

Worklist: 6891

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
M2024-2991	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2024-2146	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2024-2177	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2024-2196	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2024-2197	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2024-2210	1	UCK	AM 27 Urine Cannabinoids Confirmation by LC-QQQ
P2024-2213	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2024-2246	1	BCK	AM 27 Blood THC Quant by LC-QQQ



TS

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

Extraction Date: 08/09/2024

Plate lot#: 240513

Mobile phase A: 0.1% Formic Acid in LCMS Water

Blank Blood Lot: Lampire 24C52816

Column: UCT Selectra DA 100 x 2.1mm 3um

Analyst: Tamara Salazar

Plate Retest Date: 11/13/2024

Mobile phase B: 0.1% Formic acid in Acetonitrile

Blank Urine Lot: POC021022

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. Using a calibrated pipette, pipette 1000µL blood or 1000µL urine in wells of analytical (standards) plate. **Pipette ID: 42**
- ☒ 3. **Urine hydrolysis add 100 ul BG turbo, and 200 ul BG turbo buffer to the urine samples in wells of the analytical plate.**
- ☒ 4. Add **500µL of 0.1% formic acid in water** in the wells of the 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: **750 µL**
- ☒ 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: 067104
- ☒ 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: 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. Did all QCs pass for each analyte? (if not, describe in comments section)
- ☒ 5. Enter QCs into control charting.
- ☒ 6. Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS: Samples were initially injected on 08/09/2024, but the run showed signs of significant ion suppression. The instrument was cleaned, and the samples were reconstituted and re-injected on 08/12/2024. The reinjection data was used for evaluation.

Calibrator 2 did not inject properly with initial injection and was reinjected.

TS

	1	2	3	4	5	6
A	IS + Cal. 1	IS + QC_1			P2024-2213-1	IS + QC_1
B	IS + Cal. 2				P2024-2197-1	IS + Cal. 7
C	IS + Cal. 3				P2024-2196-1	IS + Cal. 6
D	IS + Cal. 4				P2024-2177-1	IS + Cal. 5
E	IS + Cal. 5				P2024-2146-1	IS + Cal. 4
F	IS + Cal. 6			P2024-2210-1	M2024-2991-2	IS + Cal. 3
G	IS + Cal. 7			Neg Urine	Neg Blood	IS + Cal. 2
H	IS + QC_1			P2024-2246-1	IS + QC_1	IS + Cal. 1

All wells to contain 100 µl of residual DMSO

TS



AM #27 Cannabinoids Quant. Results

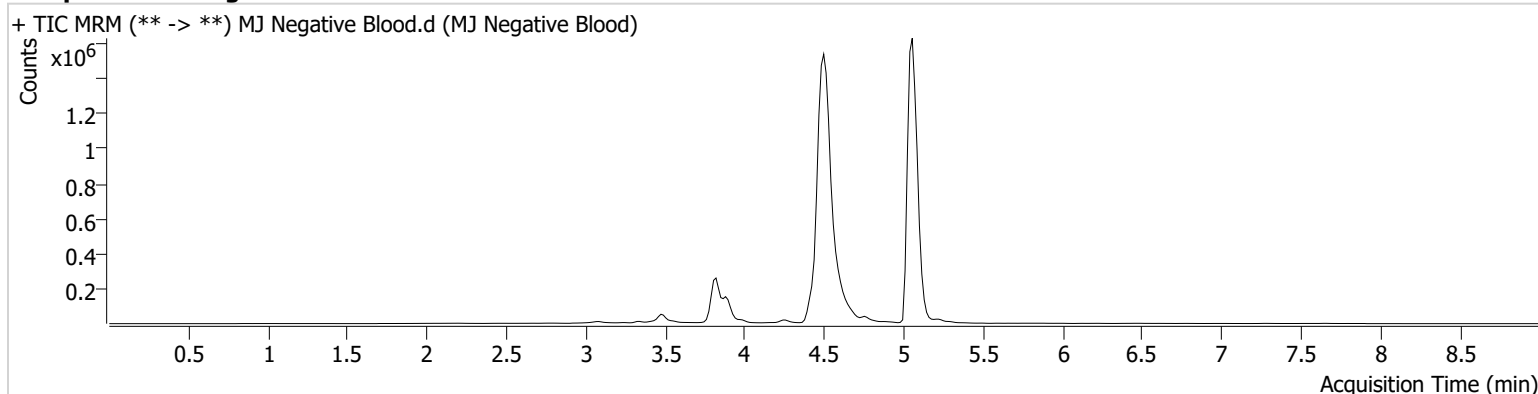
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Calibration Last Update 8/13/2024 1:19:15 PM

Instrument Falco (069901)
Type Sample
Acq. Method AM 27 Agilent Method.m
Sample Position P5-G5
Injection Volume 10
Acq. Date-Time 8/12/2024 12:40:25 PM
Sample Info.

Data File MJ Negative Blood.d
Sample MJ Negative Blood
Operator Tamara Salazar
Comment

