REVIEWED
By Tamara Salazar at 11:42 am, May 11, 2021

5/7/2021

Worklist: 4956

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION	
P2021-1379	10	URINE	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	
P2021-1379	4	BLOOD	AM 27 Blood THC Quant by LC-QQQ	
P2021-1379	9	URINE	AM 27 Urine Cannabinoids Confirmation by LC-QQQ	

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

Extraction Date: 5/03/2021 Analyst: Amber Gerheart
Plate lot#: 210412 Plate Expiration: 10/12/2021
Mobile phase A: 0.1% Formic Acid in LCMS Water **Mobile phase B:** 0.1% Formic acid in Acetonitrile
Blank Blood Lot: 20L20723 **Column**: UCT Selectra DA 100 x 2.1mm 3um
LCMS-QQQ ID: 069901 **Blank Urine Lot**: POC031319

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:** *42*
☒ 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 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)***
☒ 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 samples with calculated concentrations for THC at 1ng/mL or greater and OH-THC at 3ng/mL or greater may be reported quantitatively (blood only). Calculated concentrations for carboxy-THC of 5ng/mL may be reported qualitatively. 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: *Calibration Curve Range: THC 3-100 ng/mL, THC-OH 3-100 ng/mL*

	1	2	3	4	5	6
A	IS + Cal. 1	Negative Blood	IS + Sample	IS + Sample	IS + Sample	IS + QC_1
B	IS + Cal. 2	Negative Urine	IS + Sample	IS + Sample	IS + Sample	IS + Cal. 7
C	IS + Cal. 3	Urine External	IS + Sample	IS + Sample	IS + Sample	IS + Cal. 6
D	IS + Cal. 4	P2021-1379-4	IS + Sample	IS + Sample	IS + Sample	IS + Cal. 5
E	IS + Cal. 5	P2021-1379-9	IS + Sample	IS + Sample	IS + Sample	IS + Cal. 4
F	IS + Cal. 6	P2021-1379-10	IS + Sample	IS + Sample	IS + Sample	IS + Cal. 3
G	IS + Cal. 7	IS + Sample	IS + Sample	IS + Sample	IS + Sample	IS + Cal. 2
H	IS + QC_1	IS + Sample	IS + Sample	IS + Sample	IS + Sample	IS + Cal. 1

All wells to contain 100 µl of residual DMSO

Idaho State Police
Forensic Services

AM #26 Screening of THC and Metabolites and AM #27 Confirmation of THC and Metabolites Blood External Control Prep Sheet

Methanol External Control Solution (Lot: WS03052021)

10 µL of 1mg/mL THC, 100 µL of 100 µg/mL THC-OH, C-THC in 9790 µL MeOH

Approximate concentration 1ug/mL.

Component	***Source***	***Source Lot Number***	***Expiration Date***
Methanol (LCMS)	Fisher	200921	
THC	Cerilliant	FE01041701	03/31/2022
C-THC	Cerilliant	FE08011801	08/31/2023
THC-OH	Cerilliant	FE07221601	07/31/2021
Prepared:	03/05/2021		
Prepared By:	Tamara Salazar/Amber Gerheart		

Urine External Control Solution (Lot: 04232021)

200 ul of methanol external control solution was added to 9800 ul of blood.

Approximately 20ng/mL each

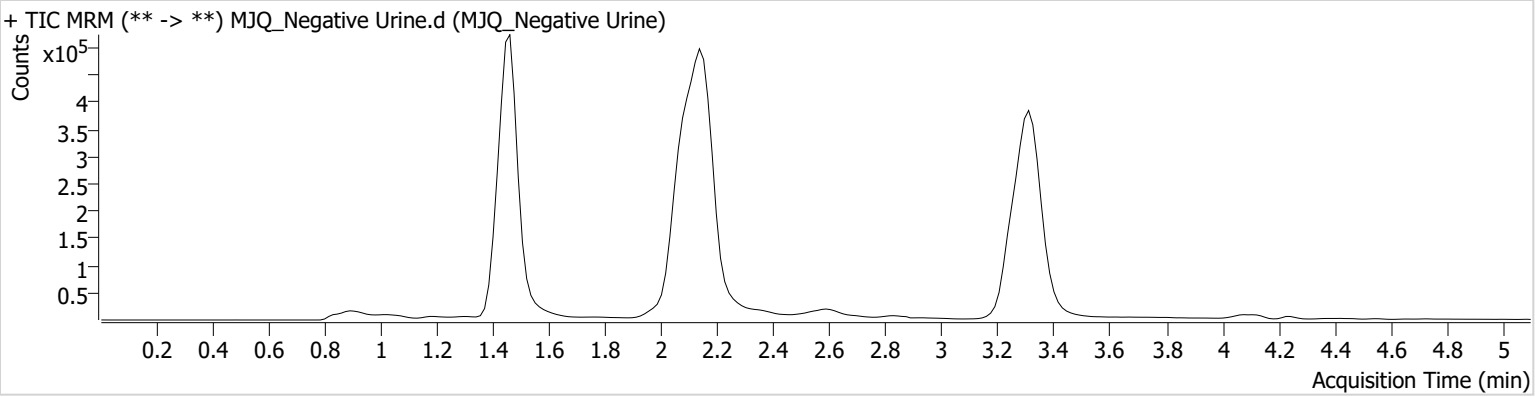
Component	***Source***	***Source Lot Number***
Negative Blood	Lampire	20L20724
Methanol External Control Solution	-	WS03052021
Prepared:	04/23/2021	
Prepared by:	Sarah Collins	

AM #27 Cannabinoid Quant. Results

Batch results	D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update	5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_Negative Urine.d
Type	Sample	**Sample**	MJQ_Negative Urine
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-B2	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 3:01:11 PM		
Sample Info.			

