REVIEWED
By Tamara Salazar at 12:21 pm, Jul 26, 2021

7/22/2021

Worklist: 5115

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION
M2021-2376	4	BCK	AM 27 Blood THC Quant by LC-QQQ
M2021-2691	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2021-2803	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2021-3160	3	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2307	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2343	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2359	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2367	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2378	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2381	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2382	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2385	2	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2406	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2418	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-2420	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: 07/21/21
Plate lot#: IDP-108-2-210609
Mobile phase A: 0.1% Formic Acid in LCMS Water
Blank Blood Lot: Lampire 20L20723
LCMS-QQQ ID: 069901

Analyst: Sarah Collins
Plate Expiration: 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.

	1	2	3	4	5	6
A	IS + Cal. 1	negative blood	p2021-2367-1			
B	IS + Cal. 2	m2021-2376-4	p2021-2378-1			
C	IS + Cal. 3	m2021-2691-1	p2021-2381-1			
D	IS + Cal. 4	m2021-2803-1	p2021-2382-1			
E	IS + Cal. 5	m2021-3160-3	p2021-2385-2			
F	IS + Cal. 6	p2021-2307-1	p2021-2406-1			
G	IS + Cal. 7	p2021-2343-1	p2021-2418-1			
H	IS + QC_1	p2021-2359-1	p2021-2420-1			

All wells to contain 100 µl of residual DMSO

Position of samples on analytical plate.

	1	2	3	4	5	6
A				IS + Cal. 1	negative blood	p2021-2367-1
B				IS + Cal. 2	m2021-2376-4	p2021-2378-1
C				IS + Cal. 3	m2021-2691-1	p2021-2381-1
D				IS + Cal. 4	m2021-2803-1	p2021-2382-1
E				IS + Cal. 5	m2021-3160-3	p2021-2385-2
F				IS + Cal. 6	p2021-2307-1	p2021-2406-1
G				IS + Cal. 7	p2021-2343-1	p2021-2418-1
H				IS + QC_1	p2021-2359-1	p2021-2420-1

All wells to contain 100 µl of residual DMSO

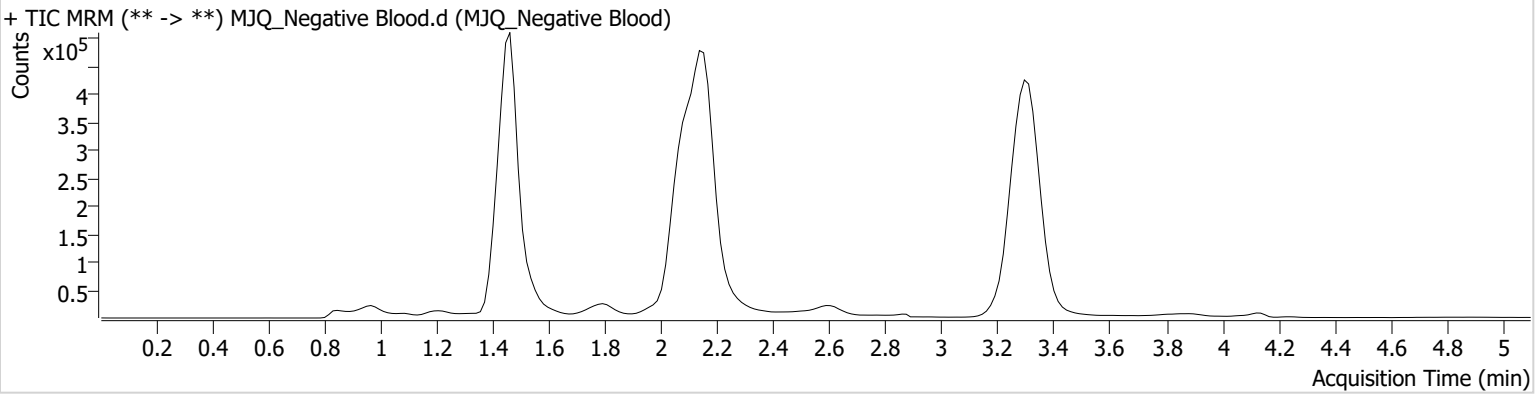
Sample positions on SLE plate and catcher plate

AM #27 Cannabinoid Quant. Results

Batch results	D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update	7/22/2021 10:06:47 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-A5	**Comment**	
Injection Volume	10		
Acq. Date-Time	7/21/2021 3:56:48 PM		
Sample Info.			