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Sample Chromatogram



TS



AM #27 Cannabinoids Quant. Results

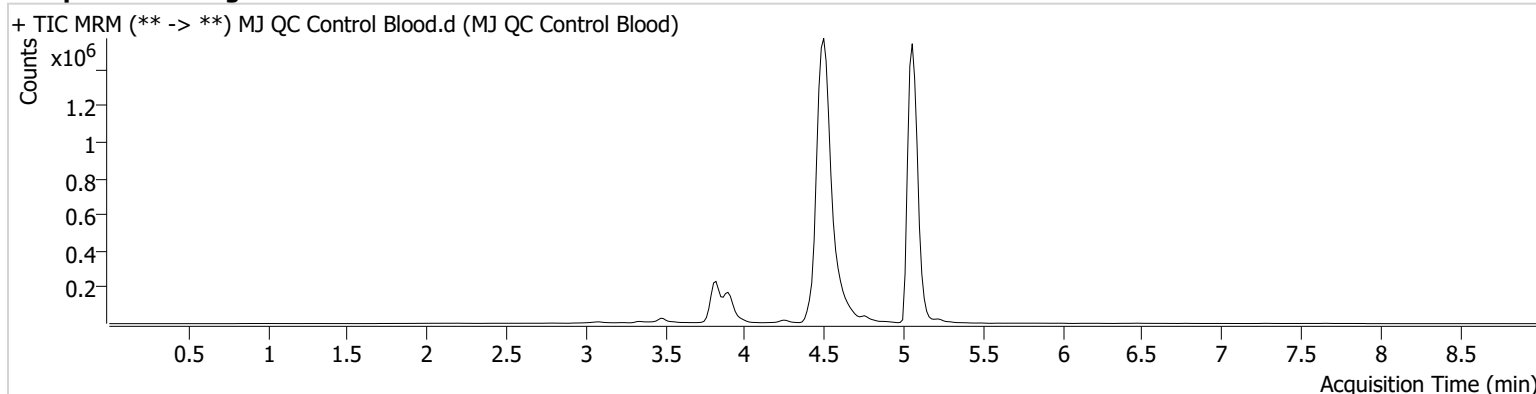
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Calibration Last Update 8/13/2024 1:19:15 PM

Instrument Falco (069901)
Type QC
Acq. Method AM 27 Agilent Method.m
Sample Position P5-A6
Injection Volume 10
Acq. Date-Time 8/12/2024 12:14:12 PM
Sample Info.

Data File MJ QC Control Blood.d
Sample MJ QC Control Blood
Operator Tamara Salazar
Comment

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Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.060	273261	1411.41	25.7	∞	5965467	4.8285 ng/ml
THC-COOH	3.909	36427	∞	228.3	876.05	404707	15.4599 ng/ml
THC-OH	3.820	63691	∞	14.6	∞	893357	4.7493 ng/ml

TS



AM #27 Cannabinoids Quant. Results

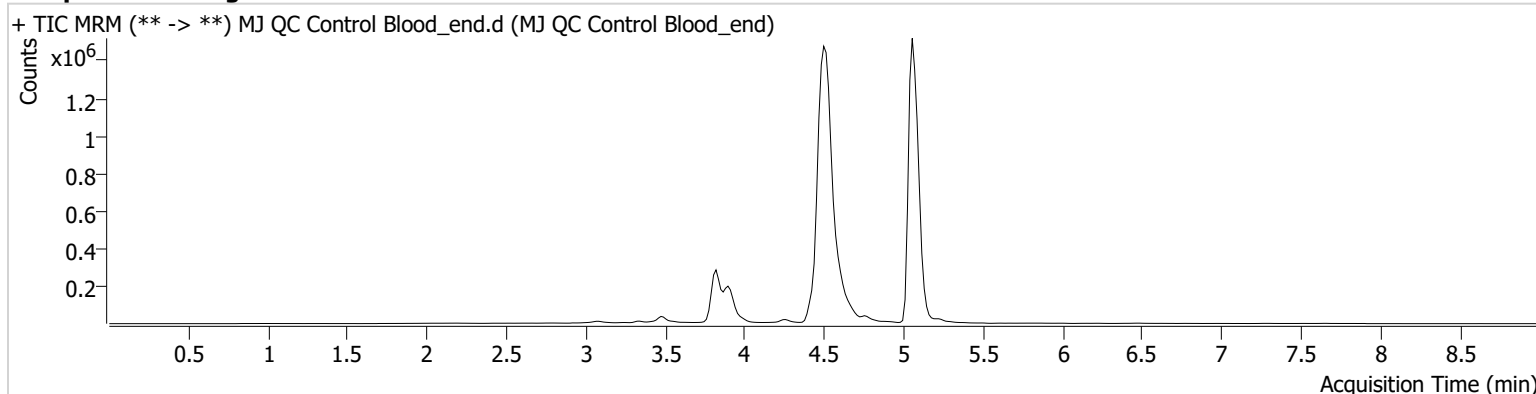
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Calibration Last Update 8/13/2024 1:19:15 PM

Instrument Falco (069901)
Type QC
Acq. Method AM 27 Agilent Method.m
Sample Position P5-A6
Injection Volume 10
Acq. Date-Time 8/12/2024 5:28:48 PM
Sample Info.

Data File MJ QC Control Blood_end.d
Sample MJ QC Control Blood_end
Operator Tamara Salazar
Comment

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Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.075	281073	1925.69	26.0	∞	5872598	5.0329 ng/ml
THC-COOH	3.909	40565	∞	237.9	∞	462277	15.1016 ng/ml
THC-OH	3.820	79028	∞	13.2	∞	1062238	4.9420 ng/ml

TS



AM #27 Cannabinoids Quant. Results

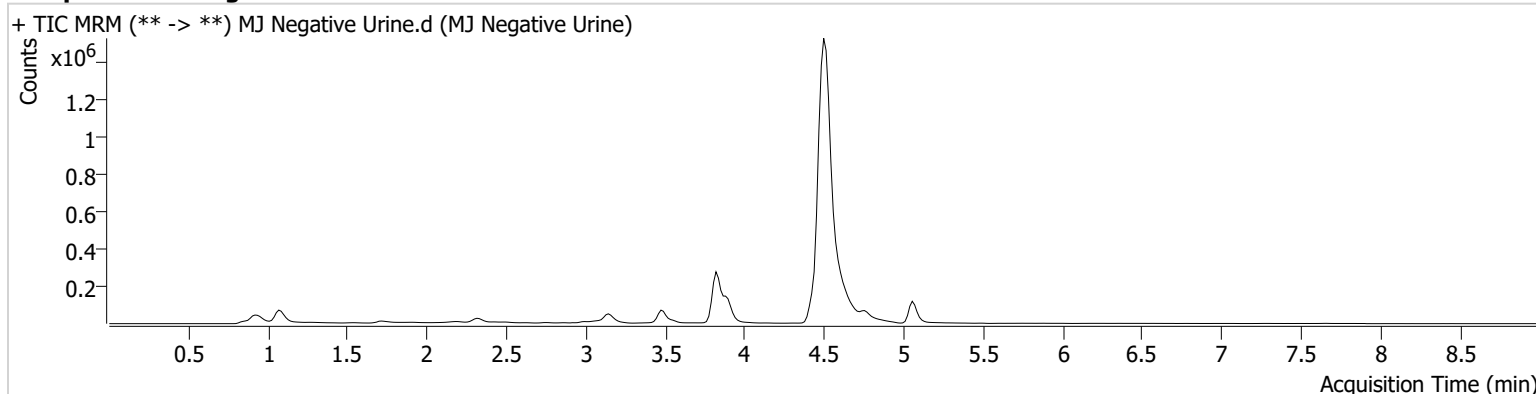
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Calibration Last Update 8/13/2024 1:19:15 PM

