REVIEWED
By Anne Nord at 3:32 pm, Nov 20, 2020

11/20/2020

Worklist: 4616

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION
M2020-3661	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-3765	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-3973	2	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-4219	1	BCK	AM 27 Blood THC Quant by LC-QQQ
M2020-4262	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-2986	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3119	4	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3139	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3188	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3202	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3204	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3206	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3207	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3225	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3231	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3233	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3236	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3239	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3240	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3241	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3242	1	BCK	AM 27 Blood THC Quant by LC-QQQ

Worklist: 4616

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION
P2020-3244	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3257	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2020-3290	1	BCK	AM 27 Blood THC Quant by LC-QQQ

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

Extraction Date: 11/19/20 Analyst: Sarah Pickle
Plate lot#: IDP-108-2-200723 Plate Expiration: 01/23/21

Mobile phase A: 0.1% Formic Acid in LCMS Water **Mobile phase B:** 0.1% Formic acid in Acetonitrile
MTBE LCMS Methanol Hexane
Blank Blood Lot: Lampire 20K20702 **Column**: UCT Selectra DA 100 x 2.1mm 3um
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. Pipette **1000 µL blood (calibrated pipette)** in wells of analytical (standards) plate. **Pipette ID: 3382167**
☒ 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
☒ 4. Pipette **500 µL 0.1% formic acid in LCMS water** in wells of analytical plate.
☒ 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
☒ 6. Transfer **800 µL of blood+acid** mixture to corresponding wells of SLE+ plate.
☒ 7. Apply positive pressure for approx. 10-15 seconds (or until no liquid remains on top of sorbent). ***(Load at 85-95 PSI- Selector to the right)***
☒ 8. Wait 5 minutes.
☒ 9. Add **2.25 mL MTBE. (*Add in 3 increments of 750 µL)***
☒ 10. Wait 5 minutes.
☒ 11. Apply positive pressure for approx. 15 seconds. ***(12-15 PSI- Selector to the left).***
☒ 12. Add **2.25 mL Hexane. (*Add in 3 increments of 750 µL)***
☒ 13. Wait 5 minutes.
☒ 14. Apply positive pressure for approx. 15 seconds. ***(12-15 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 **100 µL 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 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? Y / N
☒ 6 Enter QCs into control charting.
☒ 7 Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

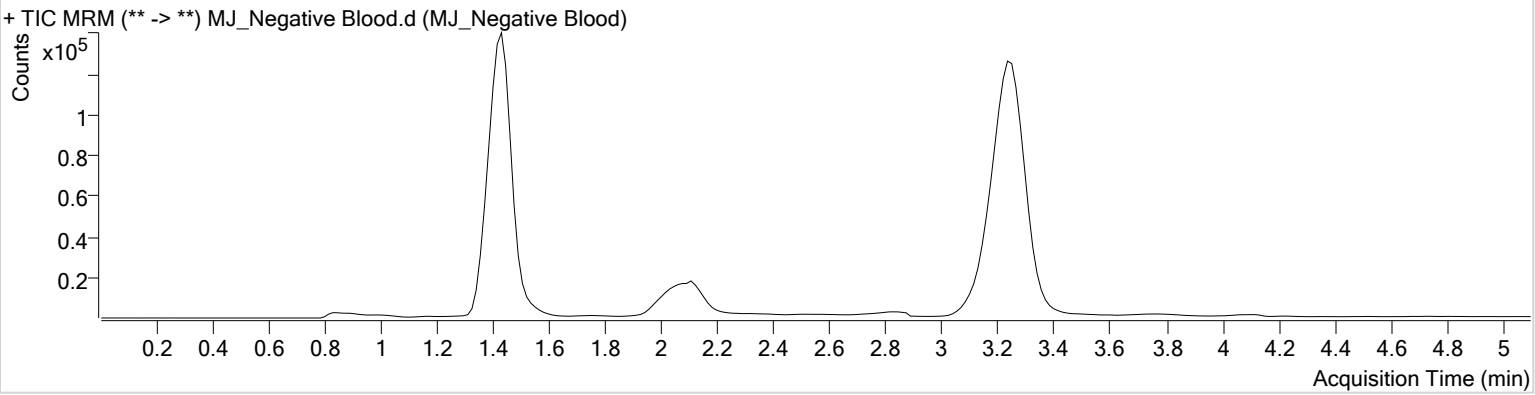
COMMENTS:

AM #27 Cannabinoid Quant. Results

Batch results	D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin
Calibration Last Update	11/20/2020 12:39:05 PM

Instrument	Instrument 1	**Data File**	MJ_Negative Blood.d
Type	Sample	**Sample**	MJ_Negative Blood
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Pickle
Sample Position	P1-A2	**Comment**	
Injection Volume	10		
Acq. Date-Time	11/19/2020 11:56:53 PM		
Sample Info.			

Sample Chromatogram

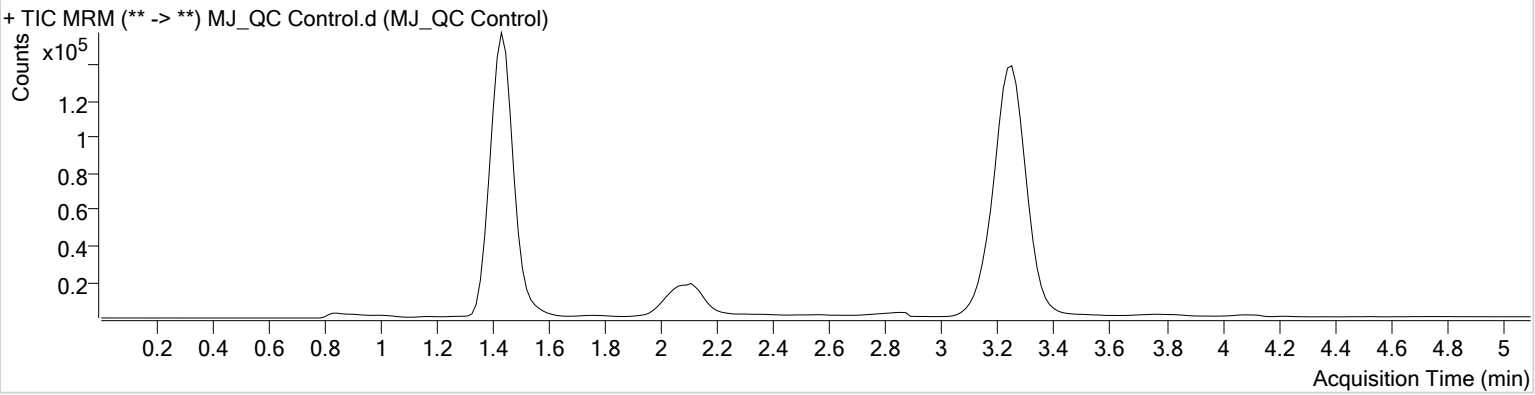


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin
Calibration Last Update 11/20/2020 12:39:05 PM

Instrument	Instrument 1	**Data File**	MJ_QC Control.d
Type	Sample	**Sample**	MJ_QC Control
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Pickle
Sample Position	P1-H1	**Comment**	
Injection Volume	10		
Acq. Date-Time	11/19/2020 11:41:42 PM		
Sample Info.			

