

Worklist: 6224

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>	
M2022-4665	2	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
M2022-5034	3	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
M2022-5416	2	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-3604	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-3611	2	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
P2022-3614	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2022-3810	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
P2023-0040	2	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-0060	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-0064	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-0089	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-0090	1	BCK	AM 27 Blood THC Quant by LC-QQQ	
P2023-0110	2	BCK	AM 27 Blood THC Quant by LC-QQQ	

SC

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

Extraction Date: 01/23/2023
Plate lot#: IDP-108-3-220802
Mobile phase A: 0.1% Formic Acid in LCMS Water
Blank Blood Lot: Lampire 23A52593
Column: UCT Selectra DA 100 x 2.1mm 3um
LCMS-QQQ ID: 069901

Analyst: Sarah Collins
Retest Date: 02/02/2023
Mobile phase B: 0.1% Formic acid in Acetonitrile
Blank Urine Lot: POC021022

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.~~ 2/10/23 SC
- 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: THC-OH evaluated qualitative due to potential coeluding peak in case samples.

SC

	1	2	3	4	5	6
A	IS + Cal. 1	IS + QC_2 urine	p2023-0064-1	p2023-0060-1		
B	IS + Cal. 2	negative blood	p2023-0089-1	p2023-0090-1		
C	IS + Cal. 3	negative urine	p2023-0090-1*			
D	IS + Cal. 4	m2022-5416-2	p2023-0110-2			
E	IS + Cal. 5	p2022-3604-1	m2022-4665-2			
F	IS + Cal. 6	p2022-3614-1	m2022-5034-3			
G	IS + Cal. 7	p2023-0040-2	p2022-3611-2			
H	IS + QC_1 blood	p2023-0060-1*	p2022-3810-1			

All wells to contain 100 µl of residual DMSO

*Samples moved during analytical step 6 due to blood clot

SC

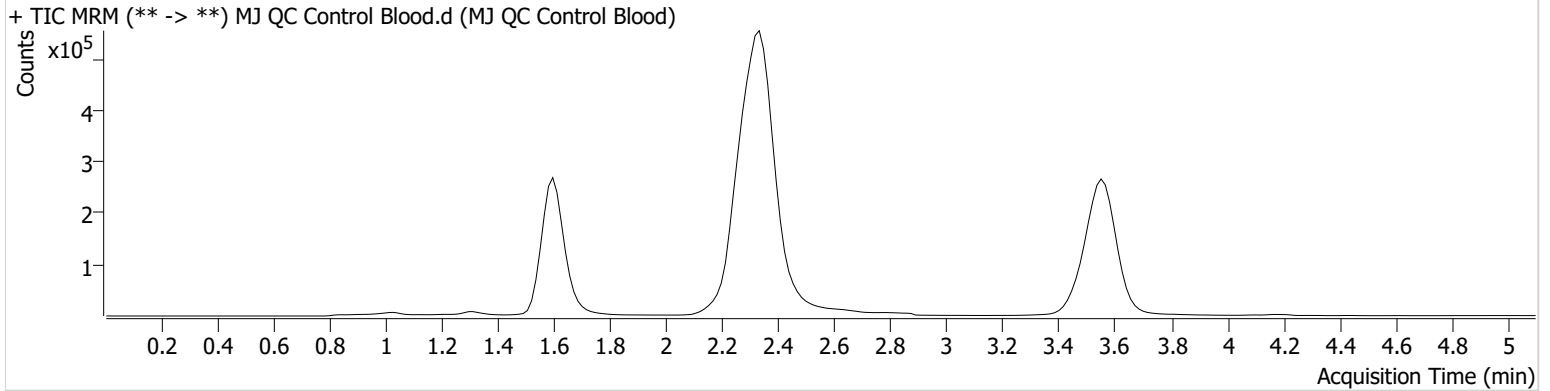


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 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	P1-H1	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 2:02:41 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	71324	∞	13.2	106.38	980343	4.7835 ng/ml
THC-COOH	1.629	22586	364.36	235.7	∞	288836	13.1963 ng/ml
THC	3.570	90596	832.19	24.3	120.66	2008988	4.8680 ng/ml

