REVIEWED
By Anne Nord at 11:14 am, Oct 27, 2021

10/26/2021

Worklist: 5328

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION
M2021-4216	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2021-4379	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2021-4380	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2021-4450	7	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-3326	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-3329	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-3376	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-3386	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-3390	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-3398	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-3445	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-3446	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-3467	1	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: 10/25/21
Plate lot#: IDP-108-2-210609
Mobile phase A: 0.1% Formic Acid in LCMS Water
Blank Blood Lot: Lampire 20L20724
LCMS-QQQ ID: 069901

Analyst: Sarah Collins
Retest Date: 12/09/2021
Mobile phase B: 0.1% Formic acid in Acetonitrile
Column: UCT Selectra DA 100 x 2.1mm 3um

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: Did not evaluate THC-OH due to interfering peak. Due to varying internal standard responses in initial injections of the calibrators, the run was stopped and the calibrators were reinjected with no further issues.

	1	2	3	4	5	6
A	IS + Cal. 1			p2021-3329-1	p2021-3376-1	IS + QC_1
B	IS + Cal. 2			p2021-3326-2	p2021-3329-1*	IS + Cal. 7
C	IS + Cal. 3			p2021-3467-1	p2021-3326-2*	IS + Cal. 6
D	IS + Cal. 4			p2021-3446-1	m2021-4450-7	IS + Cal. 5
E	IS + Cal. 5			p2021-3445-1*	m2021-4380-1	IS + Cal. 4
F	IS + Cal. 6			p2021-3398-1	m2021-4379-1	IS + Cal. 3
G	IS + Cal. 7			p2021-3390-1	m2021-4216-1	IS + Cal. 2
H	IS + QC_1		p2021-3445-1	p2021-3386-1	negative blood	IS + Cal. 1

All wells to contain 100 µl of residual DMSO

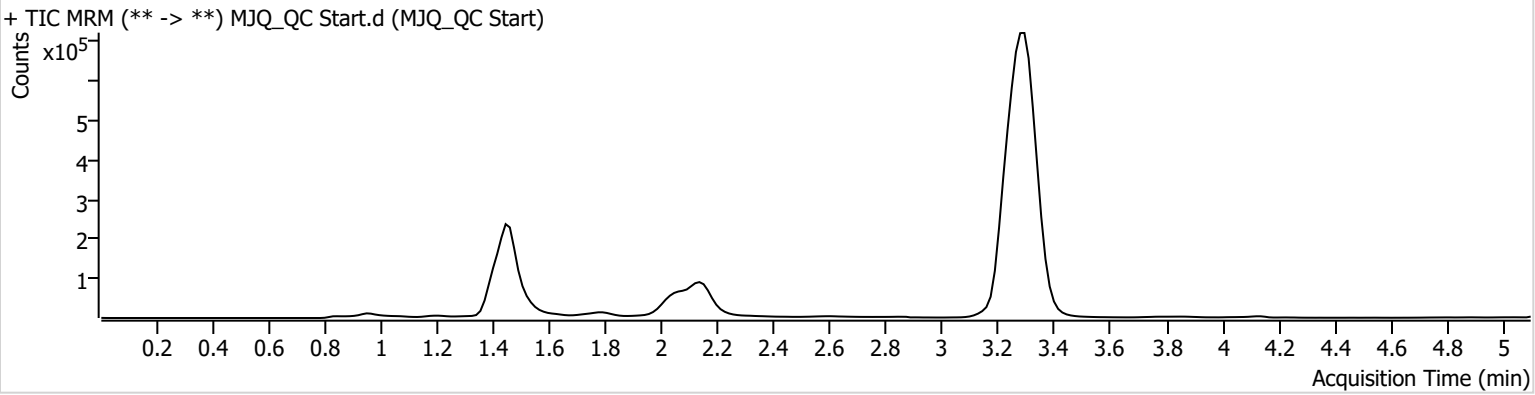
*Samples moved during analytical step 6 due to blood clot

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/26/2021 11:07:17 AM

Instrument	Falco (069901)	**Data File**	MJQ_QC Start.d
Type	Sample	**Sample**	MJQ_QC Start
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-A6	**Comment**	
Injection Volume	10		
Acq. Date-Time	10/25/2021 5:55:25 PM		
Sample Info.			