Instrument Falco (069901)
Type Sample
Acq. Method AM 27 Agilent Method.m
Sample Position P5-G4
Injection Volume 10
Acq. Date-Time 8/12/2024 4:10:10 PM
Sample Info.

Data File MJ Negative Urine.d
Sample MJ Negative Urine
Operator Tamara Salazar
Comment

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Sample Chromatogram



TS



AM #27 Cannabinoids Quant. Results

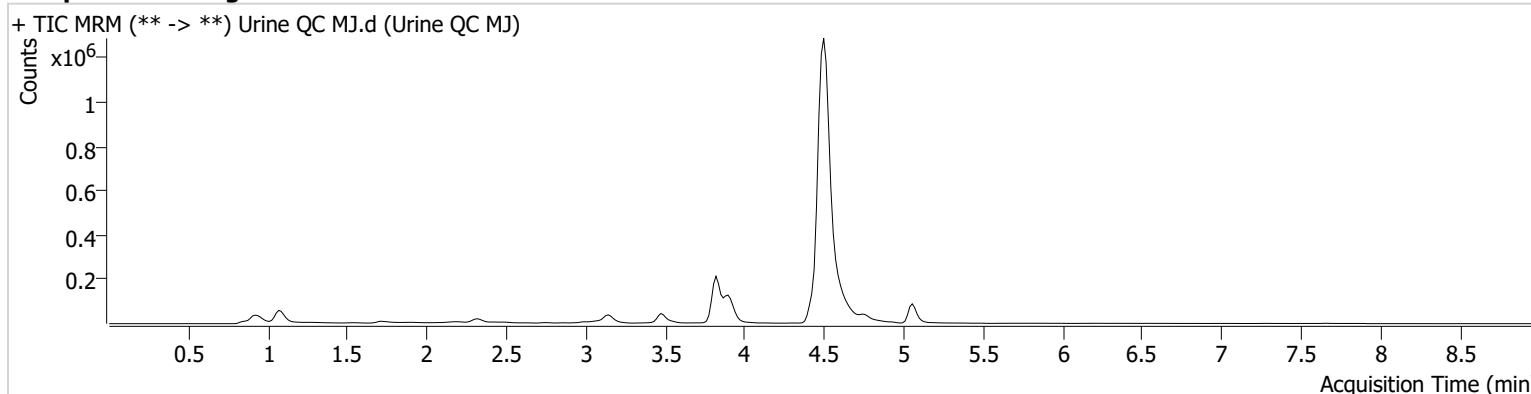
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Calibration Last Update 8/13/2024 1:19:15 PM

Instrument Falco (069901)
Type QC
Acq. Method AM 27 Agilent Method.m
Sample Position P5-H5
Injection Volume 10
Acq. Date-Time 8/12/2024 5:02:35 PM
Sample Info.

Data File Urine QC MJ.d
Sample Urine QC MJ
Operator Tamara Salazar
Comment

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Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.060	14171	264.15	31.5	21.69	298304	4.9975 ng/ml
THC-COOH	3.909	26052	∞	232.3	1021.15	316138	14.2538 ng/ml
THC-OH	3.820	57691	∞	13.1	50.27	745499	5.1276 ng/ml

TS



AM #27 Cannabinoids Quant. Calibration Curve Report

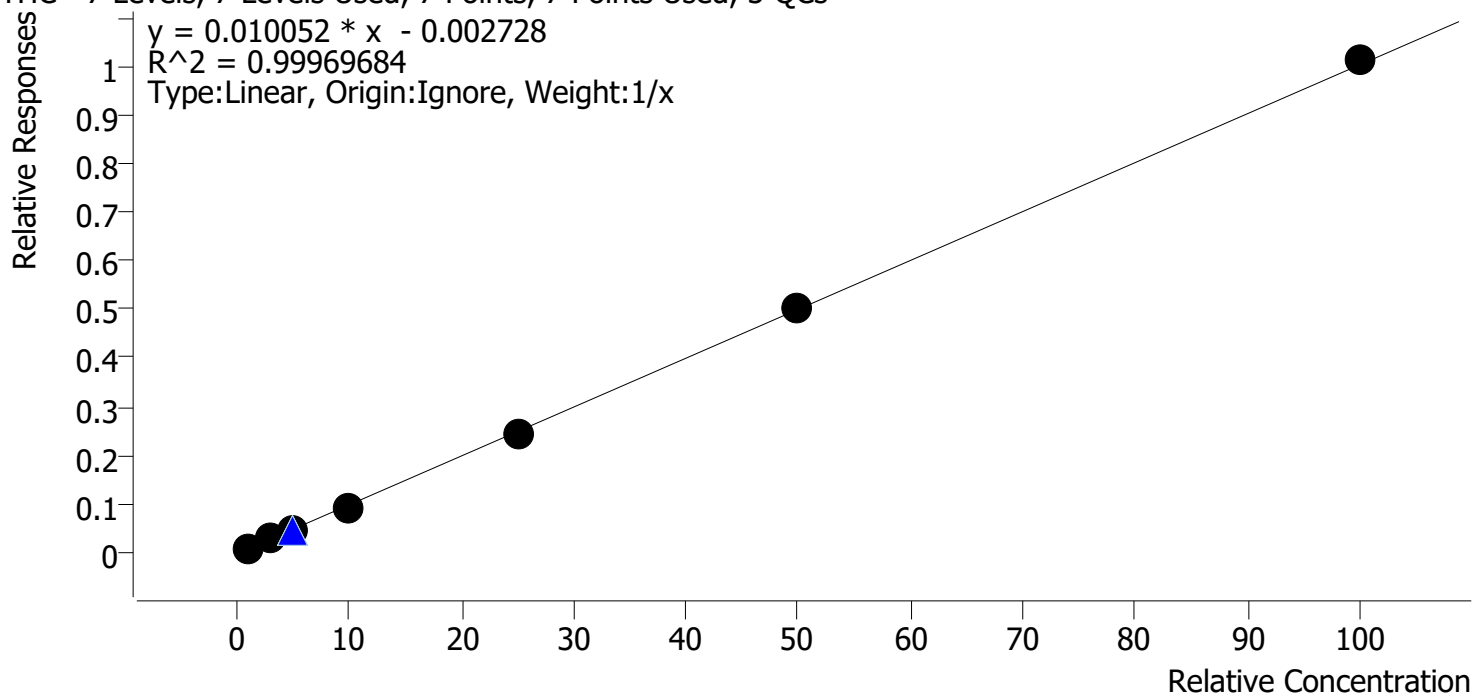
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error\QuantResults\AM 27.batch.bin