SC

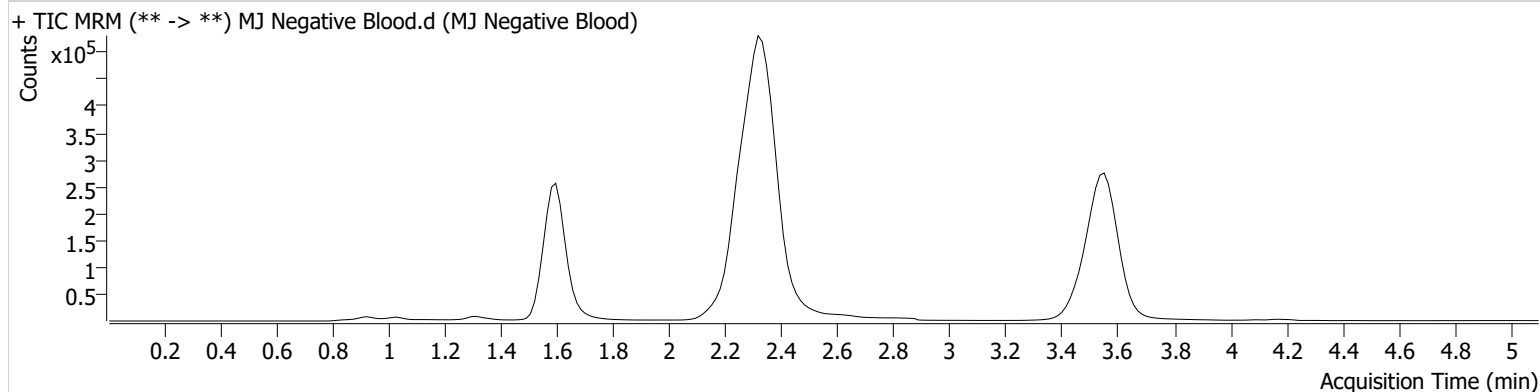


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 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	P1-B2	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 2:17:55 PM		
Sample Info.			

Sample Chromatogram



SC

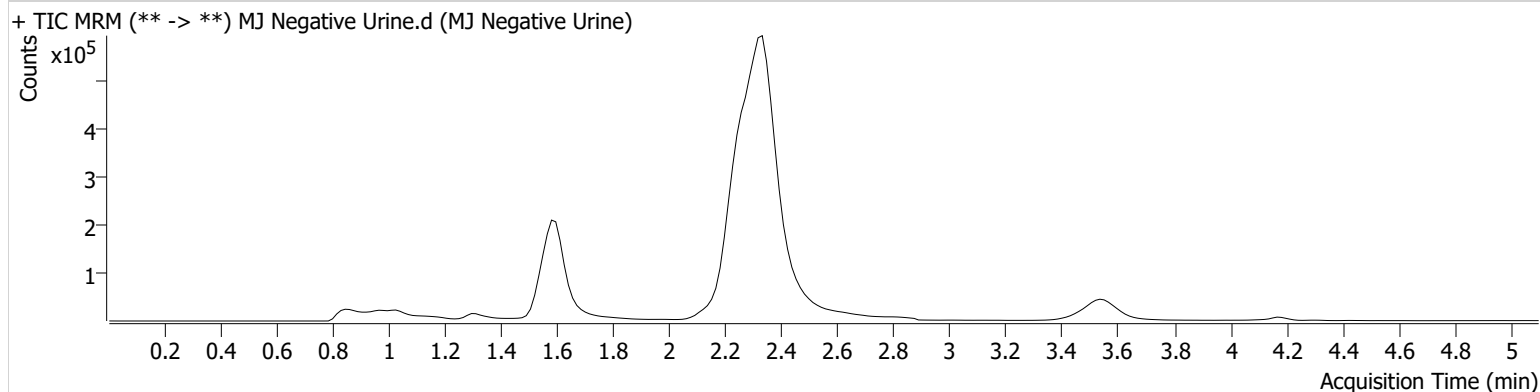


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 PM

Instrument	Falco (069901)	Data File	MJ Negative Urine.d
Type	Sample	Sample	MJ Negative Urine
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-C2	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 4:50:06 PM		
Sample Info.			