Sample Chromatogram

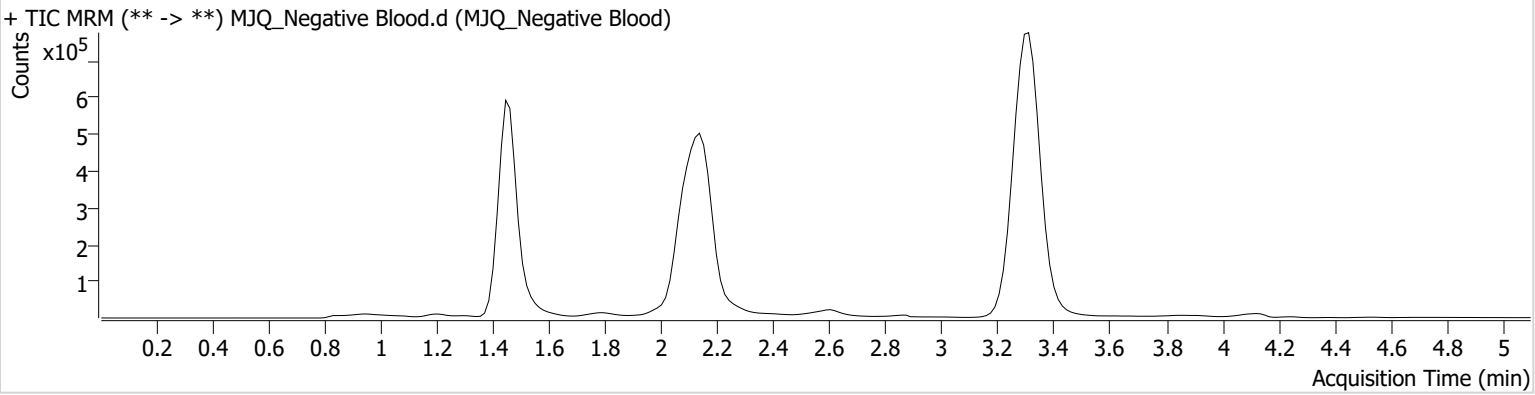


AM #27 Cannabinoid Quant. Results

Batch results	D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update	5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_Negative Blood.d
Type	Sample	**Sample**	MJQ_Negative Blood
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-A2	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 2:45:58 PM		
Sample Info.			

Sample Chromatogram

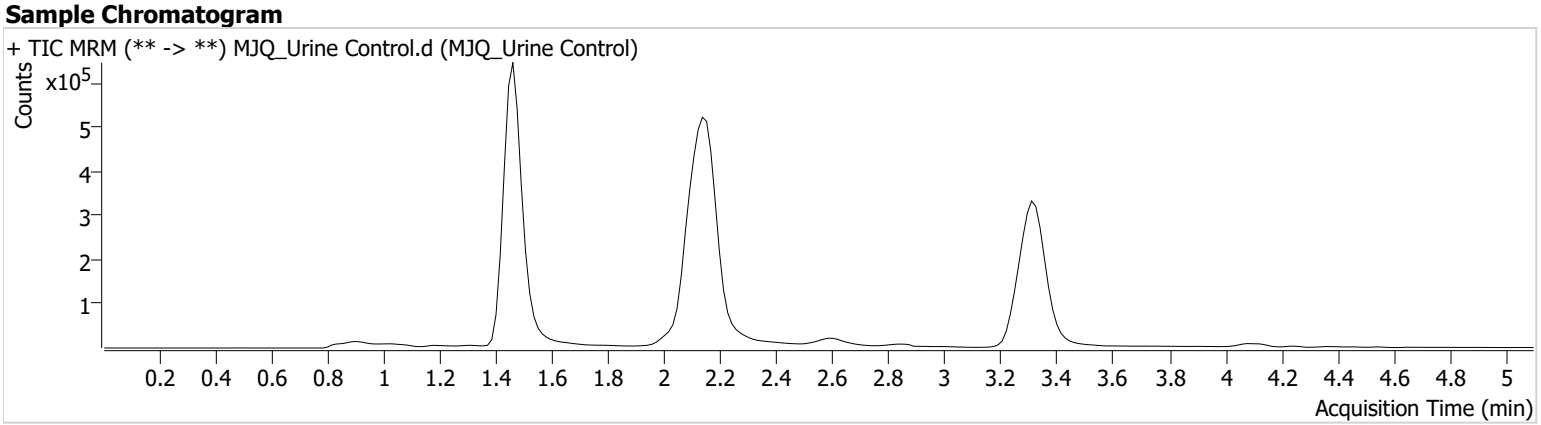


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update 5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_Urine Control.d
Type	Sample	**Sample**	MJQ_Urine Control
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-C2	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 3:16:24 PM		
Sample Info.			

Sample Chromatogram



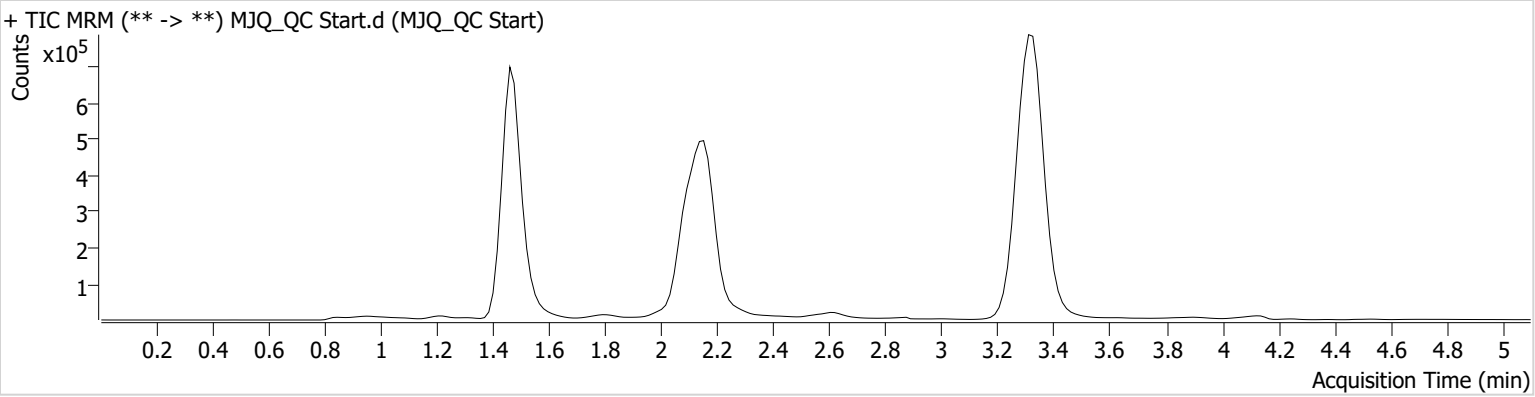
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	316894	∞	12.2	∞	1978672	9.5234	ng/ml
THC-COOH	1.489	154450	∞	48.9	∞	459920	12.6682	ng/ml
THC	3.330	215931	243.44	31.7	424.55	1968432	10.7891	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update 5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_QC Start.d
Type	Sample	**Sample**	MJQ_QC Start
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-H1	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 2:30:45 PM		
Sample Info.			