Sample Chromatogram



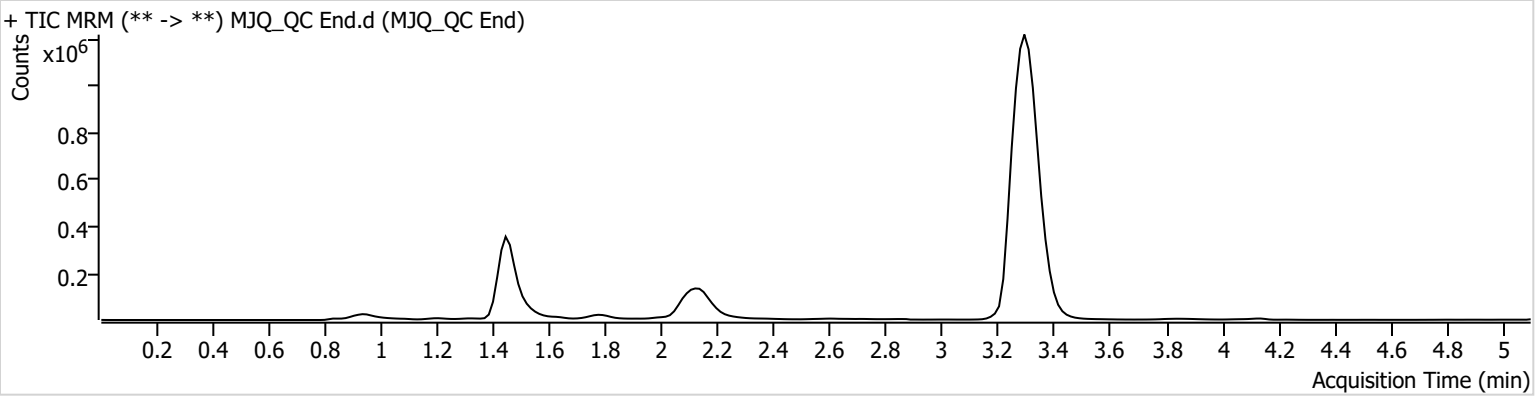
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-COOH	1.489	61466	∞	57.4	1510.39	152613	15.5773	ng/ml
THC-OH	1.468	150905	∞	8.0	∞	1010462	4.9102	ng/ml
THC	3.300	231543	1165.47	27.3	497.79	5179975	4.7531	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/26/2021 11:07:17 AM

Instrument	Falco (069901)	**Data File**	MJQ_QC End.d
Type	Sample	**Sample**	MJQ_QC End
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-A6	**Comment**	
Injection Volume	10		
Acq. Date-Time	10/25/2021 9:43:50 PM		
Sample Info.			

Sample Chromatogram



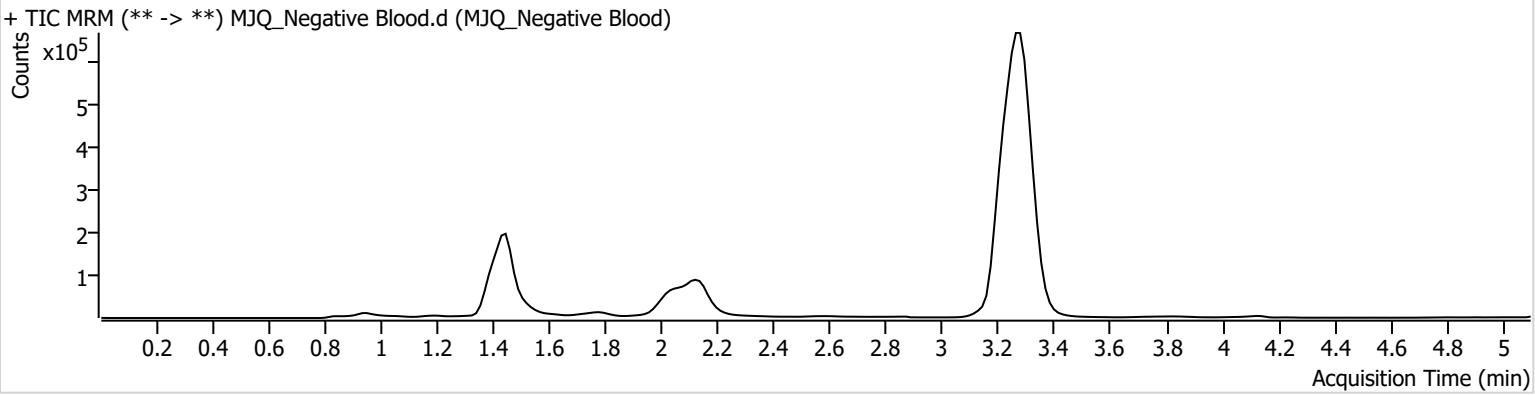
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-COOH	1.489	66365	∞	62.9	1056.64	192718	13.2883	ng/ml
THC-OH	1.513	179323	∞	6.9 **Low**	117.64	1132351	5.5997	ng/ml
THC	3.315	349758	2621.46	27.2	693.93	7723696	4.8128	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results	D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update	10/26/2021 11:07:17 AM