Sample Chromatogram



SC

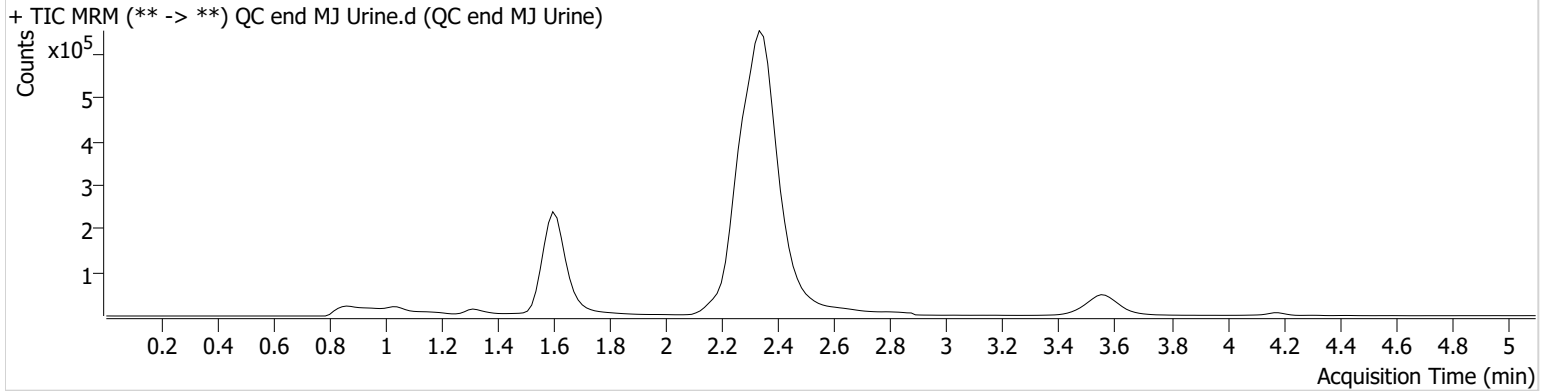


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 PM

Instrument	Falco (069901)	Data File	QC end MJ Urine.d
Type	QC	Sample	QC end MJ Urine
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-A2	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 6:06:14 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	75893	∞	12.0	165.84	988007	5.0400 ng/ml
THC-COOH	1.629	16326	∞	244.2	3120.43	208090	13.2401 ng/ml
THC	3.570	18446	∞	26.2	15.76	388424	5.1171 ng/ml

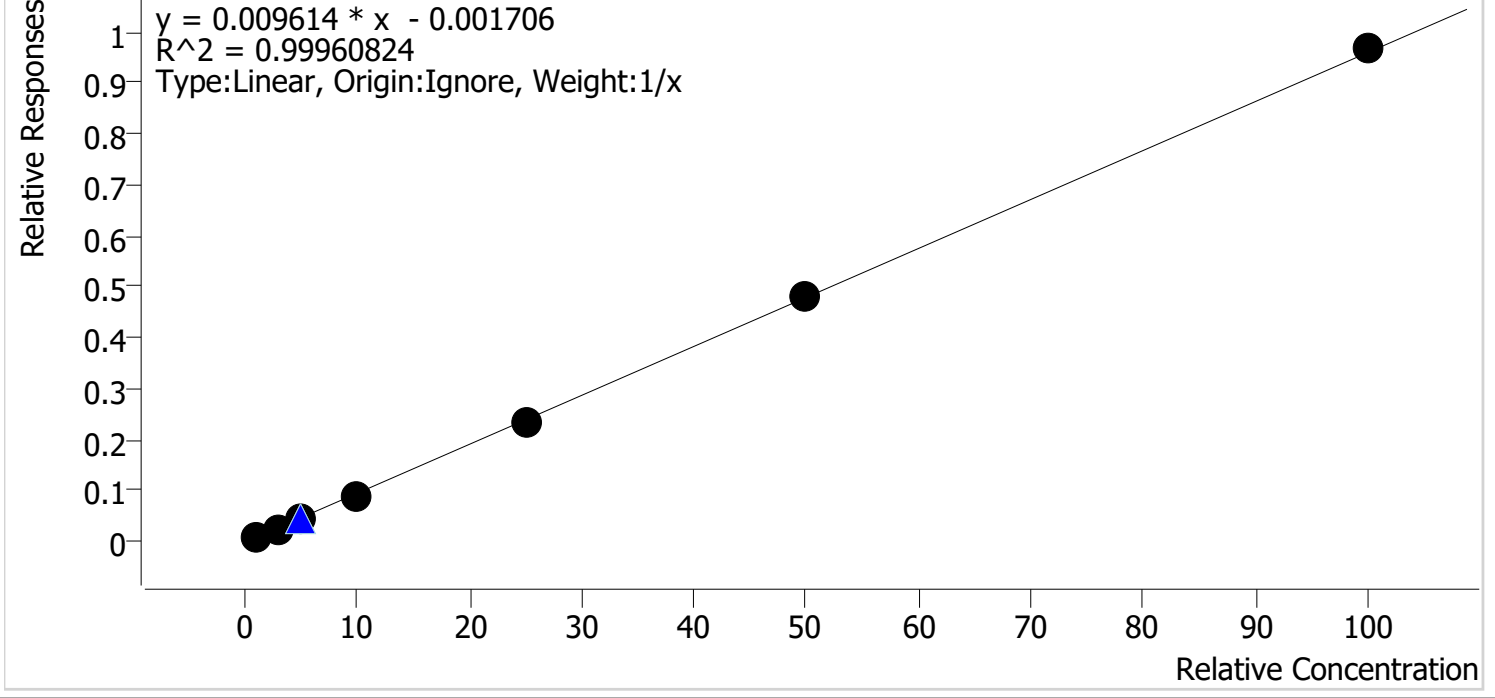
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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update 1/24/2023 12:21 PM
Analyst Name ISP\Datastor
Analyte THC **Internal Standard** THC-D3