Sample Chromatogram



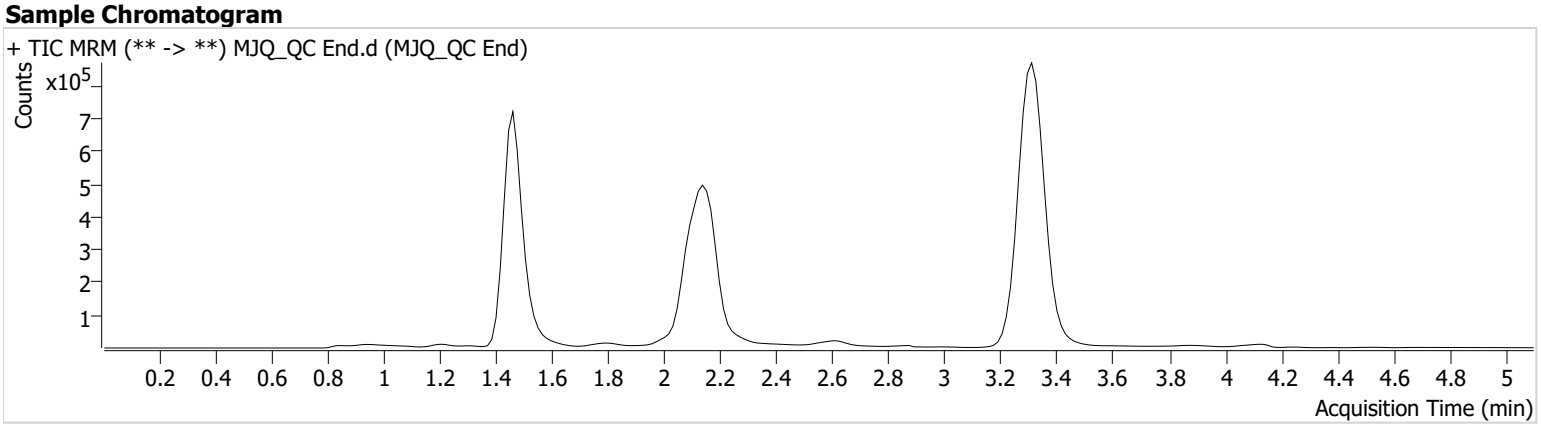
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	189162	∞	10.8	90.92	2185879	4.9867	ng/ml
THC-COOH	1.504	248317	591.55	51.6	1239.43	615242	15.1972	ng/ml
THC	3.330	228211	795.49	31.2	216.57	5136118	4.5755	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update 5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_QC End.d
Type	Sample	**Sample**	MJQ_QC End
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-H1	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 4:09:40 PM		
Sample Info.			

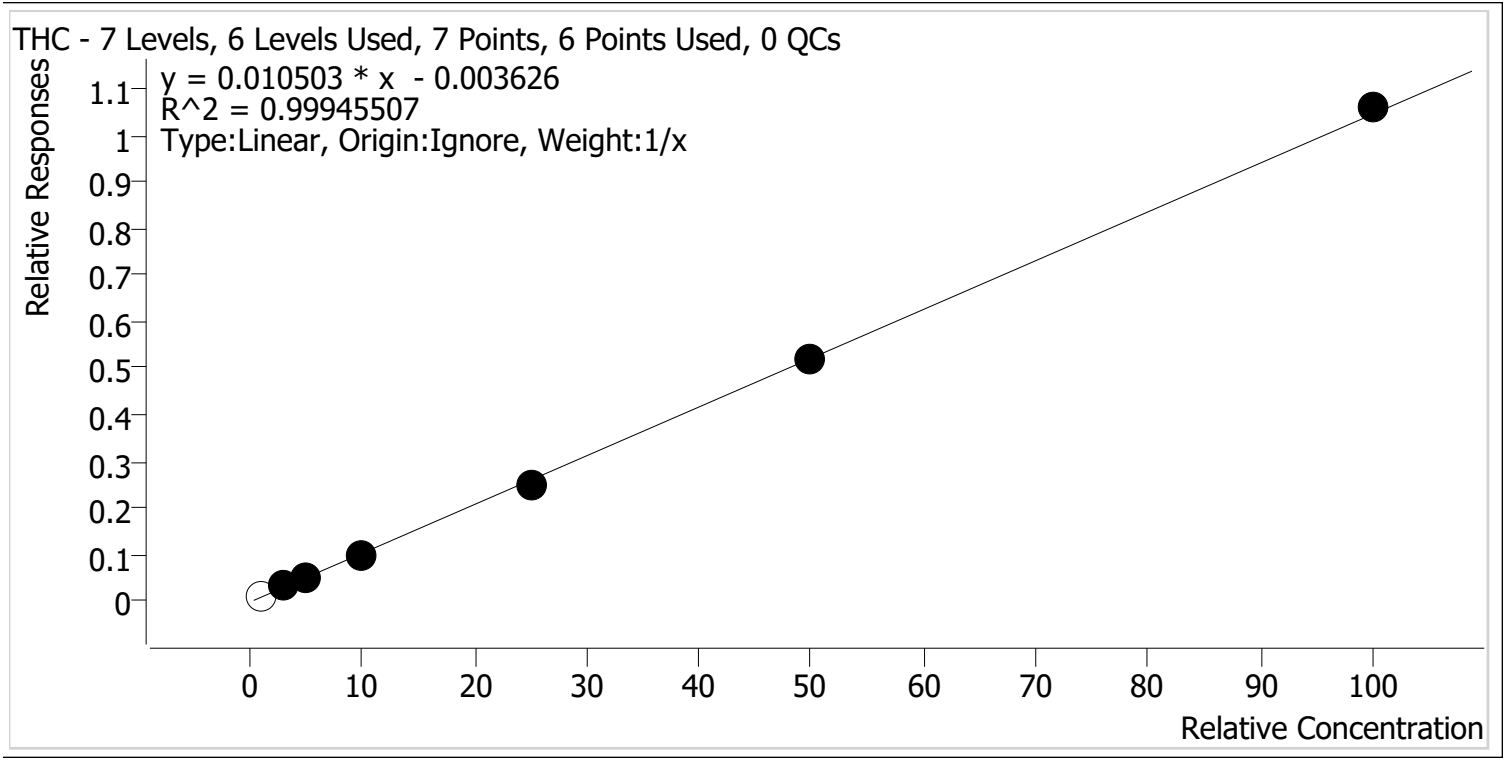
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	174503	∞	11.7	∞	2197104	4.5483	ng/ml
THC-COOH	1.489	248585	∞	52.2	1537.41	622254	15.0436	ng/ml
THC	3.330	243496	505.04	30.3	47.64	5487703	4.5696	ng/ml

AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Last Cal. Update 5/10/2021 10:24 AM
Analyst Name ISP\datastor
Analyte THC **Internal Standard** THC-D3

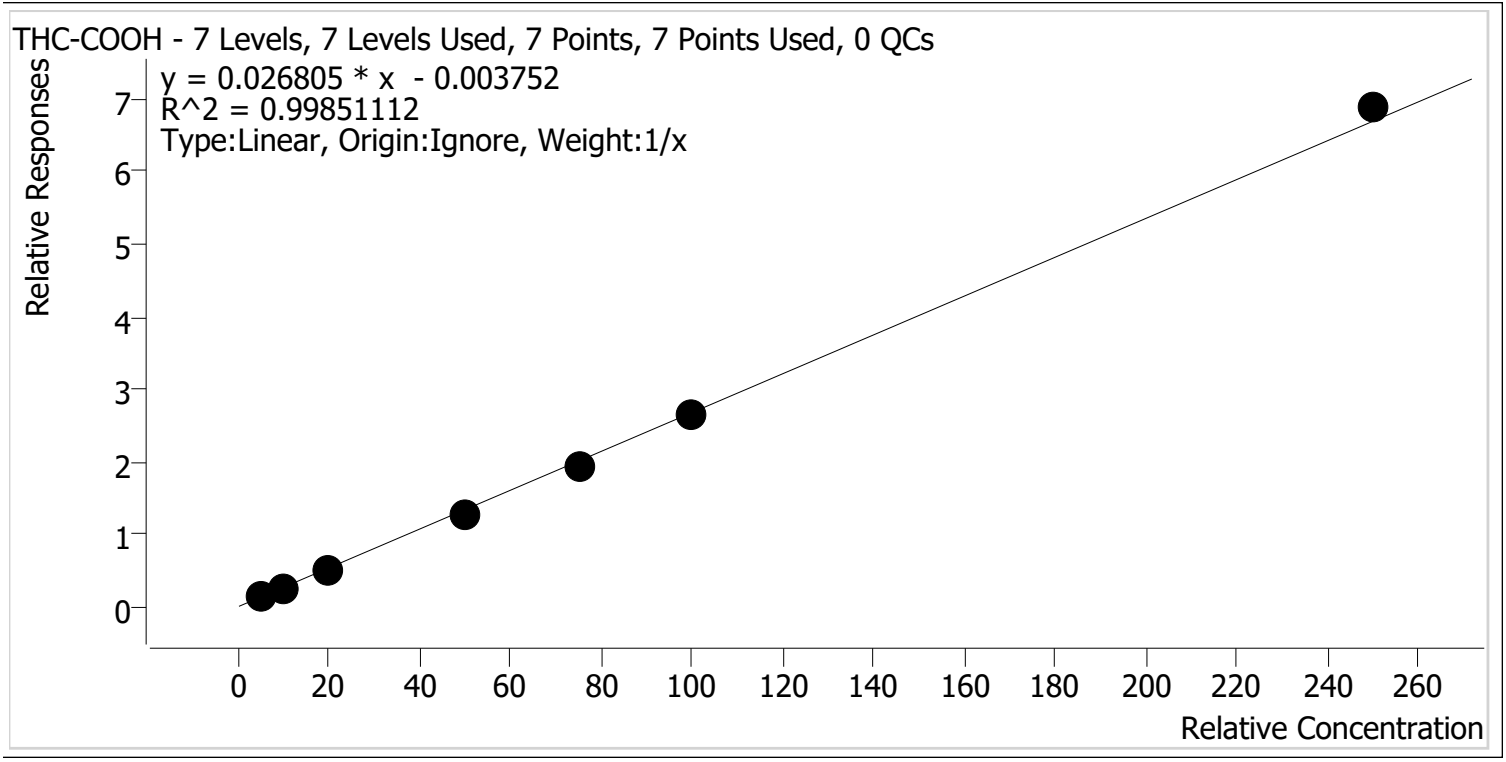


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
* MJQ_Cal 1	1	✗	1.0	1.3	131.8
MJQ_Cal 2	2	✔	3.0	3.2	106.0
MJQ_Cal 3	3	✔	5.0	5.1	101.3
MJQ_Cal 4	4	✔	10.0	9.5	94.8
MJQ_Cal 5	5	✔	25.0	24.3	97.3
MJQ_Cal 6	6	✔	50.0	49.6	99.2
MJQ_Cal 7	7	✔	100.0	101.3	101.3