Last Cal. Update 8/13/2024 1:19 PM

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
Cal 1 MJ	1	✓	1.0	1.1	110.2
Cal 2 MJ_r	2	✓	3.0	3.0	98.6
Cal 3 MJ	3	✓	5.0	4.8	95.7
Cal 4 MJ	4	✓	10.0	9.6	95.5
Cal 5 MJ	5	✓	25.0	24.8	99.2
Cal 6 MJ	6	✓	50.0	49.9	99.8
Cal 7 MJ	7	✓	100.0	100.9	100.9

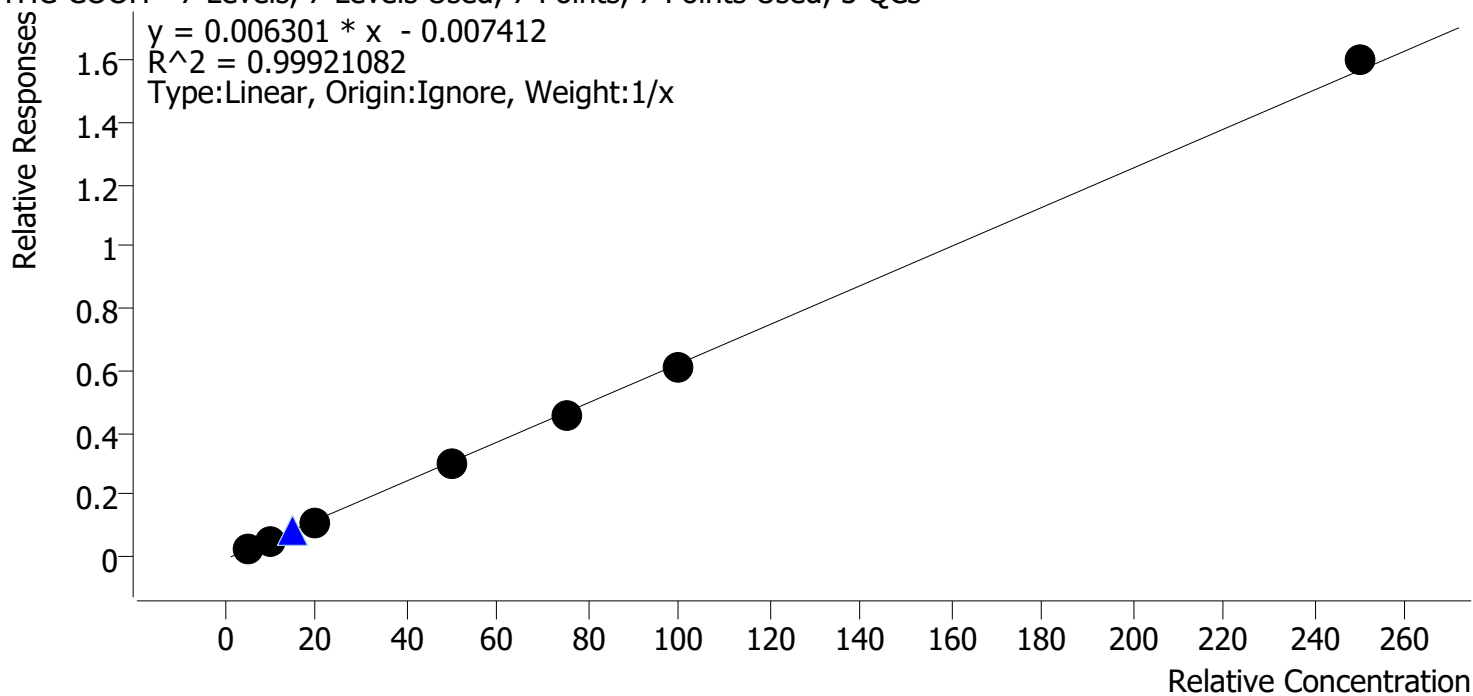
TS



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2024\AM 27 28\080924 AM 27 28 TS\AM 28 reinject after QQQ
 error\QuantResults\AM 27.batch.bin
Last Cal. Update 8/13/2024 1:19 PM
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
Cal 1 MJ	1	✓	5.0	5.5	110.5
Cal 2 MJ_r	2	✓	10.0	9.8	98.3
Cal 3 MJ	3	✓	20.0	19.0	95.0
Cal 4 MJ	4	✓	50.0	48.9	97.9
Cal 5 MJ	5	✓	75.0	73.9	98.5
Cal 6 MJ	6	✓	100.0	97.7	97.7
Cal 7 MJ	7	✓	250.0	255.1	102.1