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



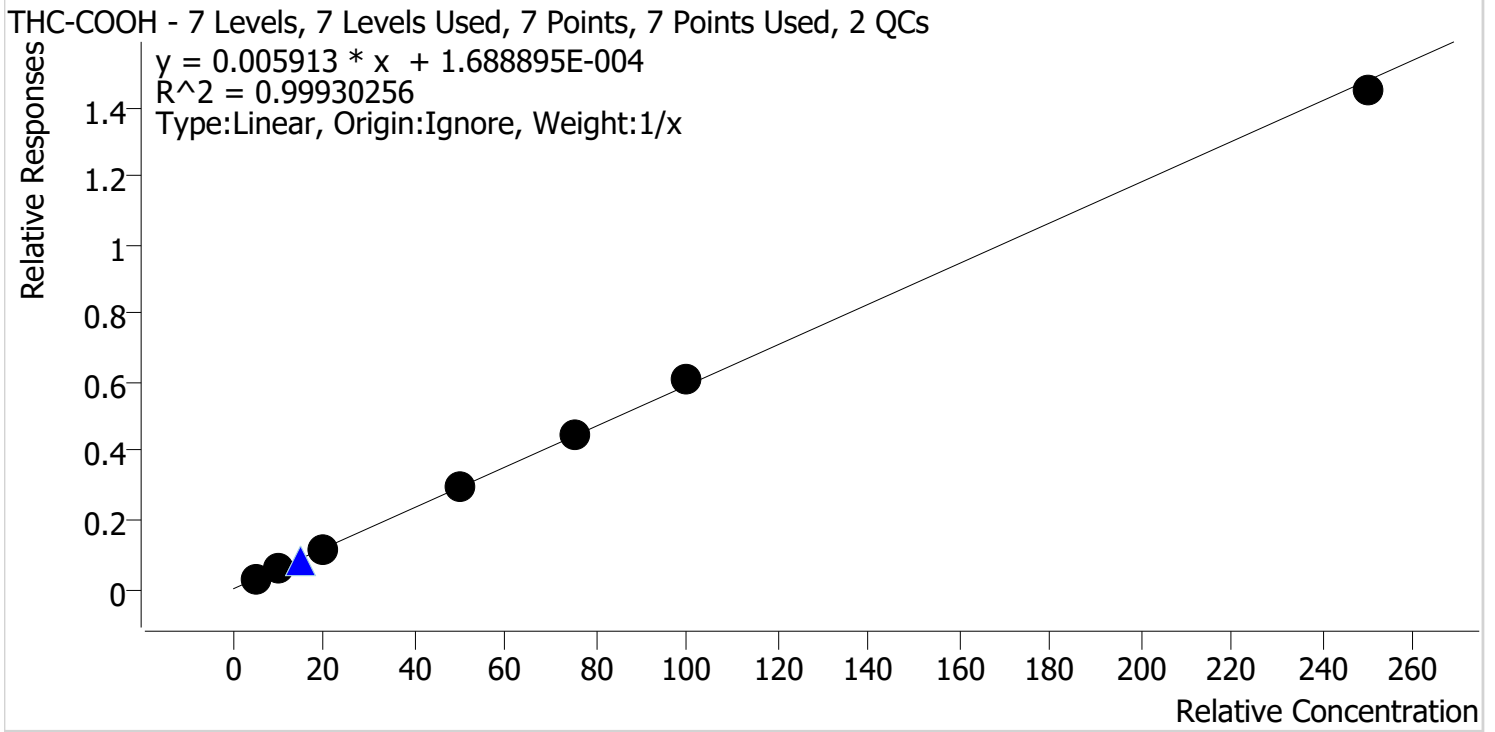
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	✓	1.0	1.1	113.3
Cal 2 MJ	2	✓	3.0	2.9	95.5
Cal 3 MJ	3	✓	5.0	4.8	95.7
Cal 4 MJ	4	✓	10.0	9.6	95.8
Cal 5 MJ	5	✓	25.0	24.6	98.4
Cal 6 MJ	6	✓	50.0	50.3	100.5
Cal 7 MJ	7	✓	100.0	100.8	100.8

SC



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update 1/24/2023 12:21 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	✓	5.0	5.0	99.2
Cal 2 MJ	2	✓	10.0	9.7	97.1
Cal 3 MJ	3	✓	20.0	19.8	98.9
Cal 4 MJ	4	✓	50.0	50.7	101.4
Cal 5 MJ	5	✓	75.0	76.6	102.1
Cal 6 MJ	6	✓	100.0	103.4	103.4
Cal 7 MJ	7	✓	250.0	244.9	98.0