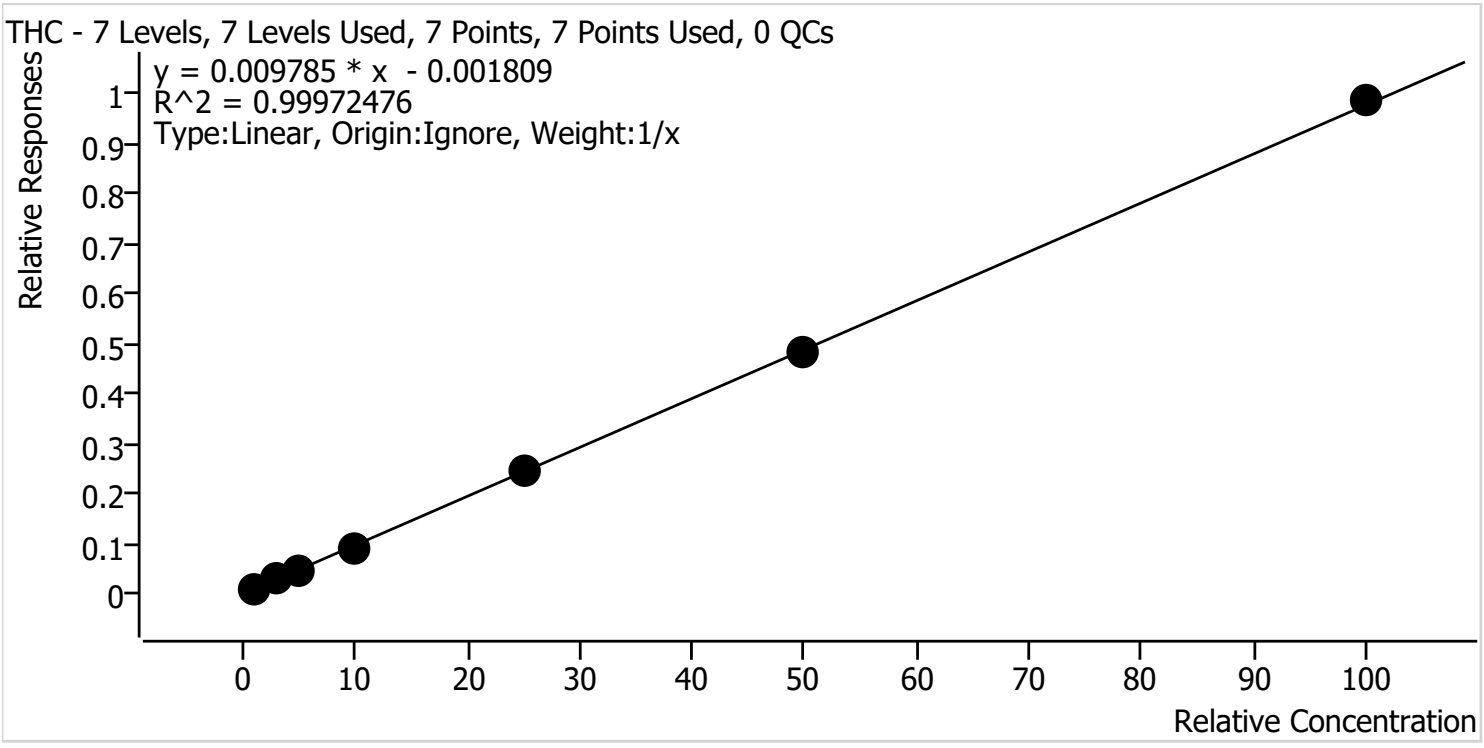
Instrument	Falco (069901)	**Data File**	MJQ_Negative Blood.d
Type	Sample	**Sample**	MJQ_Negative Blood
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-H5	**Comment**	
Injection Volume	10		
Acq. Date-Time	10/25/2021 6:10:42 PM		
Sample Info.			