Sample Chromatogram

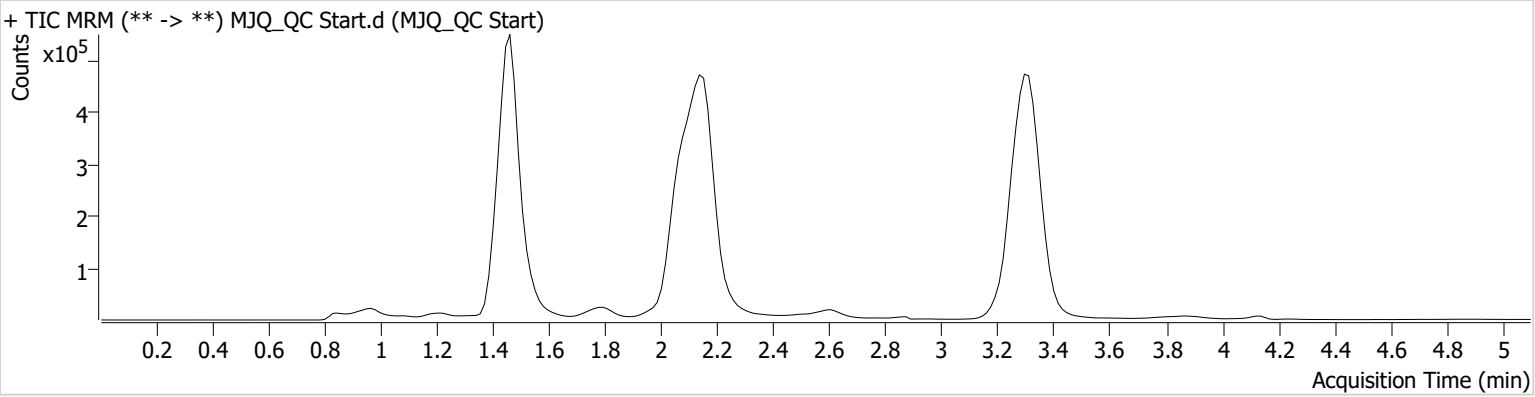


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 7/22/2021 10:06:47 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-H4	**Comment**	
Injection Volume	10		
Acq. Date-Time	7/21/2021 3:41:37 PM		
Sample Info.			

Sample Chromatogram



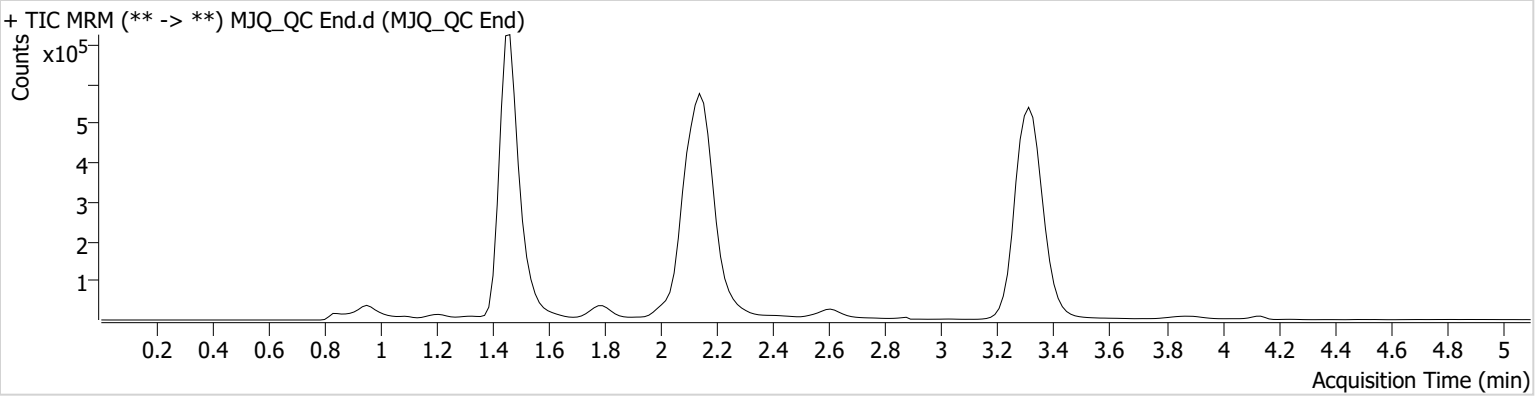
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.483	293670	34.11	6.3	∞	1936917	4.4567	ng/ml
THC-COOH	1.489	159353	310.57	54.2	∞	438712	14.8336	ng/ml
THC	3.315	150997	∞	28.7	472.82	3336111	4.6914	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 7/22/2021 10:06:47 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-H4	**Comment**	
Injection Volume	10		
Acq. Date-Time	7/21/2021 8:00:25 PM		
Sample Info.			