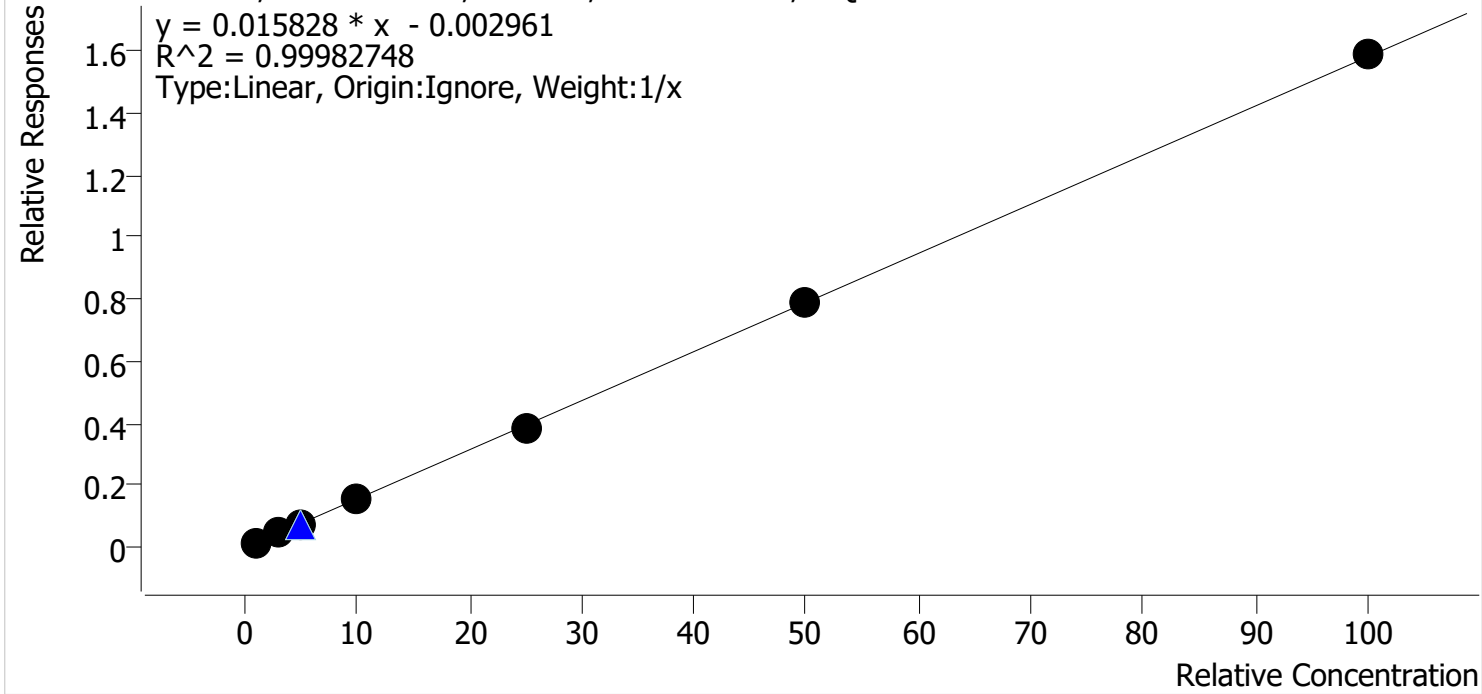
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AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update 1/24/2023 12:21 PM
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



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	✓	1.0	1.1	108.4
Cal 2 MJ	2	✓	3.0	2.9	97.2
Cal 3 MJ	3	✓	5.0	4.8	96.6
Cal 4 MJ	4	✓	10.0	9.8	98.4
Cal 5 MJ	5	✓	25.0	24.6	98.4
Cal 6 MJ	6	✓	50.0	50.3	100.6
Cal 7 MJ	7	✓	100.0	100.4	100.4

SC

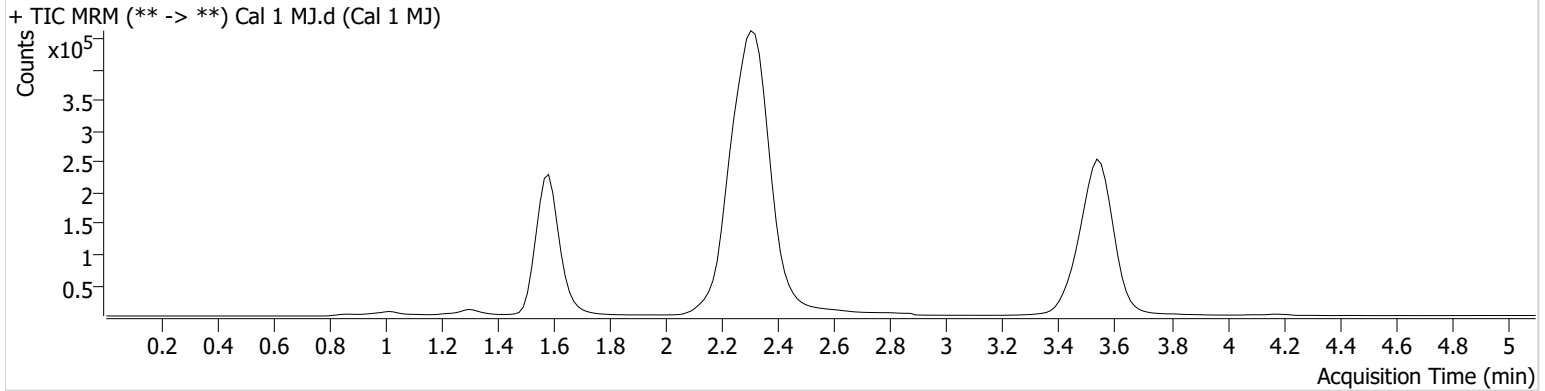


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 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	P1-A1	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 1:01:42 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.588	13853	∞	13.5	19.85	975217	1.0845 ng/ml Low
THC-COOH	1.614	8036	177.57	230.4	∞	272303	4.9621 ng/ml Low
THC	3.555	19572	∞	25.9	∞	2129714	1.1334 ng/ml