TS



AM #27 Cannabinoids Quant. Calibration Curve Report

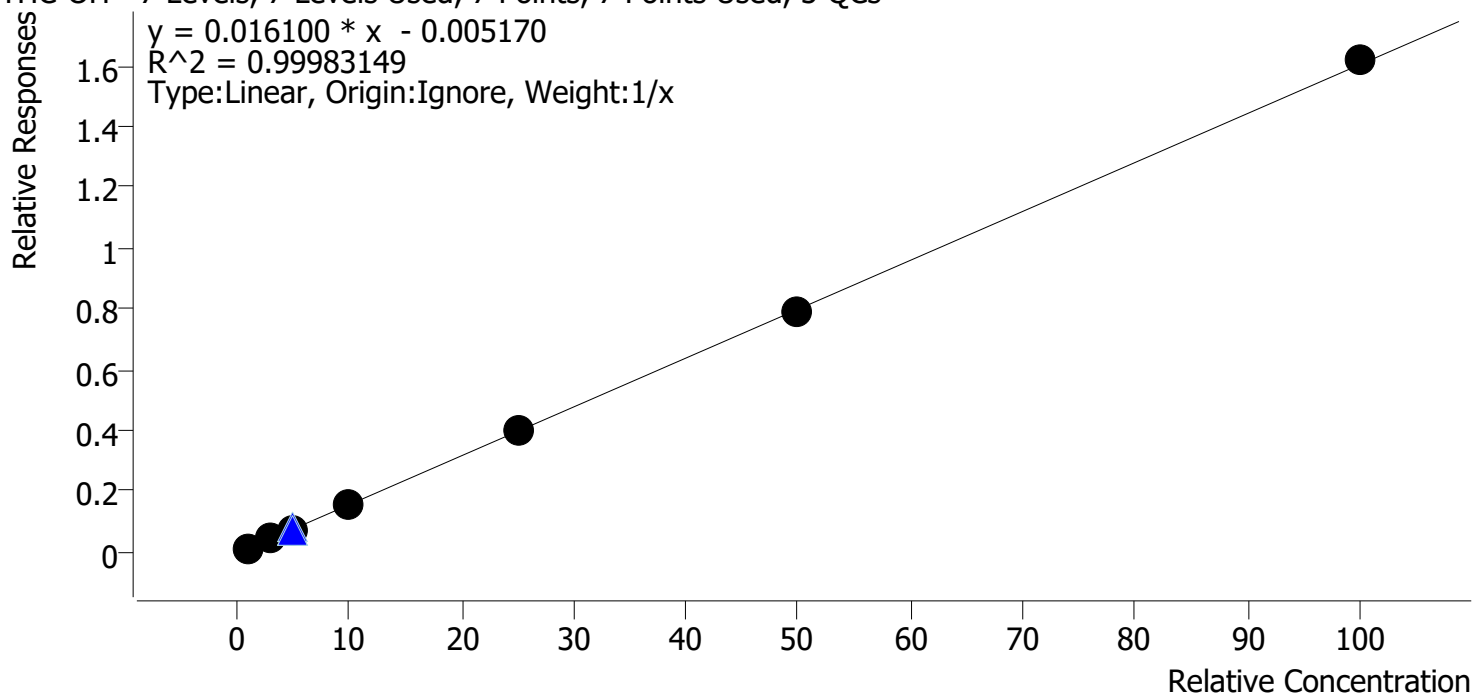
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error\QuantResults\AM 27.batch.bin

Last Cal. Update 8/13/2024 1:19 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, 3 QCs



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1 MJ	1	✓	1.0	1.1	107.6
Cal 2 MJ_r	2	✓	3.0	2.9	96.4
Cal 3 MJ	3	✓	5.0	4.9	97.1
Cal 4 MJ	4	✓	10.0	9.9	99.4
Cal 5 MJ	5	✓	25.0	24.9	99.7
Cal 6 MJ	6	✓	50.0	49.5	98.9
Cal 7 MJ	7	✓	100.0	100.9	100.9

TS



AM #27 Cannabinoids Quant. Results

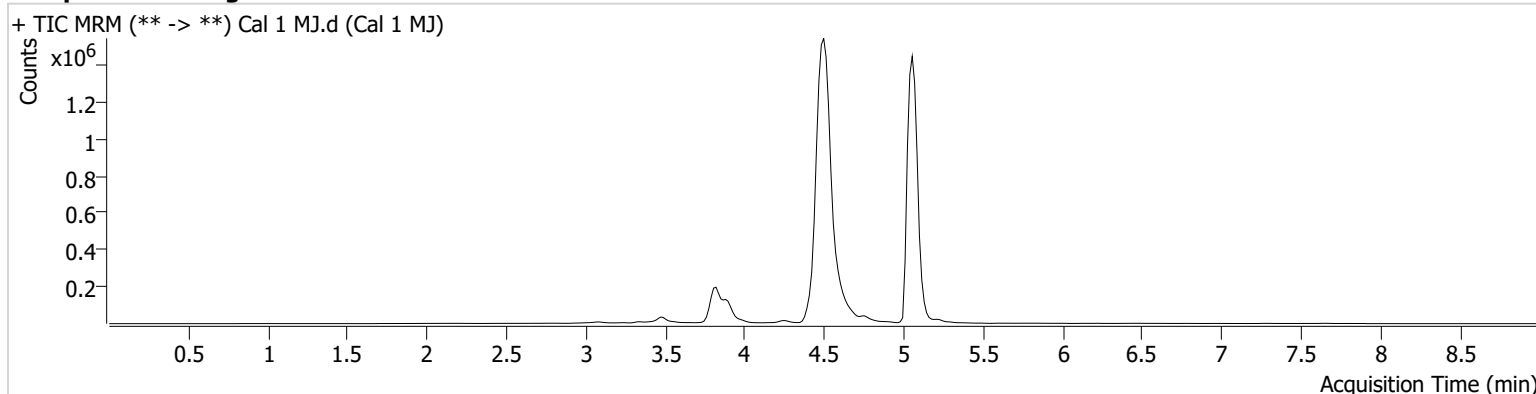
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Calibration Last Update 8/13/2024 1:19:15 PM

Instrument Falco (069901)
Type Cal
Acq. Method AM 27 Agilent Method.m
Sample Position P5-H6
Injection Volume 10
Acq. Date-Time 8/12/2024 10:15:51 AM
Sample Info.