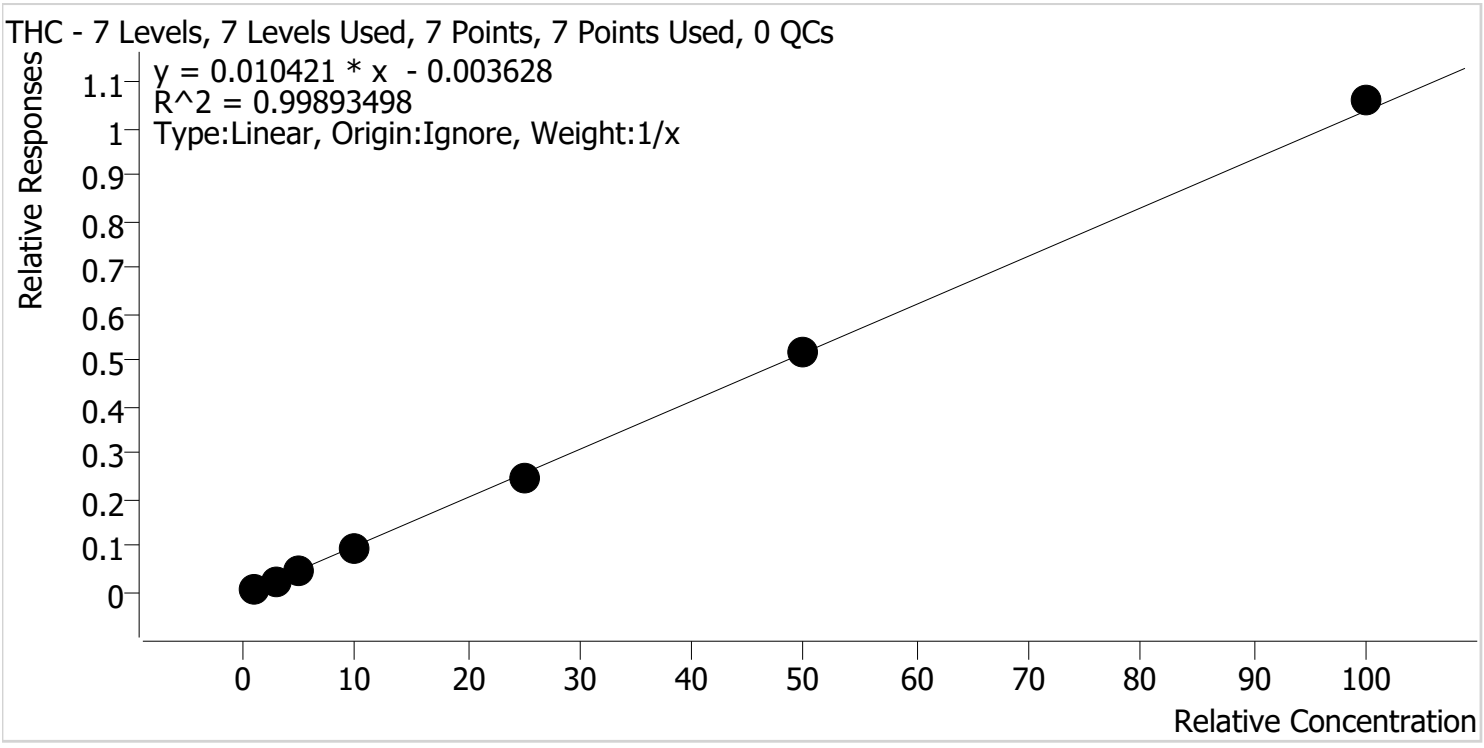
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.513 High	372219	33.27	6.0 Low	∞	2343472	4.9650	ng/ml
THC-COOH	1.489	173458	∞	56.7	1232.94	518375	13.6692	ng/ml
THC	3.330	156740	∞	30.2	∞	3600731	4.5253	ng/ml