SC

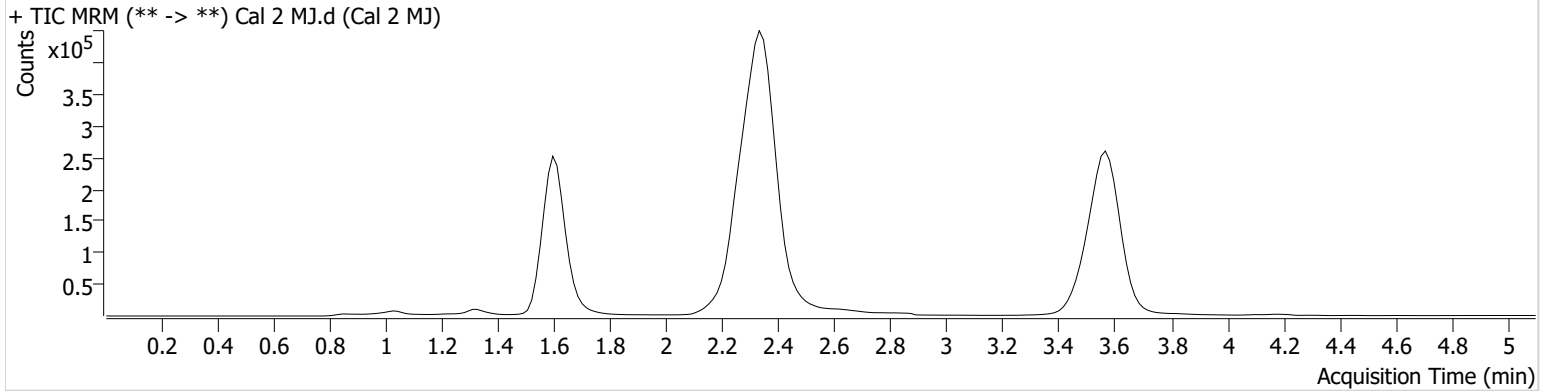


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 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	P1-B1	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 1:09:28 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	42713	∞	13.4	83.11	989247	2.9149 ng/ml Low
THC-COOH	1.629	15941	253.11	230.2	526.68	276975	9.7050 ng/ml
THC	3.586	53147	469.01	24.1	51.40	2057239	2.8646 ng/ml

SC

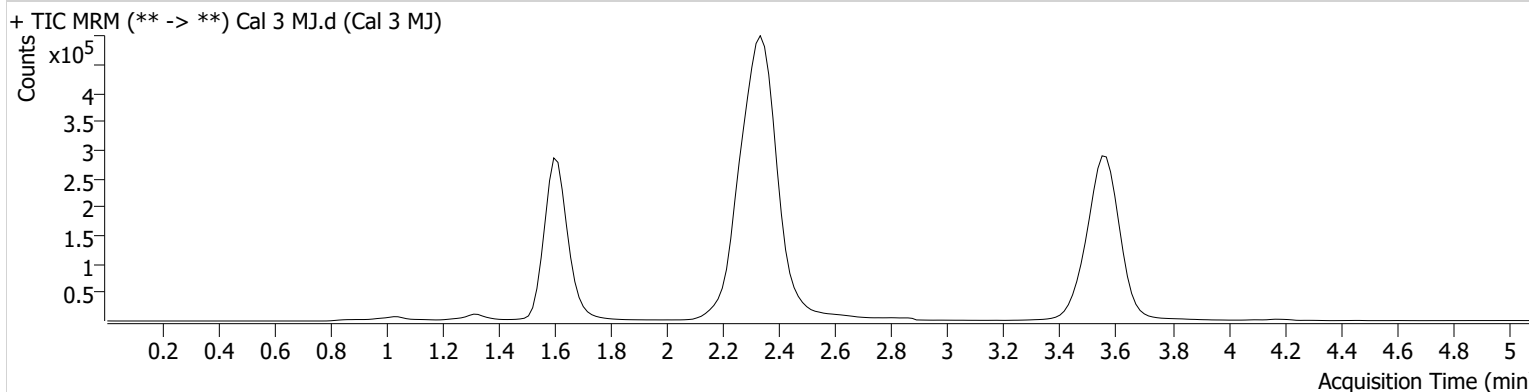


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 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	P1-C1	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 1:17:04 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	75145	897.93	13.1	175.62	1023046	4.8275 ng/ml
THC-COOH	1.629	33771	662.49	235.8	∞	288384	19.7764 ng/ml
THC	3.570	100241	∞	23.6	98.80	2263979	4.7829 ng/ml