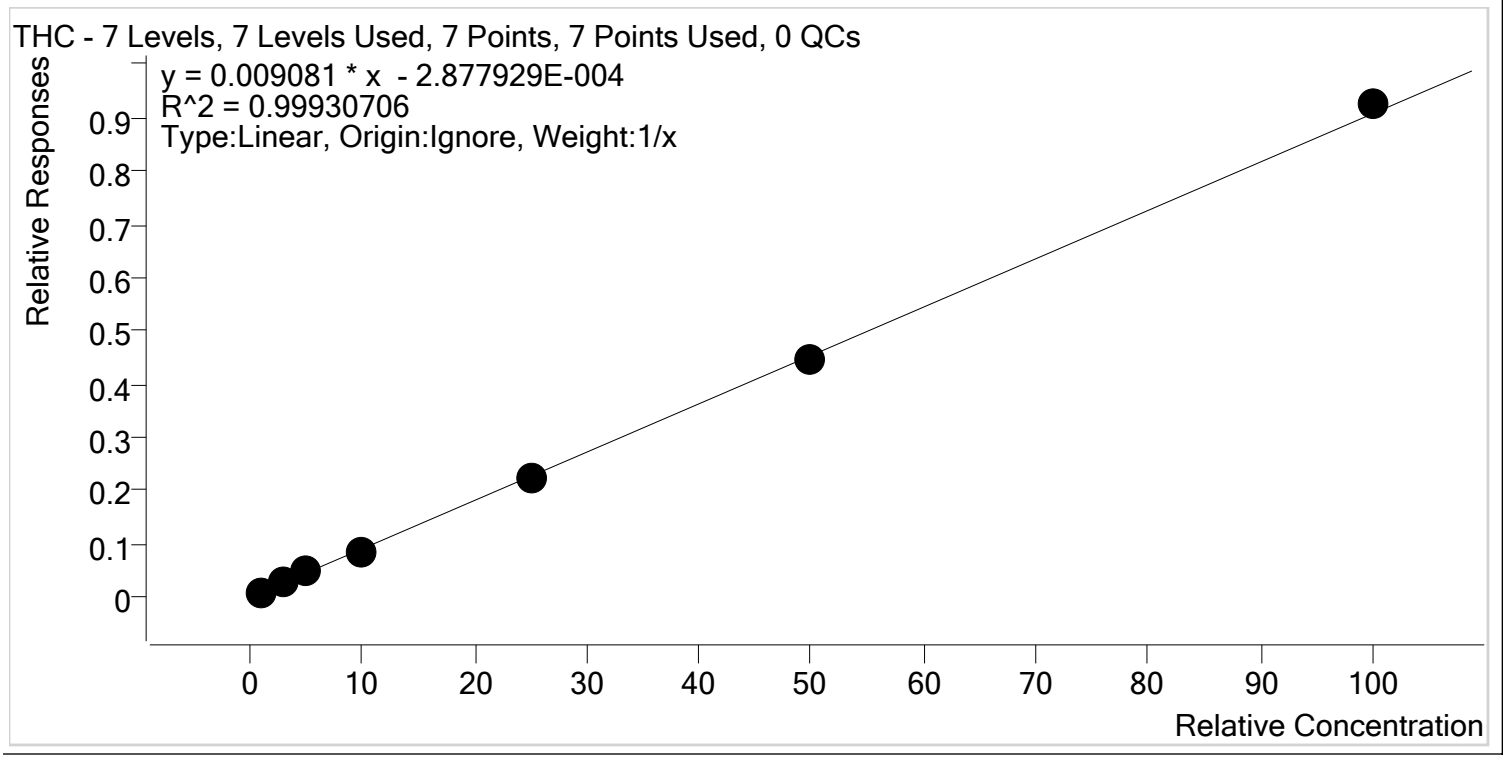
Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.438	38648	∞	14.3	83.26	576924	4.6581	ng/ml
THC-COOH	1.459	64283	∞	68.3	1031.58	161130	14.3812	ng/ml
THC	3.254	45484	329.82	31.4	110.13	1088831	4.6319	ng/ml

AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results	D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin
Last Cal. Update	11/20/2020 12:39 PM
Analyst Name	ISP\datastor
Analyte	THC
Internal Standard	THC-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	✔	1.0	1.0	100.7
MJ_Cal 2	2	✔	3.0	3.1	102.6
MJ_Cal 3	3	✔	5.0	5.2	104.6
MJ_Cal 4	4	✔	10.0	9.5	94.9
MJ_Cal 5	5	✔	25.0	24.3	97.3
MJ_Cal 6	6	✔	50.0	49.0	97.9
MJ_Cal 7	7	✔	100.0	101.9	101.9