*Cal 1 dropped due to S/N

AM #27 Cannabinoids Quant. Calibration Curve Report

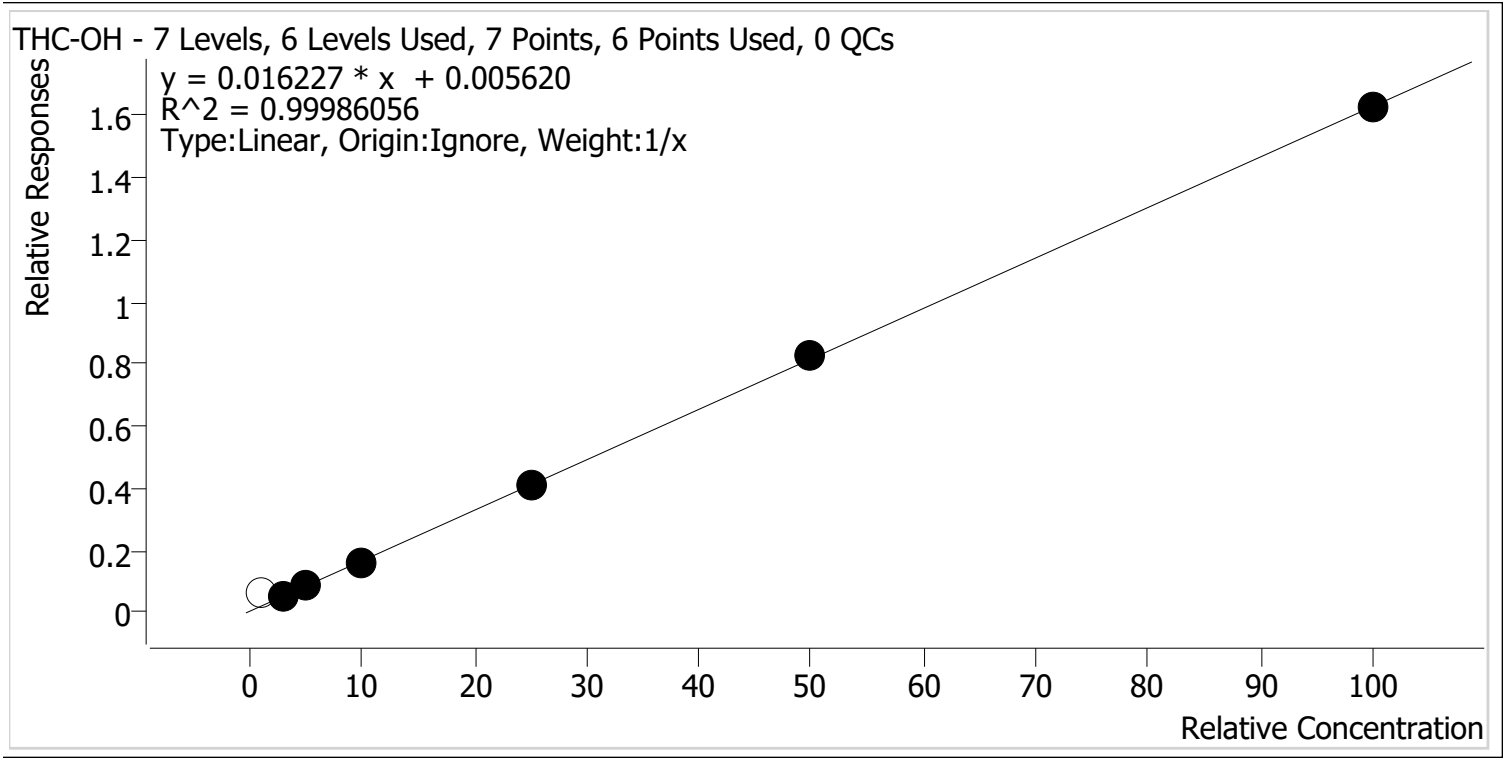
Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Last Cal. Update 5/10/2021 10:24 AM
Analyst Name ISP\datastor
Analyte THC-COOH **Internal Standard** THC-COOH-D9



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJQ_Cal 1	1	✔	5.0	5.6	112.4
MJQ_Cal 2	2	✔	10.0	9.9	99.1
MJQ_Cal 3	3	✔	20.0	19.2	96.1
MJQ_Cal 4	4	✔	50.0	47.4	94.8
MJQ_Cal 5	5	✔	75.0	71.9	95.9
MJQ_Cal 6	6	✔	100.0	98.9	98.9
MJQ_Cal 7	7	✔	250.0	257.0	102.8

AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results	D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Last Cal. Update	5/10/2021 10:24 AM
Analyst Name	ISP\datastor
Analyte	THC-OH
Internal Standard	THC-OH-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
* MJQ_Cal 1	1	✗	1.0	3.4	341.4
MJQ_Cal 2	2	✔	3.0	3.1	102.5
MJQ_Cal 3	3	✔	5.0	5.0	100.2
MJQ_Cal 4	4	✔	10.0	9.7	96.5
MJQ_Cal 5	5	✔	25.0	25.0	100.1
MJQ_Cal 6	6	✔	50.0	50.5	101.0
MJQ_Cal 7	7	✔	100.0	99.7	99.7