SC

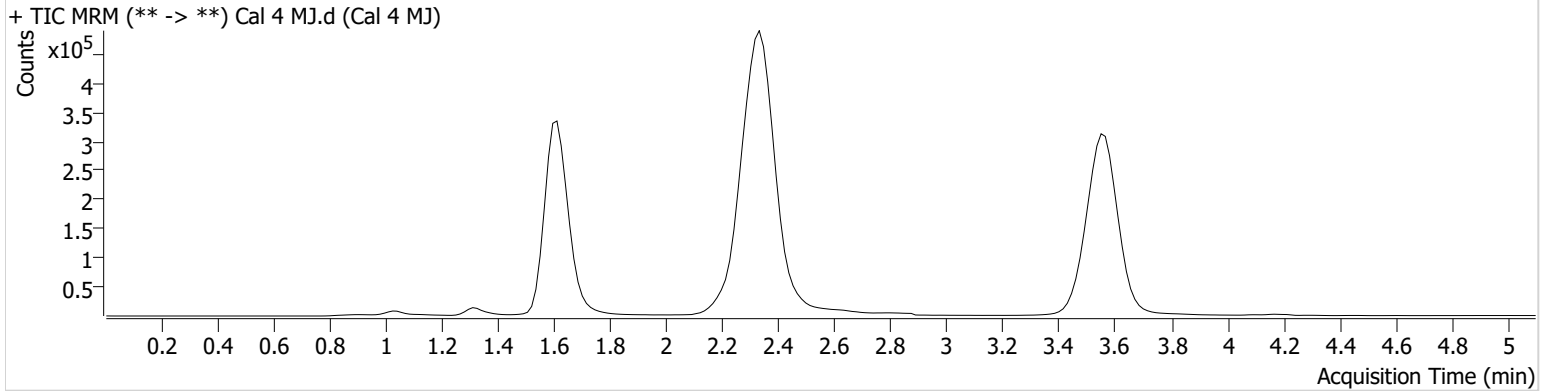


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 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	P1-D1	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 1:24:40 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	146793	∞	13.2	466.91	960525	9.8422 ng/ml
THC-COOH	1.629	79192	1973.31	234.4	4432.86	263979	50.7067 ng/ml
THC	3.570	199522	1147.49	23.6	810.78	2206431	9.5833 ng/ml

SC

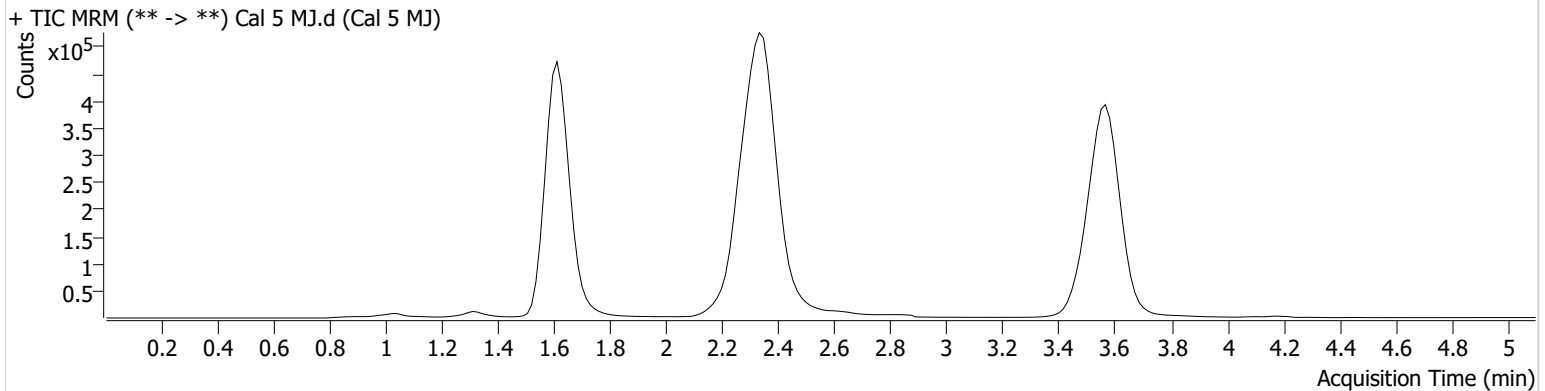


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 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	P1-E1	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 1:32:15 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	425168	756.04	13.2	864.57	1100717	24.5903 ng/ml
THC-COOH	1.629	134899	1490.14	231.1	4137.55	297894	76.5565 ng/ml
THC	3.586	580679	∞	23.0	∞	2473825	24.5927 ng/ml