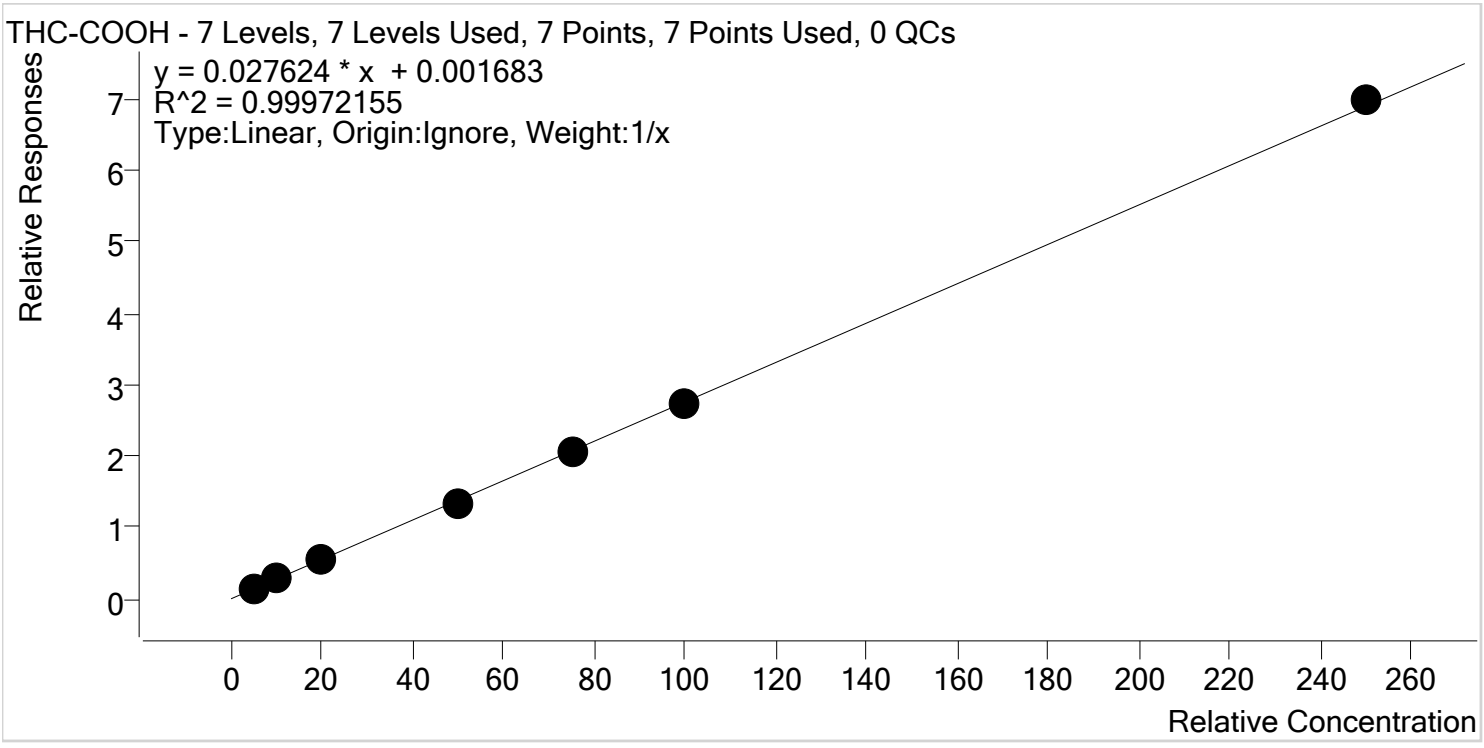
AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin

Last Cal. Update 11/20/2020 12:39 PM

Analyst Name ISP\datastor

Analyte THC-COOH **Internal Standard** THC-COOH-D9



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	✔	5.0	5.2	104.7
MJ_Cal 2	2	✔	10.0	10.0	99.5
MJ_Cal 3	3	✔	20.0	19.7	98.7
MJ_Cal 4	4	✔	50.0	48.9	97.8
MJ_Cal 5	5	✔	75.0	74.8	99.8
MJ_Cal 6	6	✔	100.0	98.3	98.3
MJ_Cal 7	7	✔	250.0	253.1	101.2