Data File Cal 1 MJ.d
Sample Cal 1 MJ
Operator Tamara Salazar
Comment

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Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.060	50280	227.01	29.0	∞	6018581	1.1025 ng/ml
THC-COOH	3.909	10278	67.37	230.5	∞	375041	5.5254 ng/ml
THC-OH	3.820	10181	∞	15.9	45.95	837535	1.0761 ng/ml

TS



AM #27 Cannabinoids Quant. Results

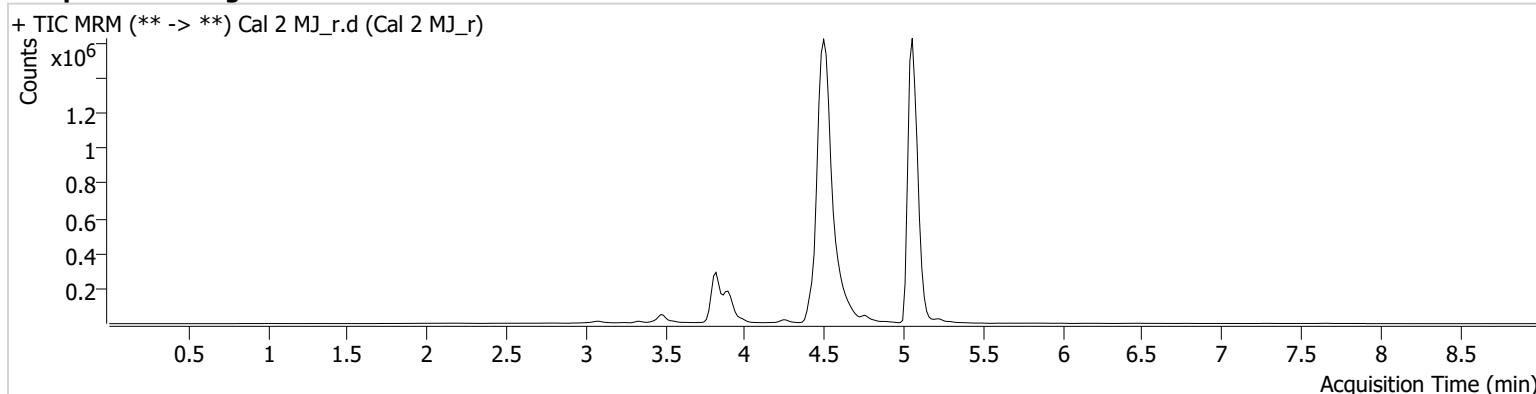
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Calibration Last Update 8/13/2024 1:19:15 PM

Instrument Falco (069901)
Type Cal
Acq. Method AM 27 Agilent Method.m
Sample Position P5-G6
Injection Volume 10
Acq. Date-Time 8/12/2024 11:48:00 AM
Sample Info.

Data File Cal 2 MJ_r.d
Sample Cal 2 MJ_r
Operator Tamara Salazar
Comment

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Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.060	170419	∞	26.7	∞	6308789	2.9588 ng/ml
THC-COOH	3.909	26039	∞	245.8	231.16	477330	9.8333 ng/ml
THC-OH	3.820	45202	∞	14.7	70.86	1092276	2.8915 ng/ml

TS



AM #27 Cannabinoids Quant. Results

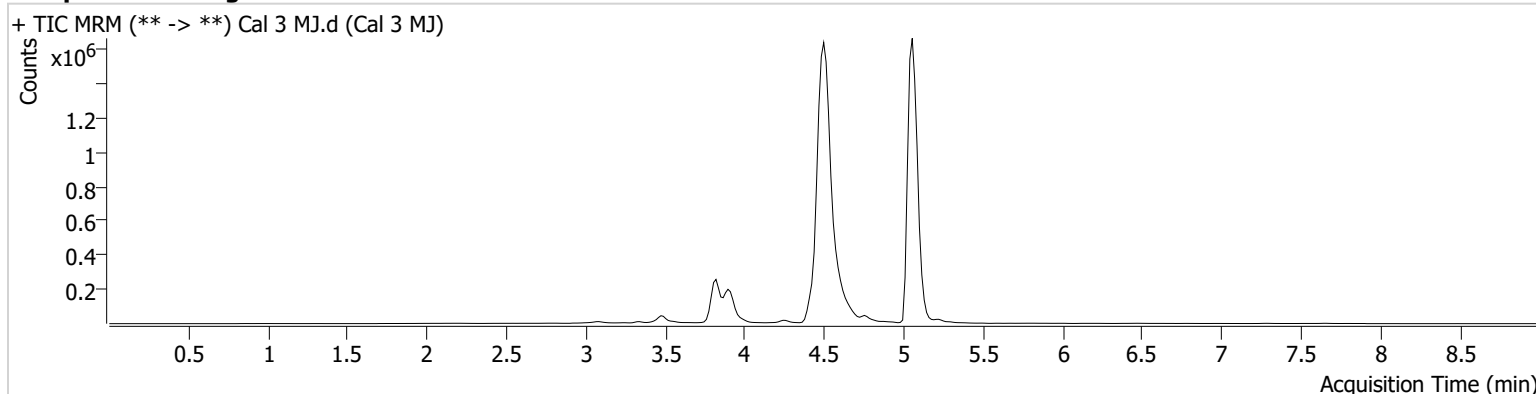
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Instrument Falco (069901)
Type Cal
Acq. Method AM 27 Agilent Method.m
Sample Position P5-F6
Injection Volume 10
Acq. Date-Time 8/12/2024 10:42:13 AM
Sample Info.