AM #27 Cannabinoids Quant. Calibration Curve Report

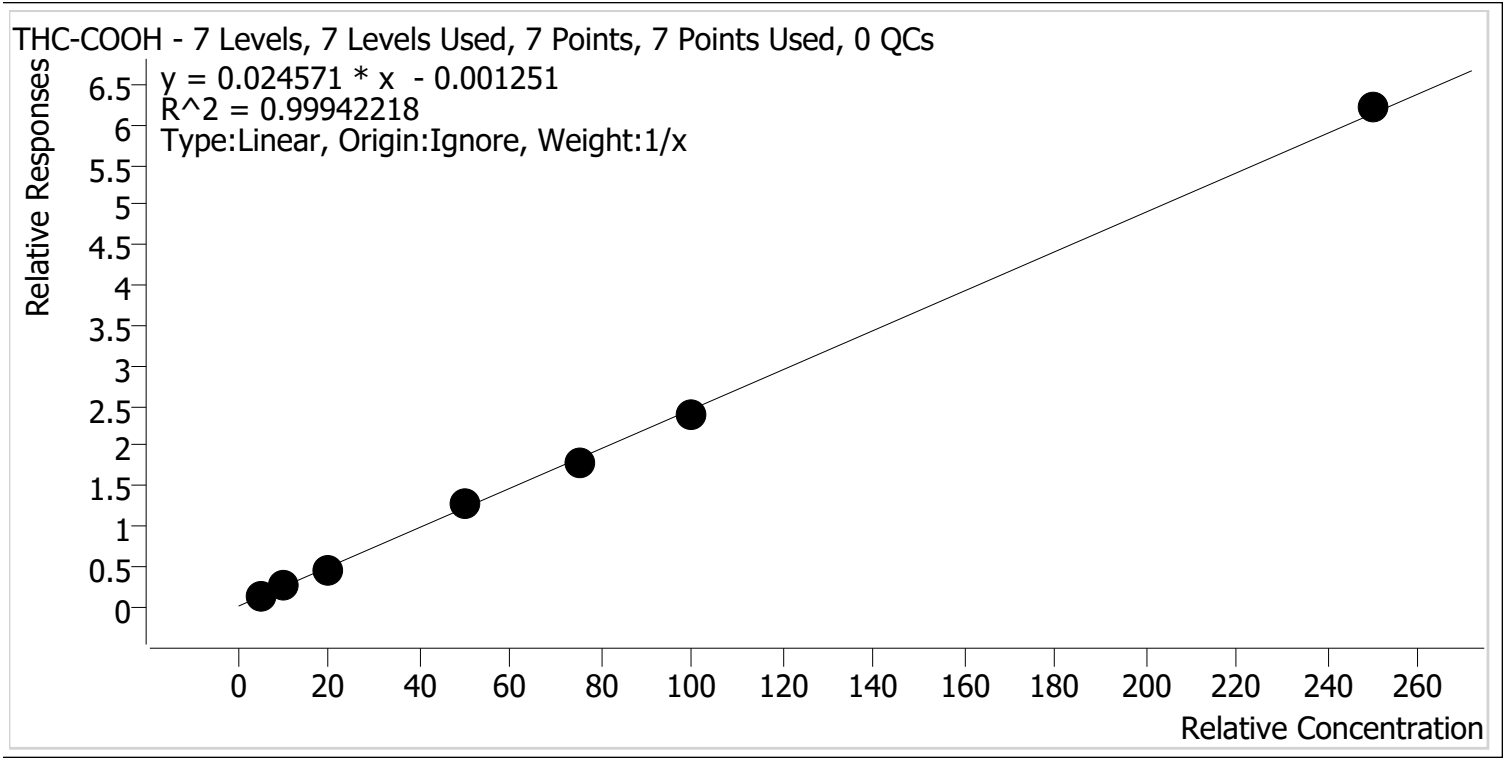
Batch results	D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update	7/22/2021 10:06 AM
Analyst Name	ISP\datastor
Analyte	THC
Internal Standard	THC-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJQ_Cal 1	1	✔	1.0	1.2	116.8
MJQ_Cal 2	2	✔	3.0	2.9	98.1
MJQ_Cal 3	3	✔	5.0	4.7	93.7
MJQ_Cal 4	4	✔	10.0	9.4	93.9
MJQ_Cal 5	5	✔	25.0	24.0	95.8
MJQ_Cal 6	6	✔	50.0	49.7	99.5
MJQ_Cal 7	7	✔	100.0	102.1	102.1

AM #27 Cannabinoids Quant. Calibration Curve Report

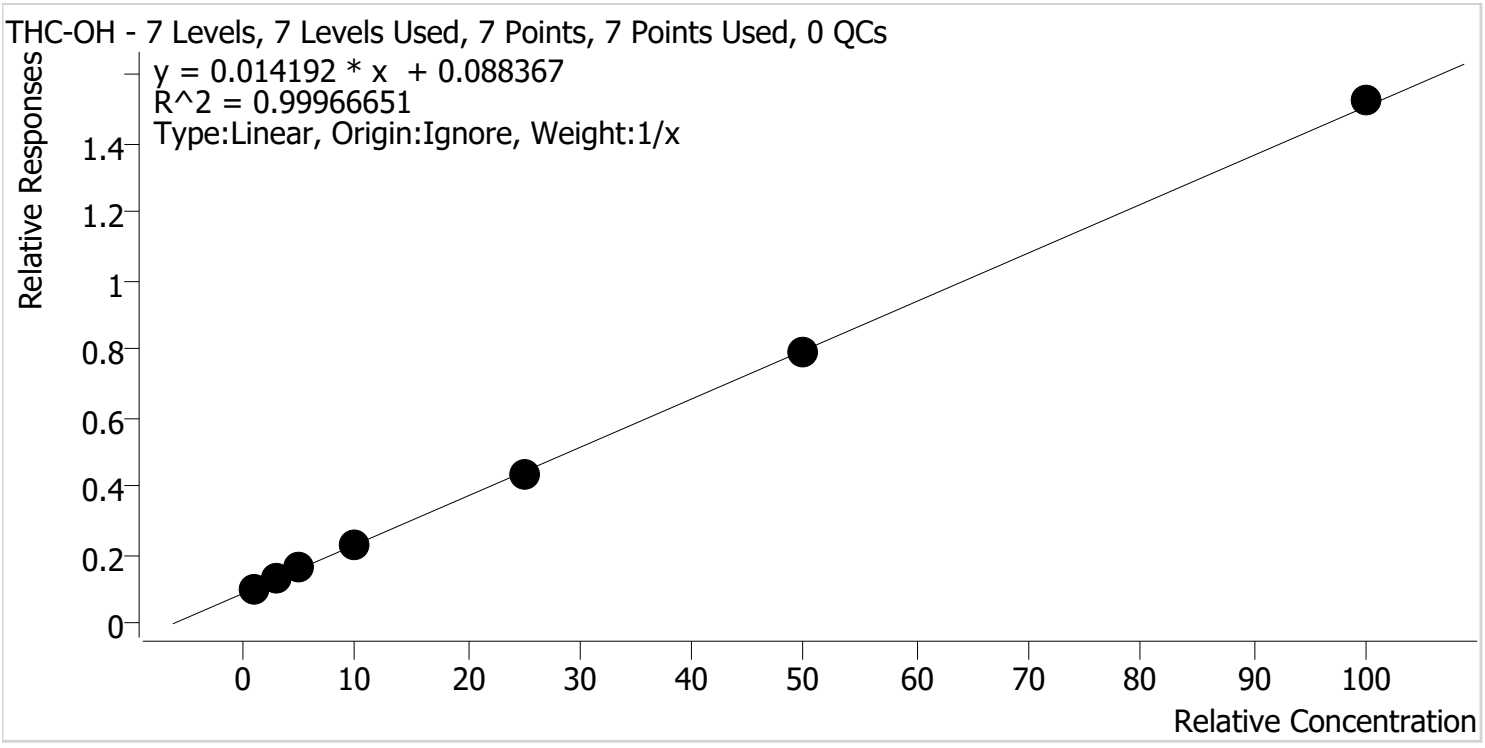
Batch results	D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update	7/22/2021 10:06 AM
Analyst Name	ISP\datastor
Analyte	THC-COOH
Internal Standard	THC-COOH-D9