SC

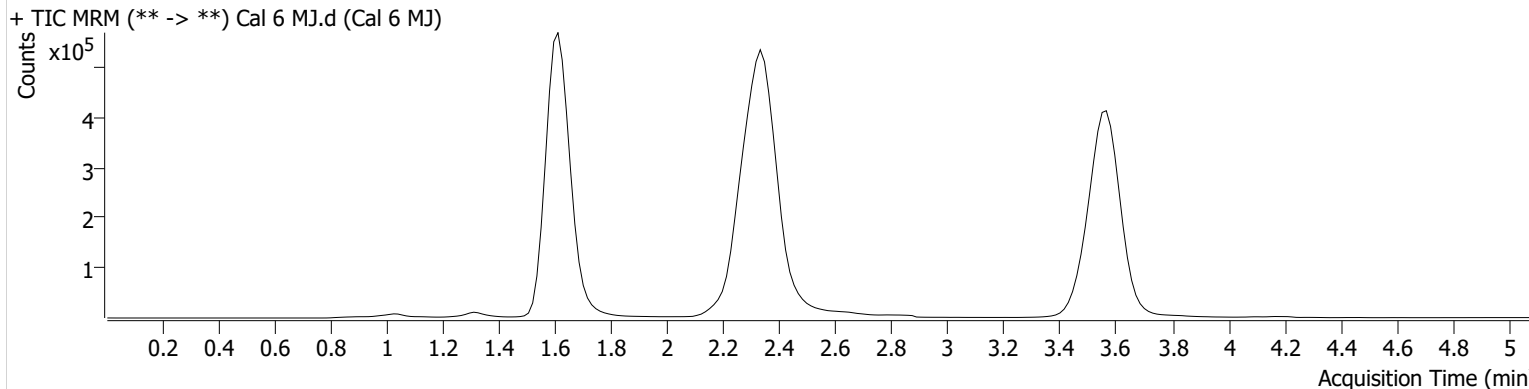


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 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	P1-F1	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 1:39:50 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	826296	∞	13.2	3805.16	1041343	50.3177 ng/ml
THC-COOH	1.629	167818	430.47	228.6	4073.72	274502	103.3647 ng/ml
THC	3.570	1010310	6282.10	22.3	1088.09	2097787	50.2717 ng/ml

SC

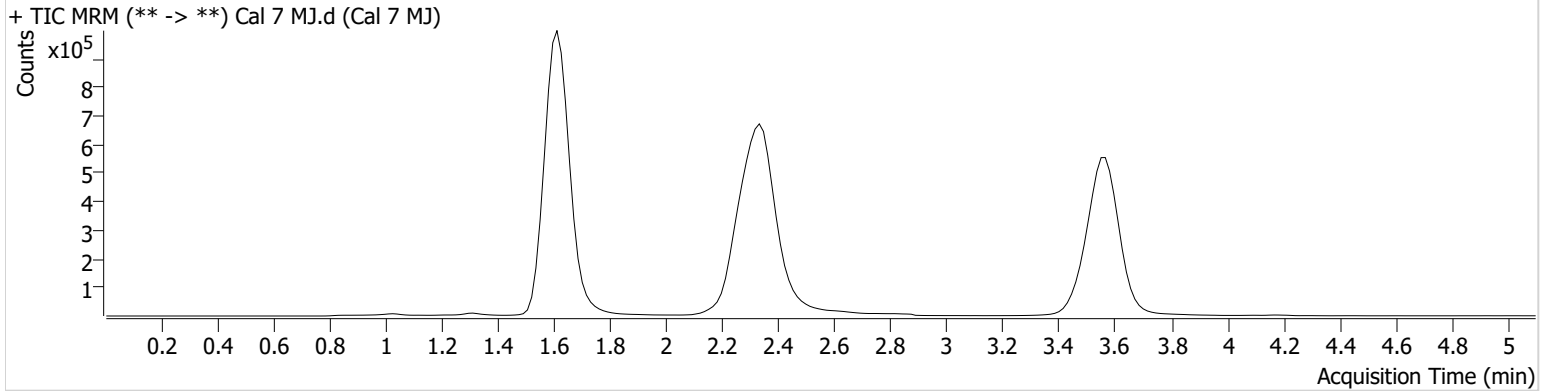


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2023\AM 27 28\012323 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 1/24/2023 12:21:28 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	P1-G1	Comment	
Injection Volume	10		
Acq. Date-Time	1/23/2023 1:47:26 PM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.603	1684593	∞	13.2	3377.03	1061776	100.4230 ng/ml
THC-COOH	1.629	417711	8669.20	232.0	∞	288393	244.9285 ng/ml
THC	3.570	1961972	12189.49	23.5	4289.69	2028688	100.7714 ng/ml