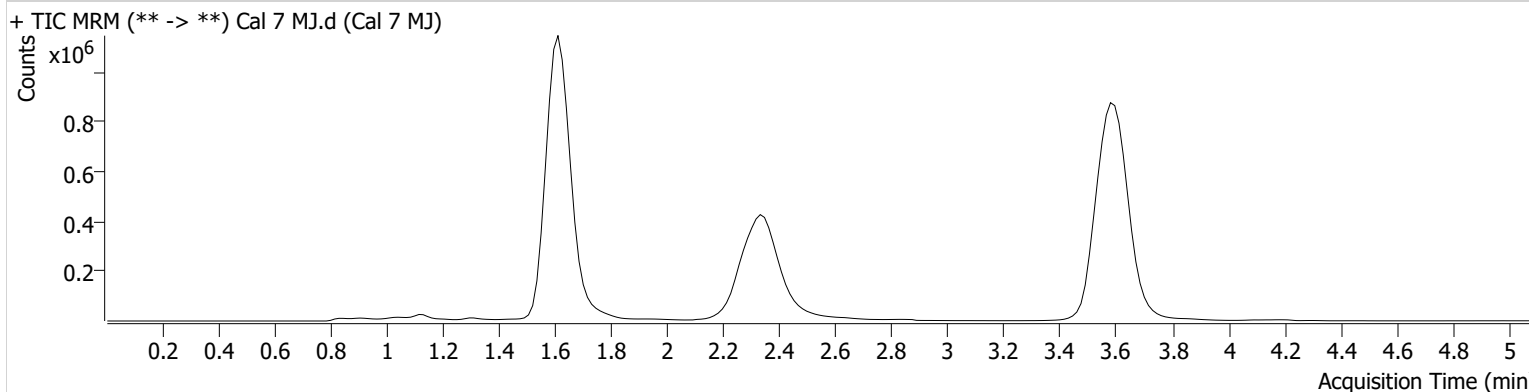


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2022\AM 27-28\100522 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/6/2022 1:15:18 PM

Instrument	Falco (069901)	Data File	Cal 7 MJ.d
Type	Cal	Sample	Cal 7 MJ
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P5-B6	Comment	
Injection Volume	10		
Acq. Date-Time	10/5/2022 1:30:00 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	2304286	∞	13.3	4206.49	1428963	100.4816 ng/ml
THC-COOH	1.640	1513290	∞	58.5	26848.7	241789	252.1993 ng/ml
THC	3.601	3072568	22070.51	28.1	∞	3150169	100.8656 ng/ml