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJQ_Cal 1	1	✔	5.0	5.3	106.4
MJQ_Cal 2	2	✔	10.0	9.9	98.5
MJQ_Cal 3	3	✔	20.0	19.0	95.1
MJQ_Cal 4	4	✔	50.0	51.4	102.9
MJQ_Cal 5	5	✔	75.0	73.6	98.1
MJQ_Cal 6	6	✔	100.0	97.8	97.8
MJQ_Cal 7	7	✔	250.0	253.0	101.2

AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results	D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update	7/22/2021 10:06 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	1.0	99.1
MJQ_Cal 2	2	✔	3.0	3.1	104.2
MJQ_Cal 3	3	✔	5.0	5.1	101.7
MJQ_Cal 4	4	✔	10.0	9.8	97.7
MJQ_Cal 5	5	✔	25.0	24.2	96.9
MJQ_Cal 6	6	✔	50.0	49.6	99.1
MJQ_Cal 7	7	✔	100.0	101.2	101.2

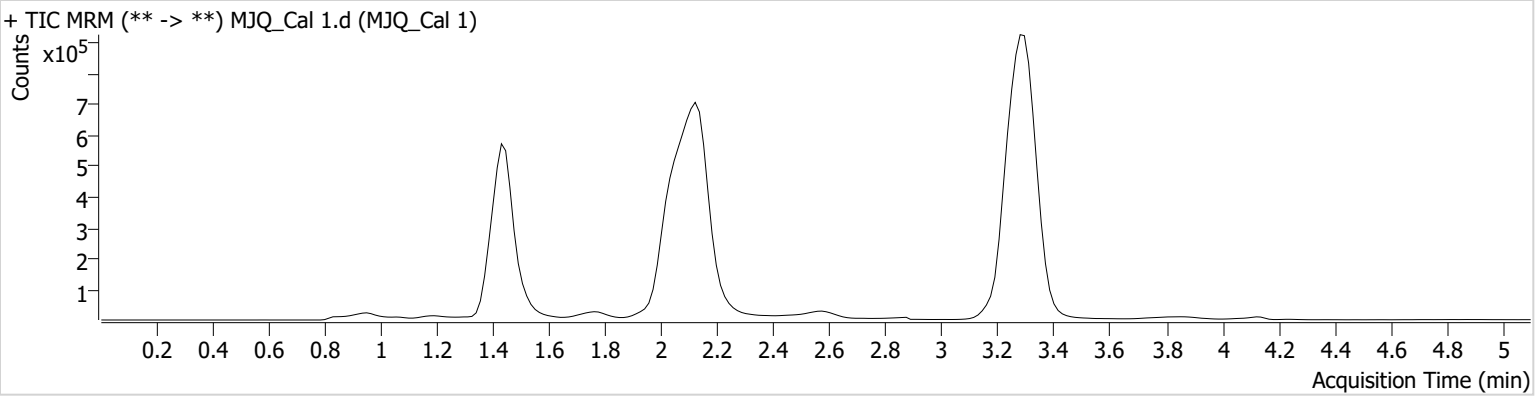
*Did not evaluate

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 7/22/2021 10:06:47 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 1.d
Type	Cal	**Sample**	MJQ_Cal 1
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-A4	**Comment**	
Injection Volume	10		
Acq. Date-Time	7/21/2021 2:40:43 PM		
Sample Info.			