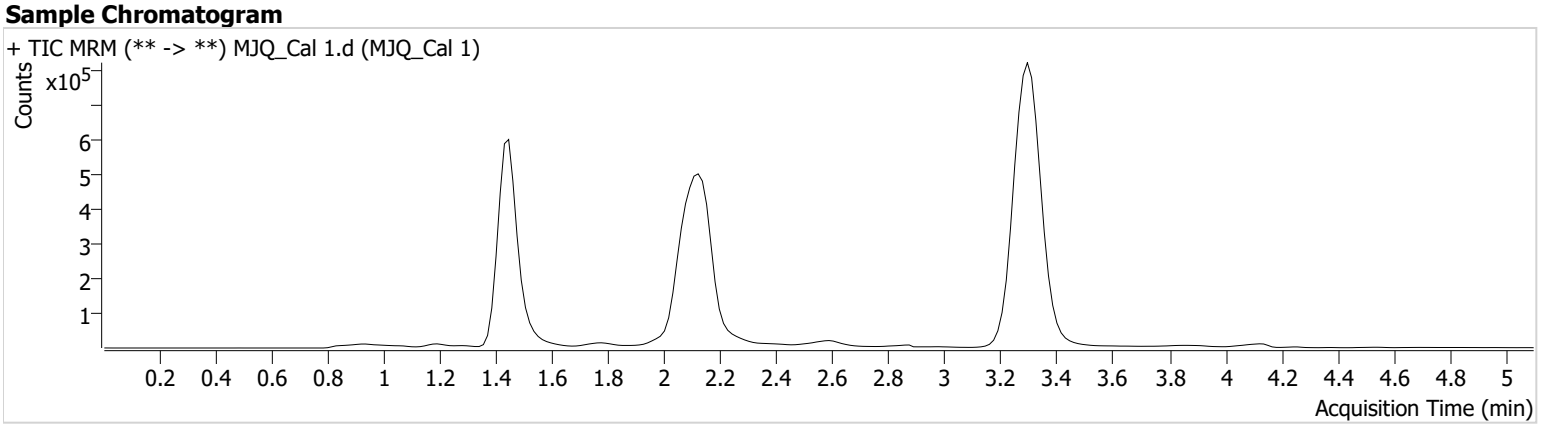
*Cal 1 dropped due to retention time

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update 5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 1.d
Type	Cal	**Sample**	MJQ_Cal 1
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-A1	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 1:29:49 PM		
Sample Info.			

Sample Chromatogram



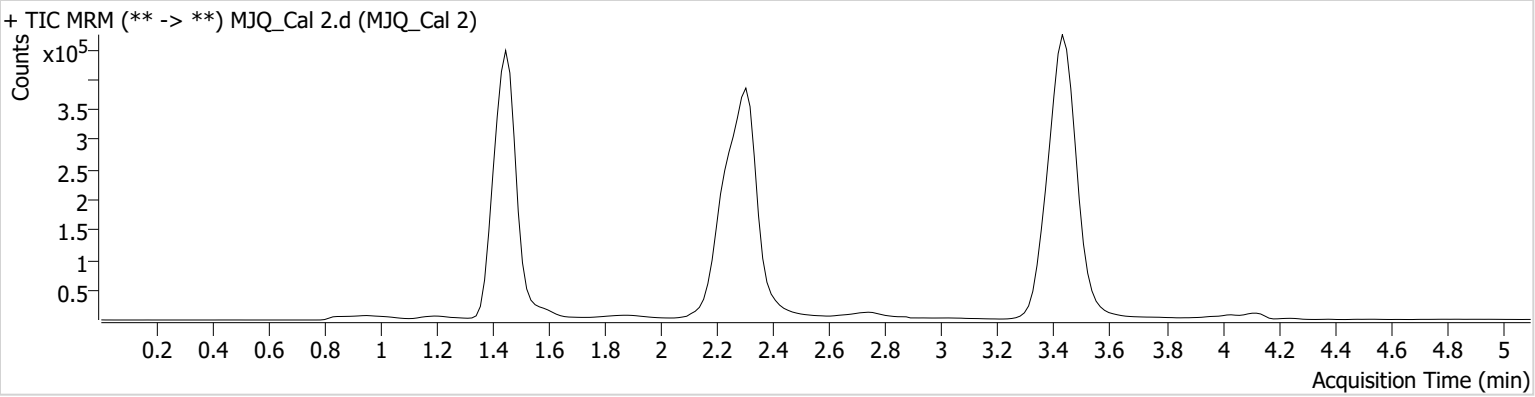
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.513 High	129894	∞	5.4 Low	32.95	2128914	3.4138	ng/ml
THC-COOH	1.474	88404	∞	46.6	∞	601819	5.6201	ng/ml
THC	3.315	57205	165.18	28.2	5.13 Low	5596434	1.3184	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update 5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 2.d
Type	Cal	**Sample**	MJQ_Cal 2
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-B1	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 1:37:35 PM		
Sample Info.			