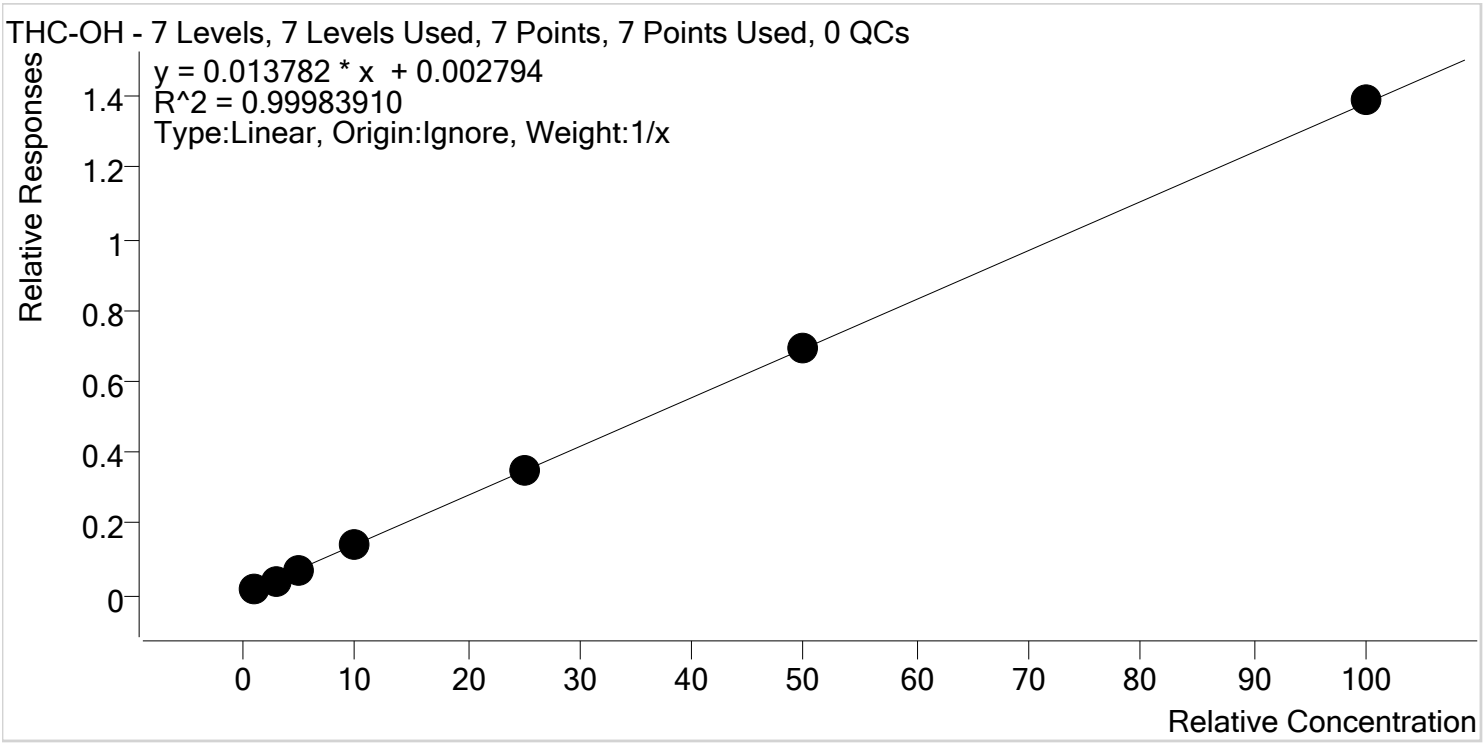
AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin

Last Cal. Update 11/20/2020 12:39 PM

Analyst Name ISP\datastor

Analyte THC-OH **Internal Standard** THC-OH-D3



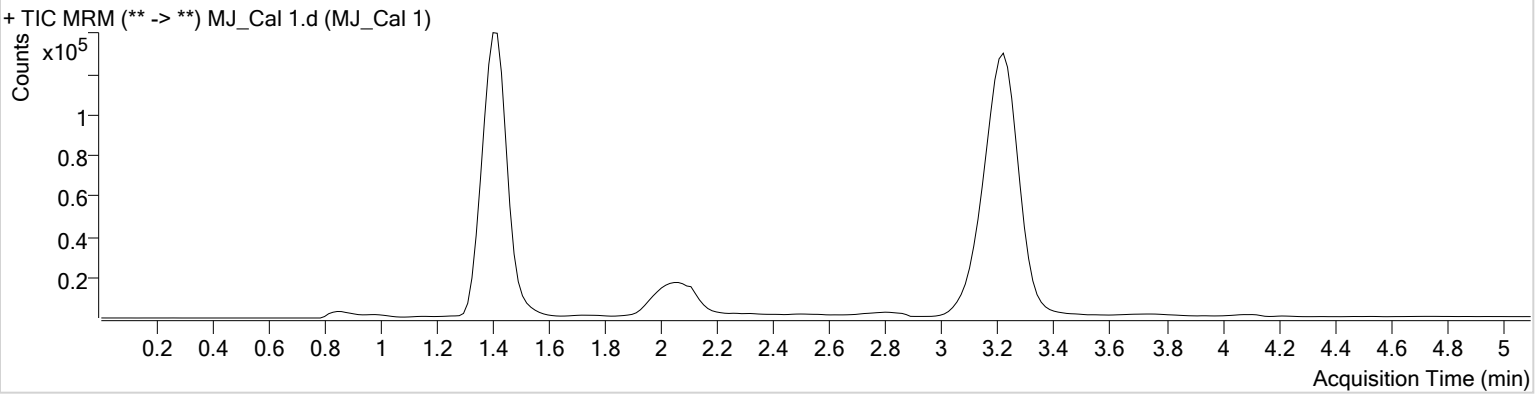
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	✔	1.0	1.1	108.6
MJ_Cal 2	2	✔	3.0	2.8	94.2
MJ_Cal 3	3	✔	5.0	5.0	99.5
MJ_Cal 4	4	✔	10.0	9.8	97.8
MJ_Cal 5	5	✔	25.0	24.9	99.7
MJ_Cal 6	6	✔	50.0	49.8	99.7
MJ_Cal 7	7	✔	100.0	100.6	100.6

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin
Calibration Last Update 11/20/2020 12:39:05 PM

Instrument	Instrument 1	**Data File**	MJ_Cal 1.d
Type	Cal	**Sample**	MJ_Cal 1
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Pickle
Sample Position	P1-A1	**Comment**	
Injection Volume	10		
Acq. Date-Time	11/19/2020 10:40:53 PM		
Sample Info.			

Sample Chromatogram



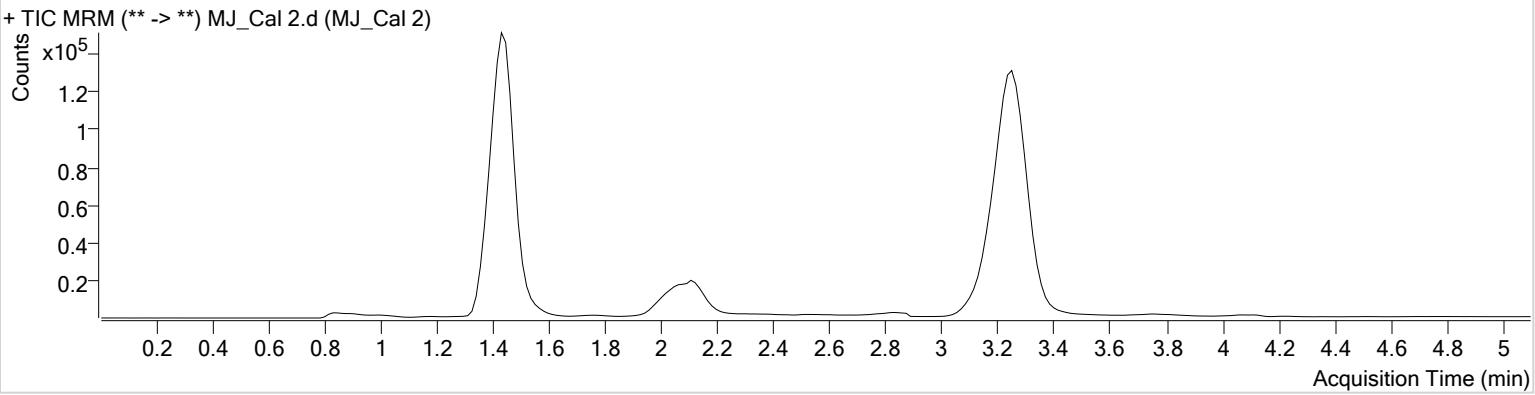
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.		
THC-OH	1.423	11299	∞	11.5	23.31	636296	1.0857	ng/ml	Low
THC-COOH	1.444	26211	∞	60.2	∞	179126	5.2361	ng/ml	
THC	3.209	9985	53.16	35.2	12.12	1126976	1.0073	ng/ml	Low