Sample Chromatogram



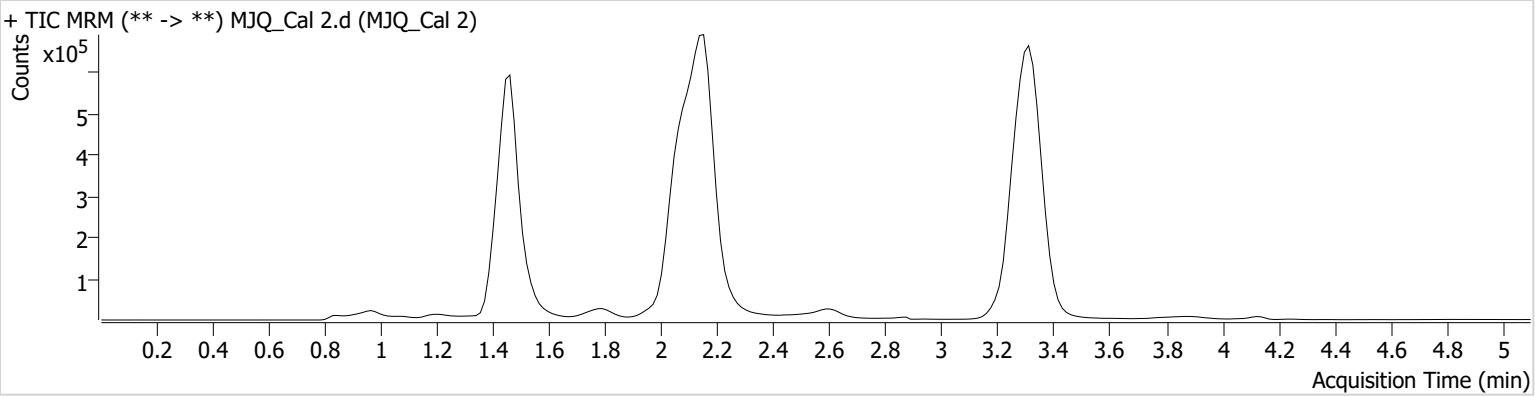
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.513 High	229396	∞	3.5 Low	∞	2239472	0.9911	ng/ml Low
THC-COOH	1.474	78490	∞	46.0	∞	606351	5.3191	ng/ml
THC	3.300	59605	522.75	32.2	∞	6975973	1.1680	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 7/22/2021 10:06:47 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 2.d
Type	Cal	**Sample**	MJQ_Cal 2
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-B4	**Comment**	
Injection Volume	10		
Acq. Date-Time	7/21/2021 2:48:28 PM		
Sample Info.			

Sample Chromatogram



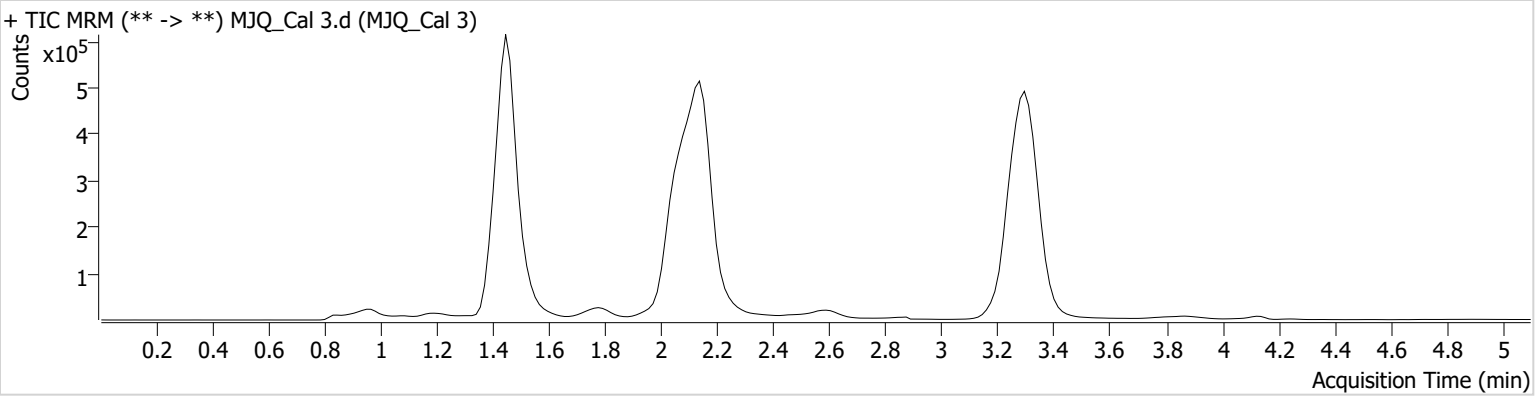
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.528 High	285729	∞	4.8 Low	43.53	2152335	3.1275	ng/ml
THC-COOH	1.489	139356	169.52	52.7	∞	578662	9.8520	ng/ml
THC	3.330	128895	662.13	28.3	∞	4763910	2.9445	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 7/22/2021 10:06:47 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 3.d
Type	Cal	**Sample**	MJQ_Cal 3
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-C4	**Comment**	
Injection Volume	10		
Acq. Date-Time	7/21/2021 2:56:04 PM		
Sample Info.			