Sample Chromatogram



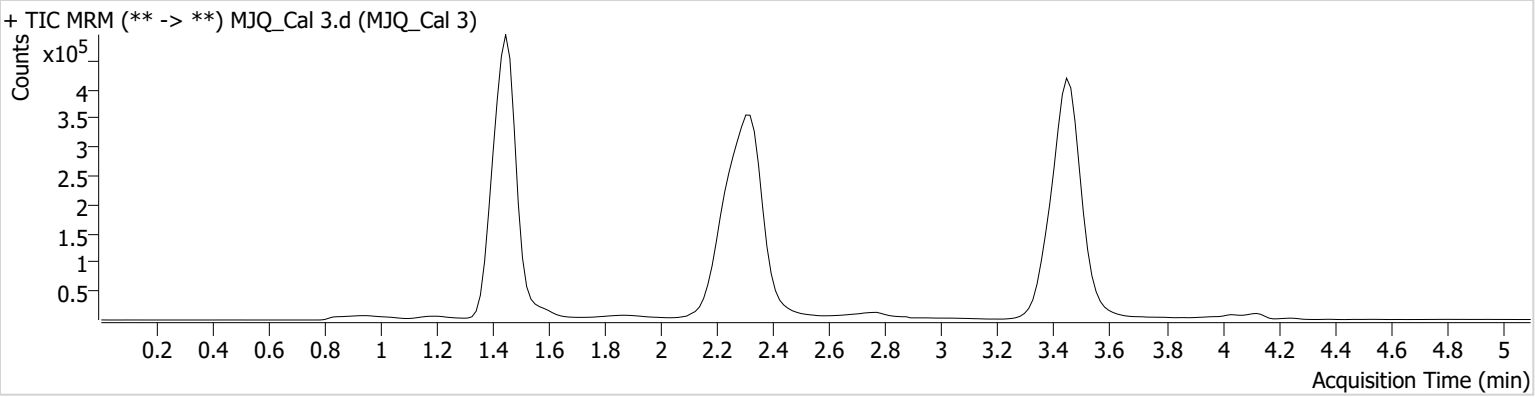
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	96401	∞	9.4	∞	1736547	3.0747	ng/ml
THC-COOH	1.459	119745	∞	46.7	1031.64	457410	9.9064	ng/ml
THC	3.435	96939	305.41	31.2	∞	3254027	3.1815	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update 5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 3.d
Type	Cal	**Sample**	MJQ_Cal 3
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-C1	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 1:45:11 PM		
Sample Info.			

Sample Chromatogram



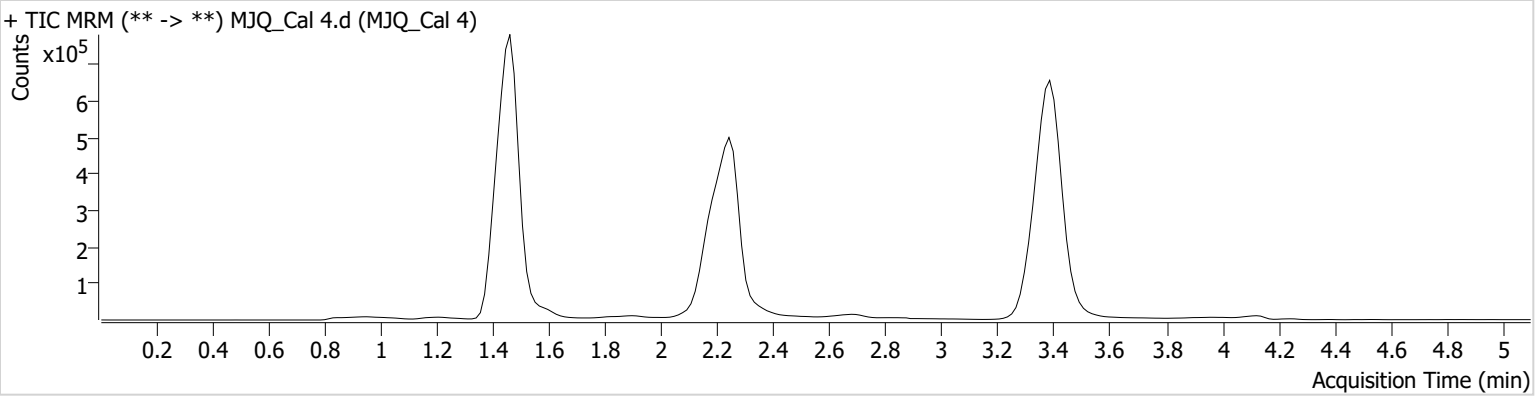
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.453	158359	∞	9.7	26.89	1822739	5.0078	ng/ml
THC-COOH	1.459	237515	∞	49.4	970.89	464435	19.2188	ng/ml
THC	3.465	146414	∞	25.2	12.42	2954471	5.0634	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update 5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 4.d
Type	Cal	**Sample**	MJQ_Cal 4
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-D1	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 1:52:47 PM		
Sample Info.			

Sample Chromatogram



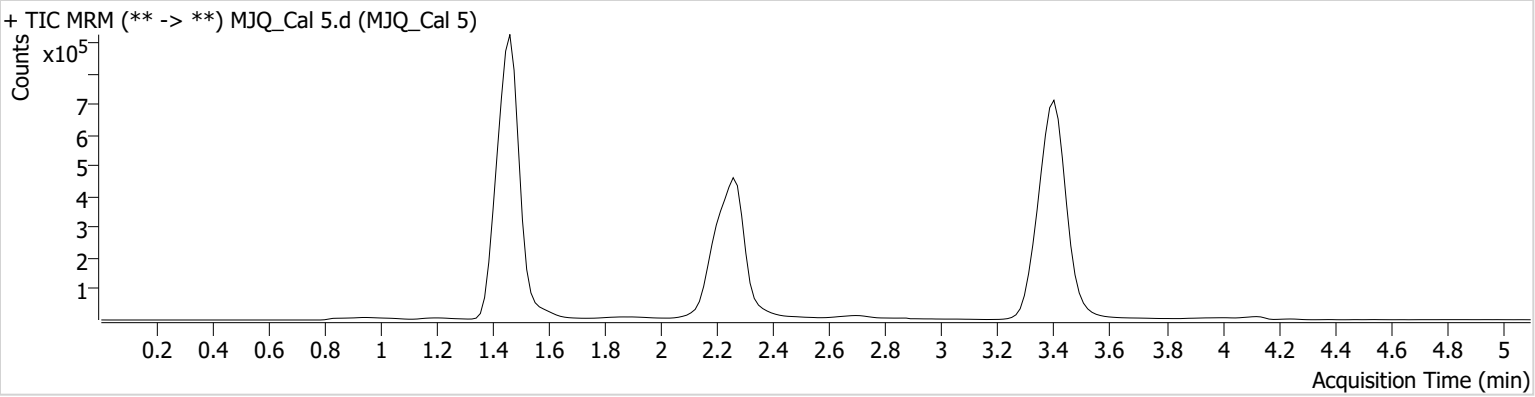
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	361264	∞	10.7	584.55	2227109	9.6502	ng/ml
THC-COOH	1.474	698685	∞	52.9	∞	551371	47.4139	ng/ml
THC	3.405	388552	499.83	26.7	133.32	4049130	9.4812	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update 5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 5.d
Type	Cal	**Sample**	MJQ_Cal 5
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-E1	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 2:00:23 PM		
Sample Info.			