Data File Cal 3 MJ.d
Sample Cal 3 MJ
Operator Tamara Salazar
Comment

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Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.060	287043	∞	26.0	∞	6327591	4.7844 ng/ml
THC-COOH	3.909	48223	∞	240.5	∞	429154	19.0080 ng/ml
THC-OH	3.820	69027	∞	13.8	∞	945265	4.8567 ng/ml

TS



AM #27 Cannabinoids Quant. Results

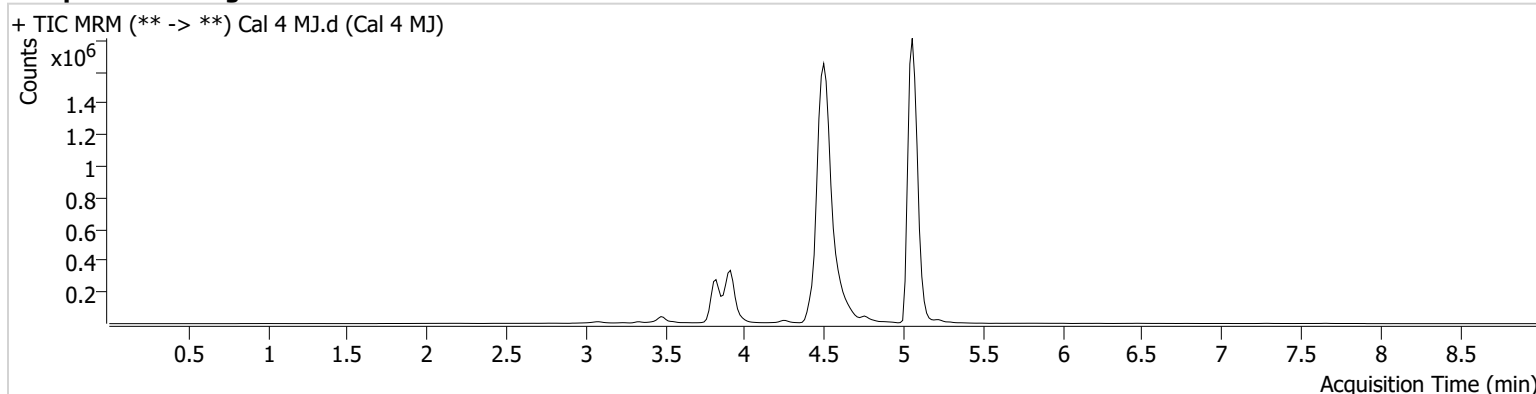
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Instrument Falco (069901)
Type Cal
Acq. Method AM 27 Agilent Method.m
Sample Position P5-E6
Injection Volume 10
Acq. Date-Time 8/12/2024 10:55:20 AM
Sample Info.

Data File Cal 4 MJ.d
Sample Cal 4 MJ
Operator Tamara Salazar
Comment

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Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.060	603516	∞	25.3	∞	6468958	9.5527 ng/ml
THC-COOH	3.909	131781	1298.43	226.6	1585.70	437936	48.9292 ng/ml
THC-OH	3.820	149002	∞	13.3	111.89	961716	9.9442 ng/ml

TS



AM #27 Cannabinoids Quant. Results

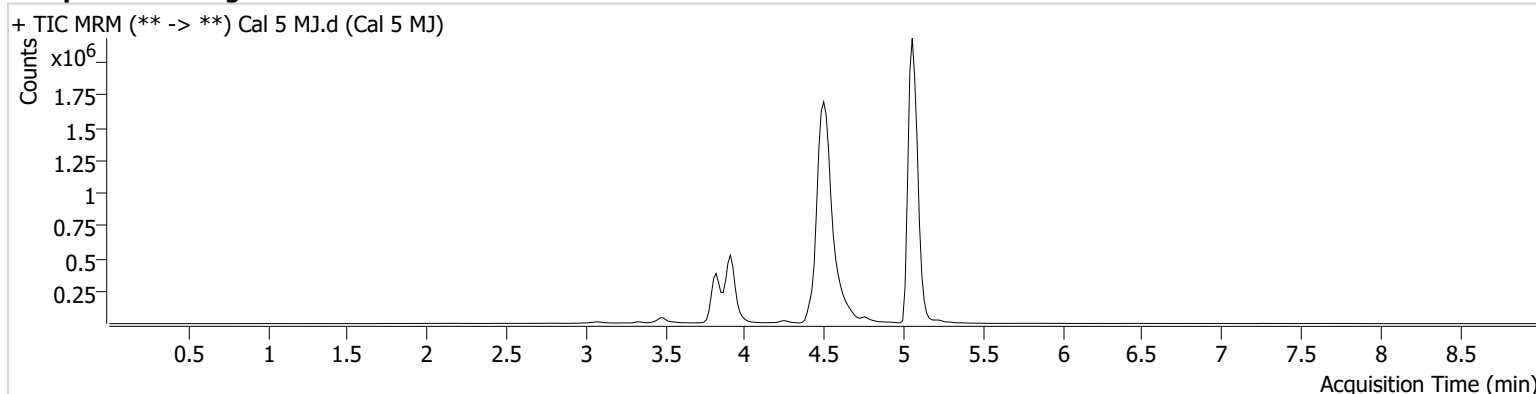
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Instrument Falco (069901)
Type Cal
Acq. Method AM 27 Agilent Method.m
Sample Position P5-D6
Injection Volume 10
Acq. Date-Time 8/12/2024 11:08:26 AM
Sample Info.