Sample Chromatogram



AM #27 Cannabinoids Quant. Calibration Curve Report

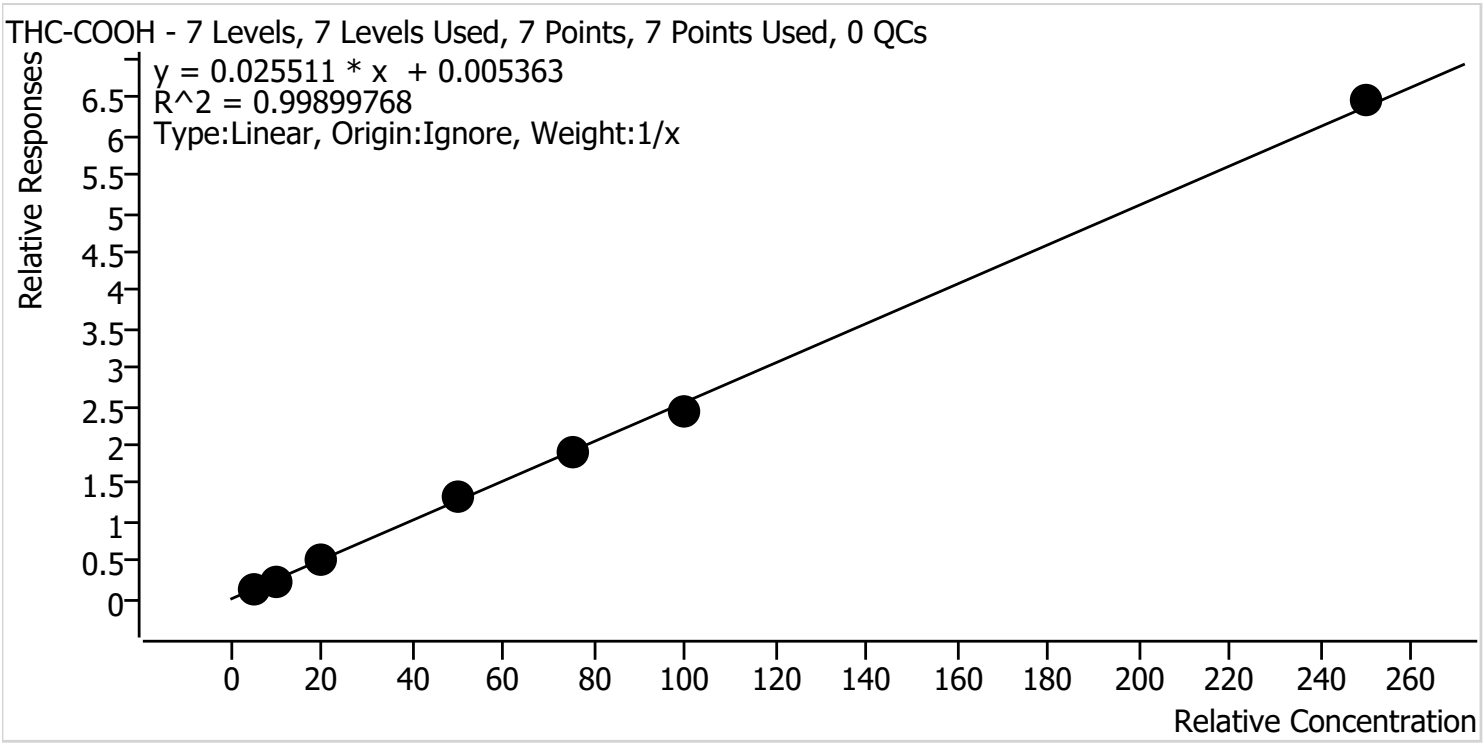
Batch results	D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update	10/26/2021 11:07 AM
Analyst Name	ISP\datastor
Analyte	THC
Internal Standard	THC-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJQ_Cal 1R	1	✔	1.0	1.1	110.3
MJQ_Cal 2R	2	✔	3.0	2.9	97.1
MJQ_Cal 3R	3	✔	5.0	4.8	96.2
MJQ_Cal 4R	4	✔	10.0	9.6	96.1
MJQ_Cal 5R	5	✔	25.0	25.0	100.0
MJQ_Cal 6R	6	✔	50.0	49.7	99.3
MJQ_Cal 7R	7	✔	100.0	100.9	100.9

AM #27 Cannabinoids Quant. Calibration Curve Report

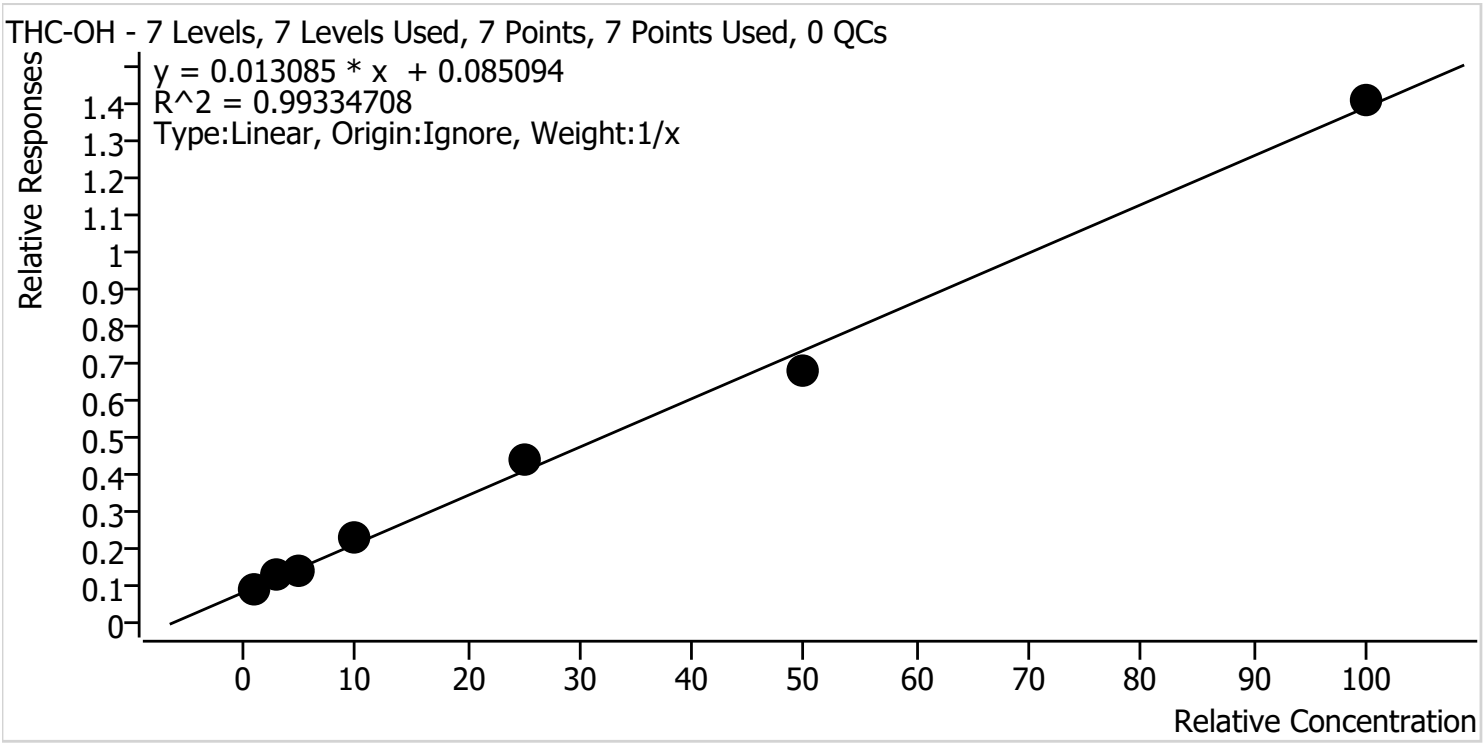
Batch results	D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update	10/26/2021 11:07 AM
Analyst Name	ISP\datastor
Analyte	THC-COOH
Internal Standard	THC-COOH-D9