Sample Chromatogram



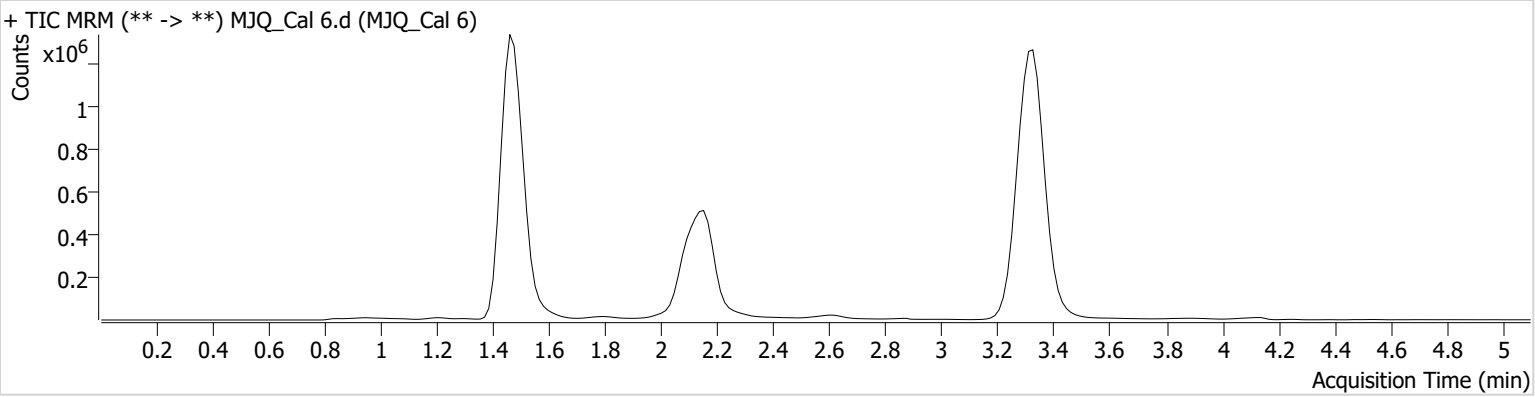
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	839192	∞	10.4	695.02	2038092	25.0287	ng/ml
THC-COOH	1.474	986137	∞	52.5	∞	512576	71.9132	ng/ml
THC	3.405	950072	554.02	26.1	260.90	3772094	24.3248	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update 5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 6.d
Type	Cal	**Sample**	MJQ_Cal 6
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-F1	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 2:07:58 PM		
Sample Info.			

Sample Chromatogram



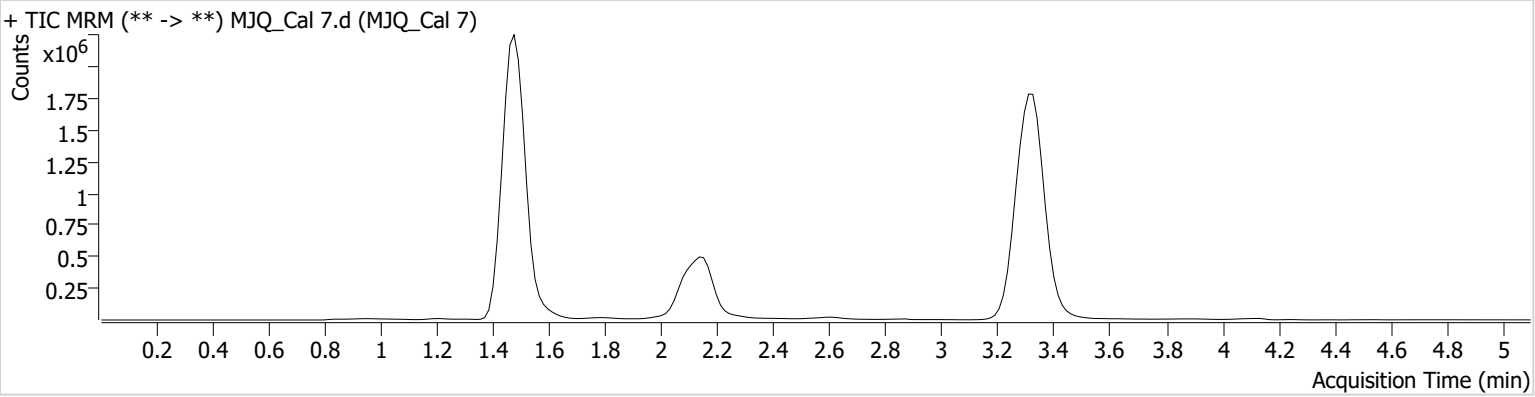
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	1790164	∞	11.1	∞	2169788	50.4981	ng/ml
THC-COOH	1.489	1534946	∞	54.4	∞	579660	98.9279	ng/ml
THC	3.330	2716277	∞	26.7	∞	5247856	49.6240	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\050321 AM 27 28 AG\QuantResults\AM 27.batch.bin
Calibration Last Update 5/10/2021 10:24:13 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 7.d
Type	Cal	**Sample**	MJQ_Cal 7
Acq. Method	AM 27 THCQ.m	**Operator**	Amber Gerheart
Sample Position	P1-G1	**Comment**	
Injection Volume	10		
Acq. Date-Time	5/3/2021 2:15:34 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	3328062	∞	11.3	∞	2049192	99.7405	ng/ml
THC-COOH	1.489	3738878	∞	53.3	3076.40	543037	256.9997	ng/ml
THC	3.330	5775716	∞	26.6	3602.78	5445506	101.3252	ng/ml