Data File Cal 5 MJ.d
Sample Cal 5 MJ
Operator Tamara Salazar
Comment

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Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.060	1641745	8318.85	25.4	∞	6658707	24.7999 ng/ml
THC-COOH	3.909	221001	1927.20	228.3	∞	482247	73.9012 ng/ml
THC-OH	3.820	424175	∞	13.8	∞	1071261	24.9146 ng/ml

TS



AM #27 Cannabinoids Quant. Results

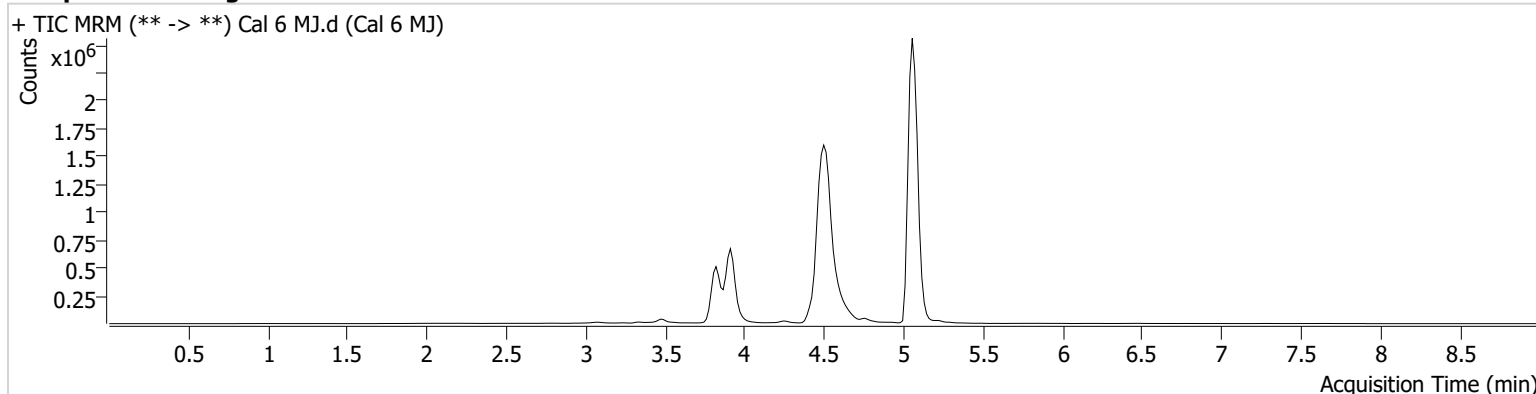
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Calibration Last Update 8/13/2024 1:19:15 PM

Instrument Falco (069901)
Type Cal
Acq. Method AM 27 Agilent Method.m
Sample Position P5-C6
Injection Volume 10
Acq. Date-Time 8/12/2024 11:21:33 AM
Sample Info.

Data File Cal 6 MJ.d
Sample Cal 6 MJ
Operator Tamara Salazar
Comment

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Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.060	3153484	∞	25.9	∞	6320203	49.9094 ng/ml
THC-COOH	3.909	294084	∞	237.2	∞	483622	97.6756 ng/ml
THC-OH	3.820	878473	∞	13.8	∞	1110527	49.4538 ng/ml

TS



AM #27 Cannabinoids Quant. Results

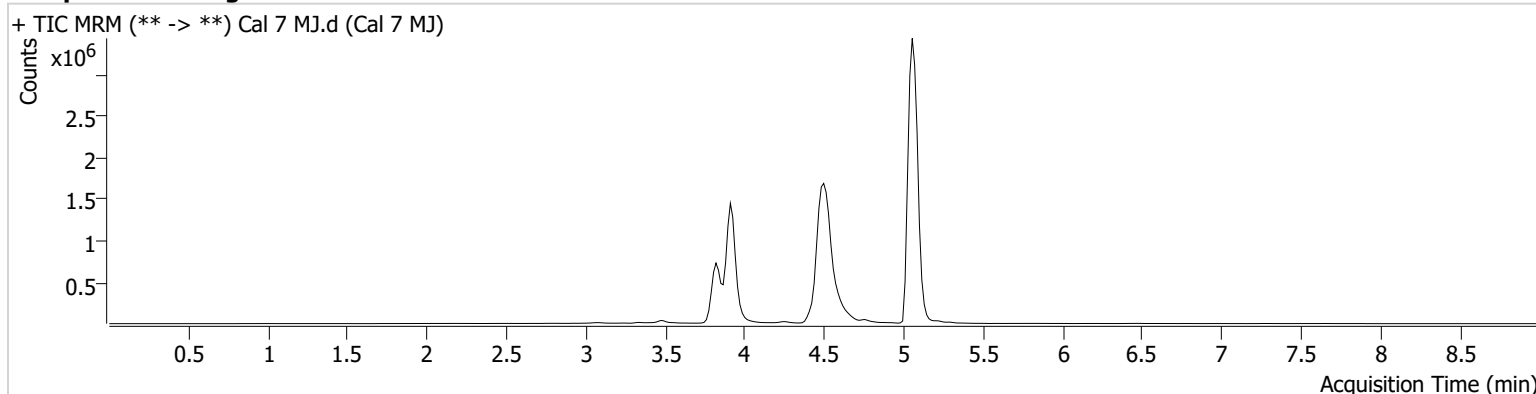
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Calibration Last Update 8/13/2024 1:19:15 PM

Instrument Falco (069901)
Type Cal
Acq. Method AM 27 Agilent Method.m
Sample Position P5-B6
Injection Volume 10
Acq. Date-Time 8/12/2024 11:34:40 AM
Sample Info.

Data File Cal 7 MJ.d
Sample Cal 7 MJ
Operator Tamara Salazar
Comment

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Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	5.060	6303792	∞	26.1	∞	6232587	100.8924 ng/ml
THC-COOH	3.909	721701	8830.75	232.5	15059.53	450989	255.1273 ng/ml
THC-OH	3.820	1741137	∞	13.7	∞	1075614	100.8631 ng/ml