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJQ_Cal 1R	1	✔	5.0	5.1	102.9
MJQ_Cal 2R	2	✔	10.0	9.4	94.3
MJQ_Cal 3R	3	✔	20.0	20.6	103.2
MJQ_Cal 4R	4	✔	50.0	52.1	104.1
MJQ_Cal 5R	5	✔	75.0	74.0	98.6
MJQ_Cal 6R	6	✔	100.0	95.6	95.6
MJQ_Cal 7R	7	✔	250.0	253.2	101.3

AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results	D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update	10/26/2021 11:07 AM
Analyst Name	ISP\datastor
Analyte	THC-OH
Internal Standard	THC-OH-D3



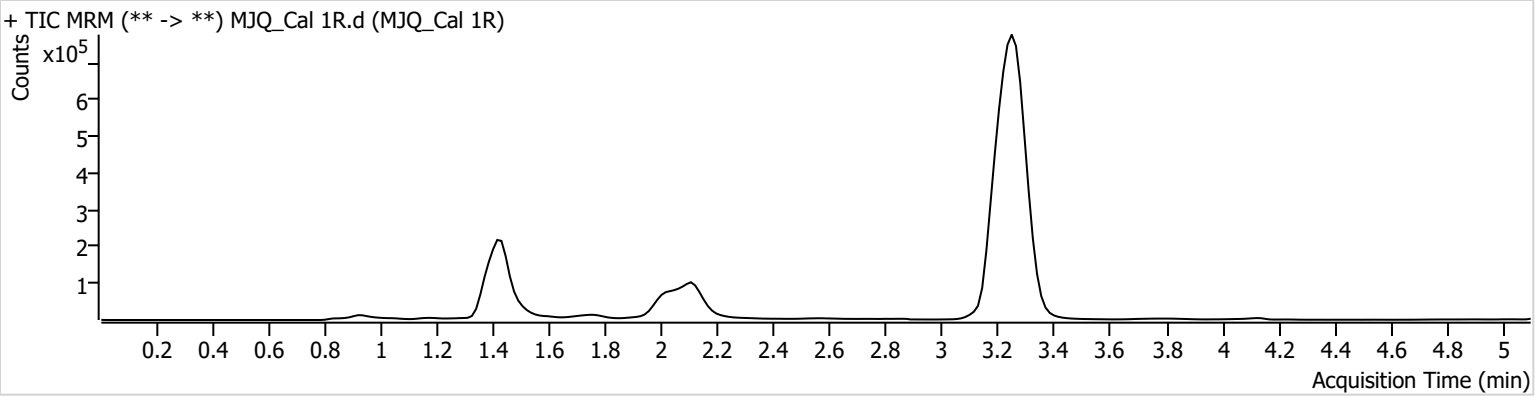
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJQ_Cal 1R	1	✔	1.0	0.8	82.2
MJQ_Cal 2R	2	✔	3.0	3.6	121.3
MJQ_Cal 3R	3	✔	5.0	4.1	82.7
MJQ_Cal 4R	4	✔	10.0	11.2	112.2
MJQ_Cal 5R	5	✔	25.0	27.3	109.2
MJQ_Cal 6R	6	✔	50.0	45.5	91.0
MJQ_Cal 7R	7	✔	100.0	101.4	101.4

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/26/2021 11:07:17 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 1R.d
Type	Cal	**Sample**	MJQ_Cal 1R
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-H6	**Comment**	
Injection Volume	10		
Acq. Date-Time	10/25/2021 4:54:28 PM		
Sample Info.			