Sample Chromatogram



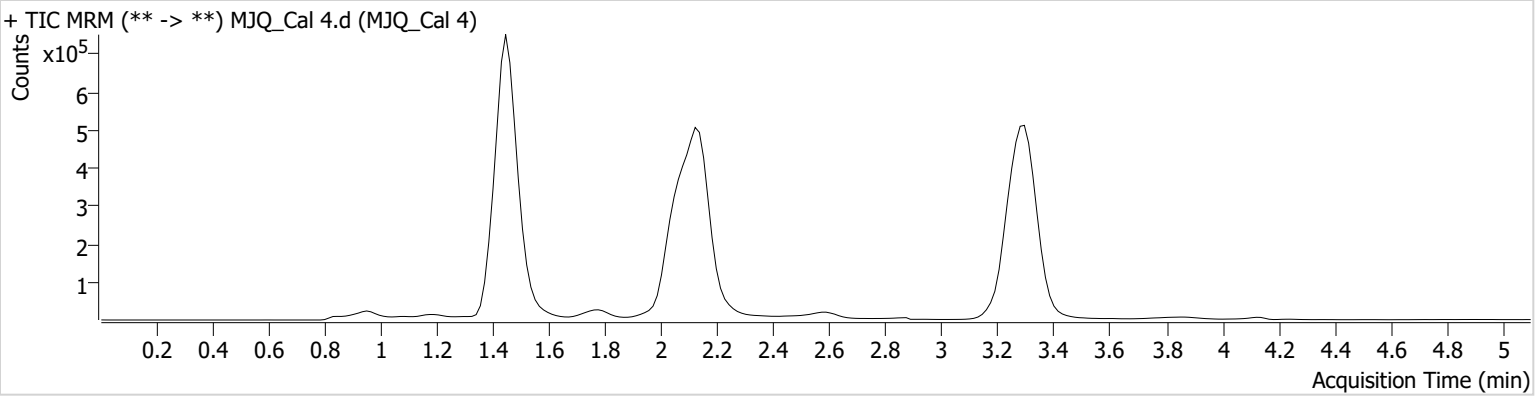
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	340213	∞	6.1	∞	2119572	5.0832	ng/ml
THC-COOH	1.474	216979	171.66	54.3	724.51	465382	19.0259	ng/ml
THC	3.300	159086	485.97	29.5	∞	3519772	4.6853	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 7/22/2021 10:06:47 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 4.d
Type	Cal	**Sample**	MJQ_Cal 4
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-D4	**Comment**	
Injection Volume	10		
Acq. Date-Time	7/21/2021 3:03:40 PM		
Sample Info.			

Sample Chromatogram



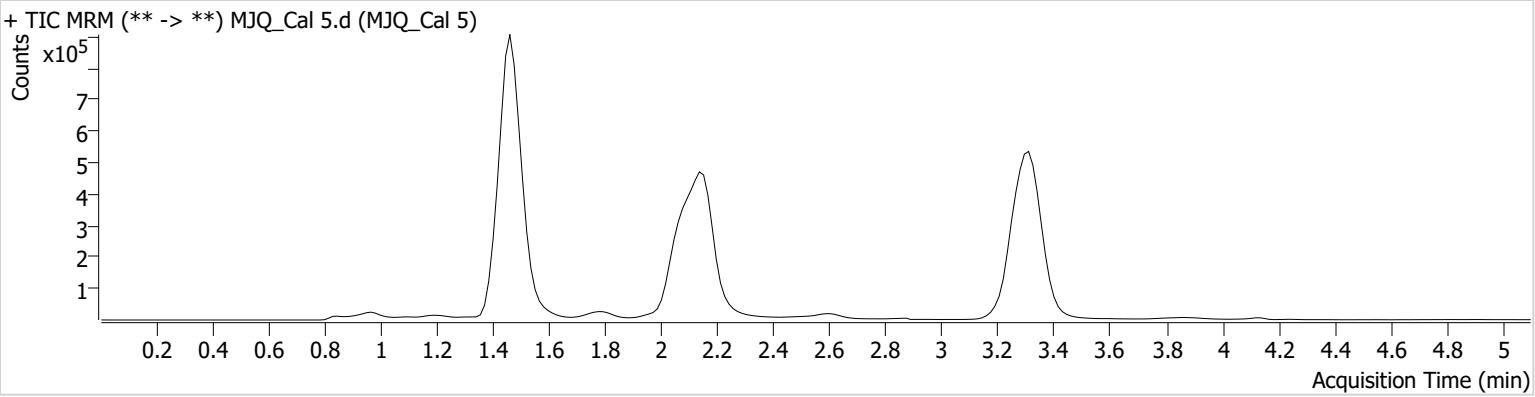
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.453	495026	∞	7.2	970.68	2180620	9.7690	ng/ml
THC-COOH	1.474	601413	∞	53.2	748.66	476306	51.4388	ng/ml
THC	3.315	325611	∞	27.9	482.91	3453837	9.3948	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 7/22/2021 10:06:47 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 5.d
Type	Cal	**Sample**	MJQ_Cal 5
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-E4	**Comment**	
Injection Volume	10		
Acq. Date-Time	7/21/2021 3:11:15 PM		
Sample Info.			