Sample Chromatogram



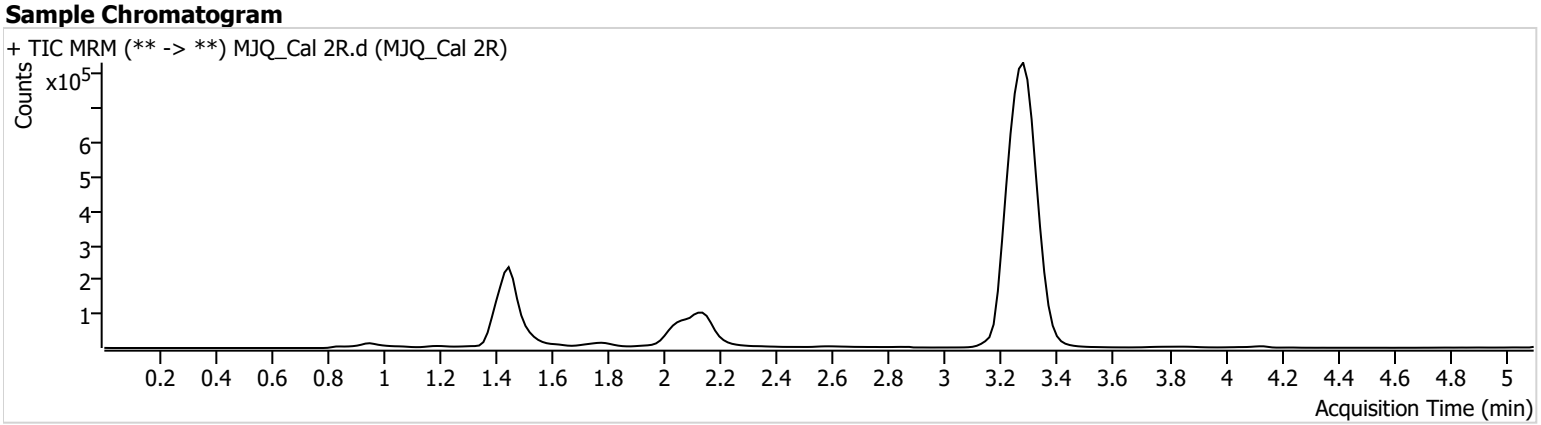
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-COOH	1.459	24425	106.08	50.7	∞	178852	5.1430	ng/ml
THC-OH	1.498	98702	∞	4.7 **Low**	19.23	1029787	0.8219	ng/ml **Low**
THC	3.270	52497	∞	32.7	∞	5844134	1.1029	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/26/2021 11:07:17 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 2R.d
Type	Cal	**Sample**	MJQ_Cal 2R
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-G6	**Comment**	
Injection Volume	10		
Acq. Date-Time	10/25/2021 5:02:15 PM		
Sample Info.			

Sample Chromatogram



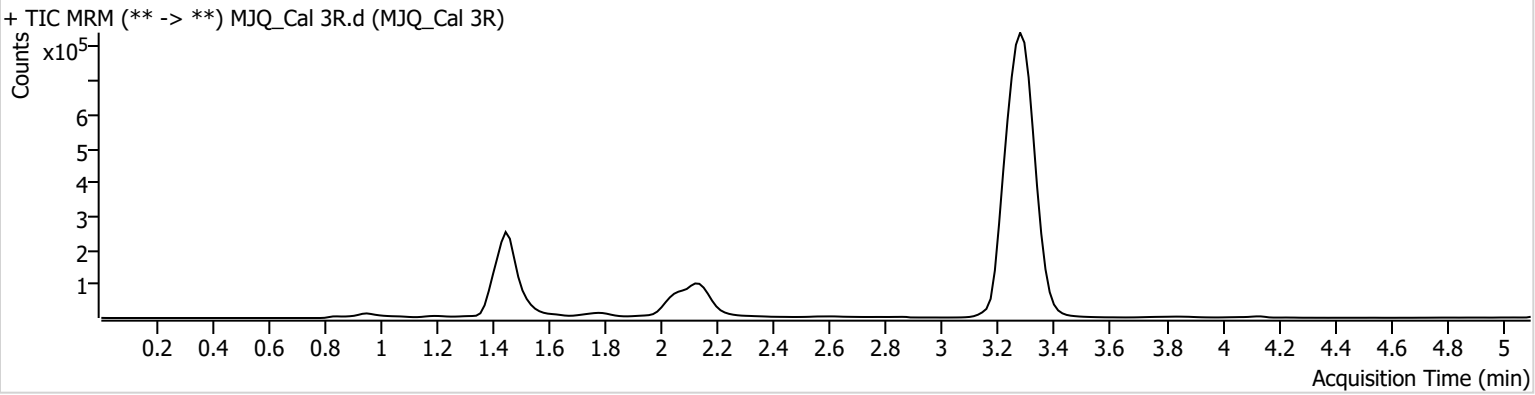
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-COOH	1.474	39383	∞	59.0	244.11	160076	9.4338	ng/ml
THC-OH	1.513	122808	∞	6.9 **Low**	∞	925327	3.6397	ng/ml
THC	3.300	159578	427.60	28.2	226.63	5974774	2.9144	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/26/2021 11:07:17 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 3R.d
Type	Cal	**Sample**	MJQ_Cal 3R
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-F6	**Comment**	
Injection Volume	10		
Acq. Date-Time	10/25/2021 5:09:50 PM		
Sample Info.			

Sample Chromatogram



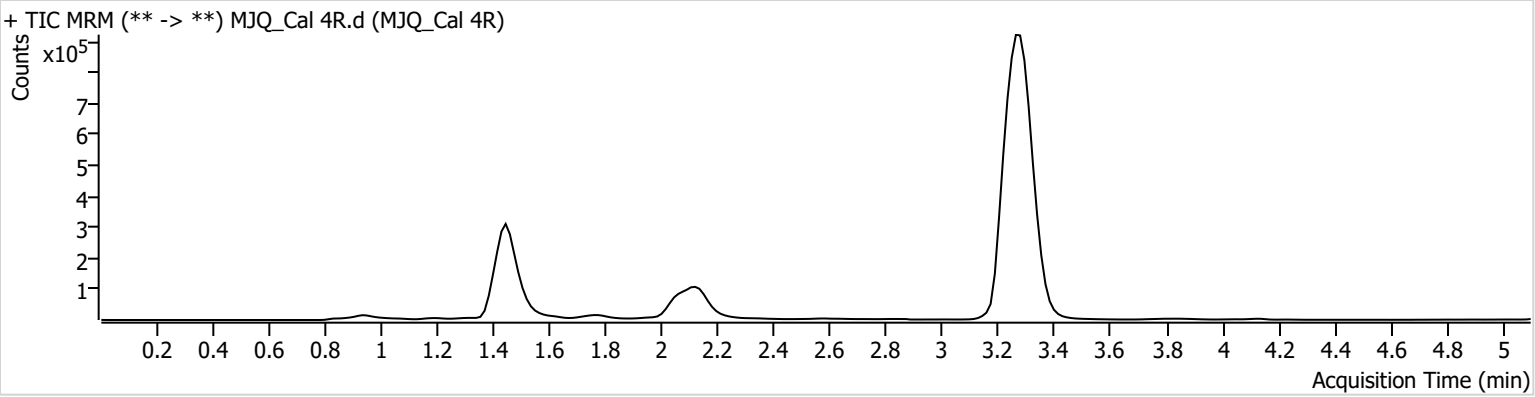
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-COOH	1.489	84072	245.45	58.6	∞	158055	20.6401	ng/ml
THC-OH	1.468	138534	∞	8.5	85.55	995221	4.1350	ng/ml
THC	3.300	265543	2018.11	27.0	263.34	5864562	4.8123	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/26/2021 11:07:17 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 4R.d
Type	Cal	**Sample**	MJQ_Cal 4R
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-E6	**Comment**	
Injection Volume	10		
Acq. Date-Time	10/25/2021 5:17:27 PM		
Sample Info.			

Sample Chromatogram



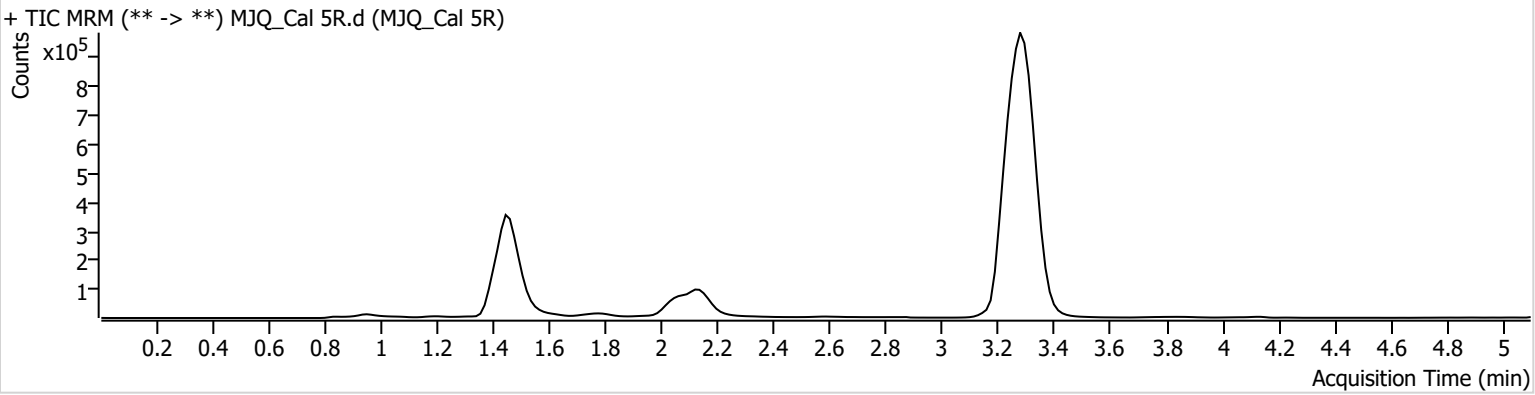
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-COOH	1.474	199995	1201.83	64.2	1810.15	150008	52.0507	ng/ml
THC-OH	1.453 Low	213888	∞	9.4	∞	922385	11.2187	ng/ml
THC	3.285	545774	∞	26.5	328.96	5918910	9.6084	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/26/2021 11:07:17 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 5R.d
Type	Cal	**Sample**	MJQ_Cal 5R
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-D6	**Comment**	
Injection Volume	10		
Acq. Date-Time	10/25/2021 5:25:02 PM		
Sample Info.			