Sample Chromatogram



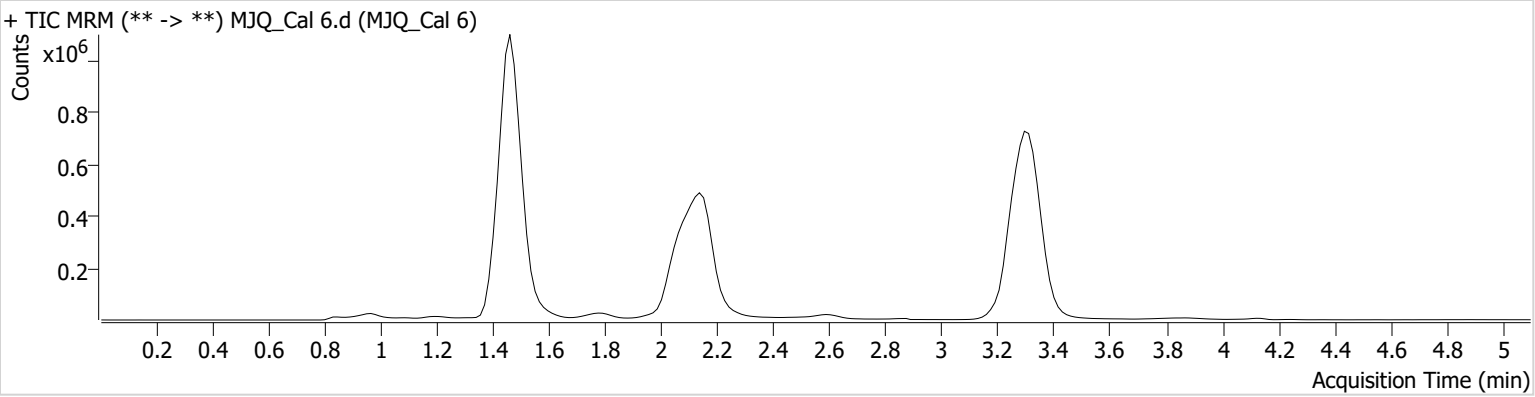
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	913701	∞	9.7 **High**	865.87	2113714	24.2317	ng/ml
THC-COOH	1.489	828798	1047.74	56.6	∞	458720	73.5825	ng/ml
THC	3.315	762264	2570.52	26.4	∞	3098891	23.9524	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 7/22/2021 10:06:47 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 6.d
Type	Cal	**Sample**	MJQ_Cal 6
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-F4	**Comment**	
Injection Volume	10		
Acq. Date-Time	7/21/2021 3:18:50 PM		
Sample Info.			

Sample Chromatogram



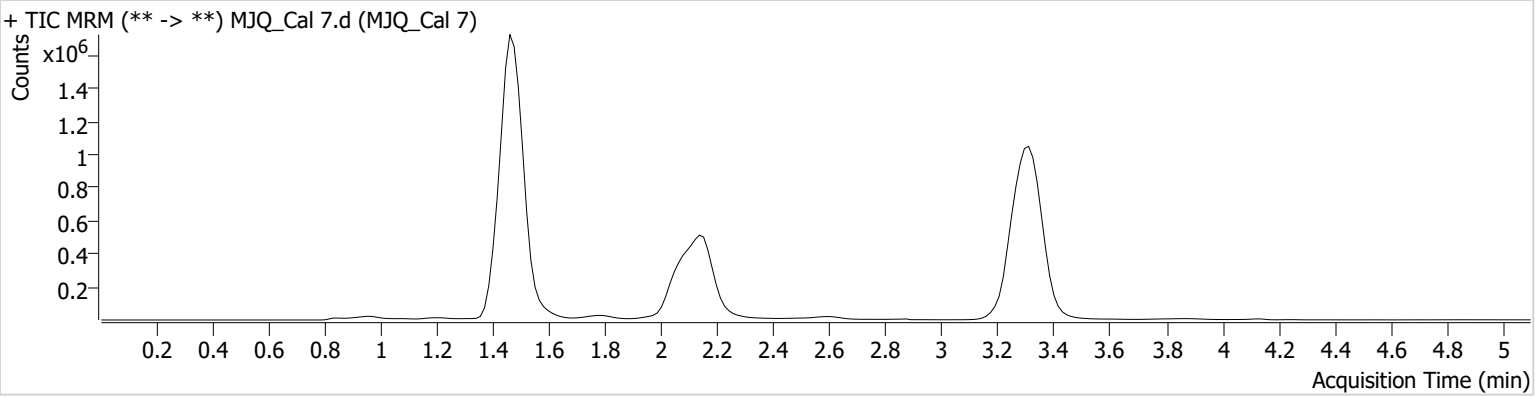
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.468	1599986	∞	10.6 **High**	∞	2020933	49.5575	ng/ml
THC-COOH	1.489	1056698	1170.99	57.5	∞	440051	97.7794	ng/ml
THC	3.315	1740825	2753.65	25.5	1196.50	3382753	49.7309	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\072121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 7/22/2021 10:06:47 AM

Instrument	Falco (069901)	**Data File**	MJQ_Cal 7.d
Type	Cal	**Sample**	MJQ_Cal 7
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Collins
Sample Position	P1-G4	**Comment**	
Injection Volume	10		
Acq. Date-Time	7/21/2021 3:26:26 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.453	2873837	∞	11.1 **High**	∞	1884230	101.2400	ng/ml
THC-COOH	1.489	2510821	∞	57.7	∞	403973	253.0023	ng/ml
THC	3.315	3693288	∞	25.7	∞	3482243	102.1241	ng/ml