Sample Chromatogram



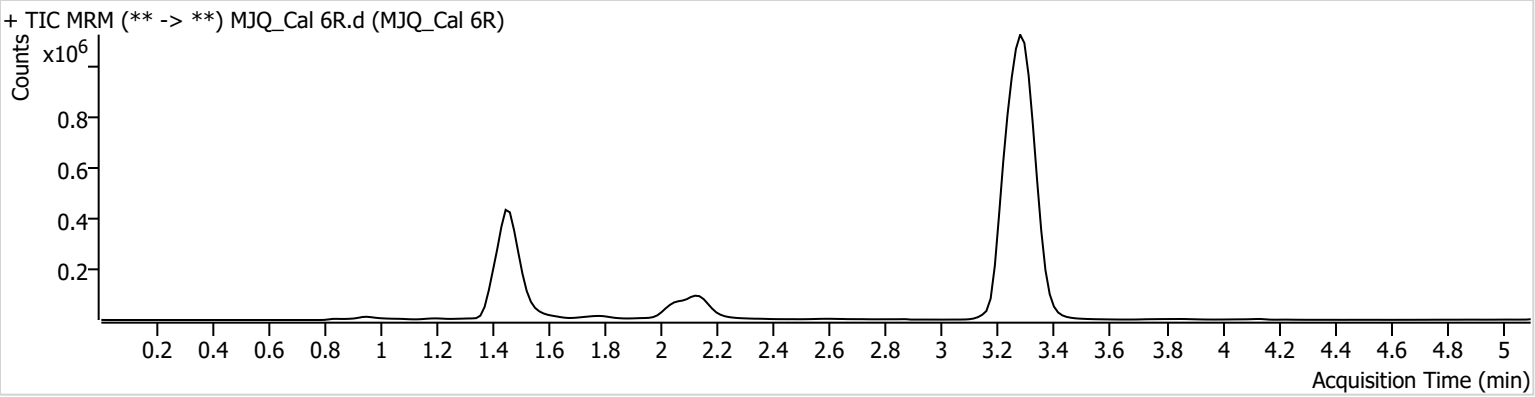
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-COOH	1.474	284875	∞	66.6	∞	150518	73.9787	ng/ml
THC-OH	1.453 Low	408224	∞	10.8	869.92	922657	27.3106	ng/ml
THC	3.300	1361457	∞	25.3	1922.71	5607438	24.9978	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/26/2021 11:07:17 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 6R.d
Type	Cal	**Sample**	MJQ_Cal 6R
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-C6	**Comment**	
Injection Volume	10		
Acq. Date-Time	10/25/2021 5:32:38 PM		
Sample Info.			

Sample Chromatogram



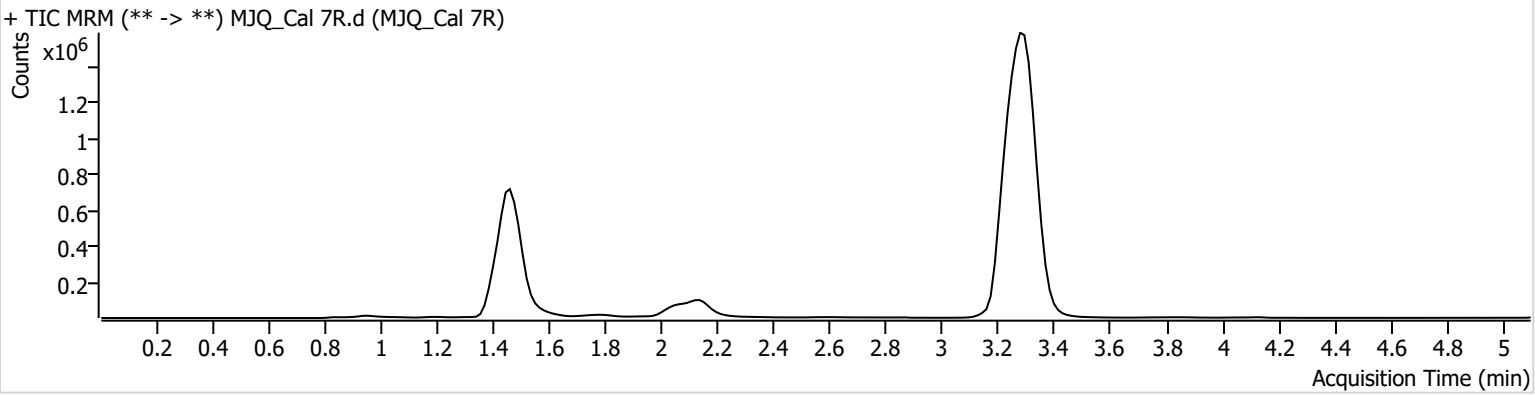
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-COOH	1.474	377112	∞	68.4	∞	154286	95.6006	ng/ml
THC-OH	1.453 Low	658197	∞	13.0 High	∞	967563	45.4861	ng/ml
THC	3.300	2576387	∞	25.9	3076.94	5320435	49.6732	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\102521 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 10/26/2021 11:07:17 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 7R.d
Type	Cal	**Sample**	MJQ_Cal 7R
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-B6	**Comment**	
Injection Volume	10		
Acq. Date-Time	10/25/2021 5:40:13 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-COOH	1.474	950142	3711.82	68.8	∞	147000	253.1531	ng/ml
THC-OH	1.453 Low	1252735	∞	13.6 High	1261.35	887383	101.3880	ng/ml
THC	3.300	5406581	∞	26.2	∞	5486636	100.8909	ng/ml