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin
Calibration Last Update 11/20/2020 12:39:05 PM

Instrument	Instrument 1	**Data File**	MJ_Cal 2.d
Type	Cal	**Sample**	MJ_Cal 2
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Pickle
Sample Position	P1-B1	**Comment**	
Injection Volume	10		
Acq. Date-Time	11/19/2020 10:48:38 PM		
Sample Info.			

Sample Chromatogram



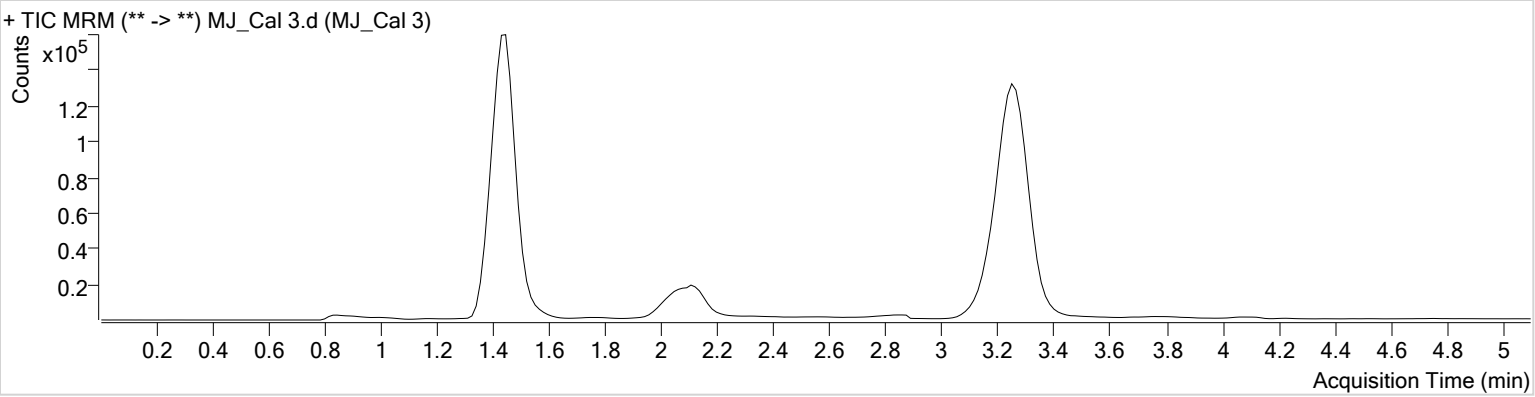
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.		
THC-OH	1.438	26149	∞	13.7	30.68	626147	2.8275	ng/ml	Low
THC-COOH	1.474	48002	∞	65.2	250.94	173564	9.9509	ng/ml	
THC	3.254	30041	281.09	36.4	48.05	1085611	3.0790	ng/ml	

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin
Calibration Last Update 11/20/2020 12:39:05 PM

Instrument	Instrument 1	**Data File**	MJ_Cal 3.d
Type	Cal	**Sample**	MJ_Cal 3
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Pickle
Sample Position	P1-C1	**Comment**	
Injection Volume	10		
Acq. Date-Time	11/19/2020 10:56:12 PM		
Sample Info.			

Sample Chromatogram



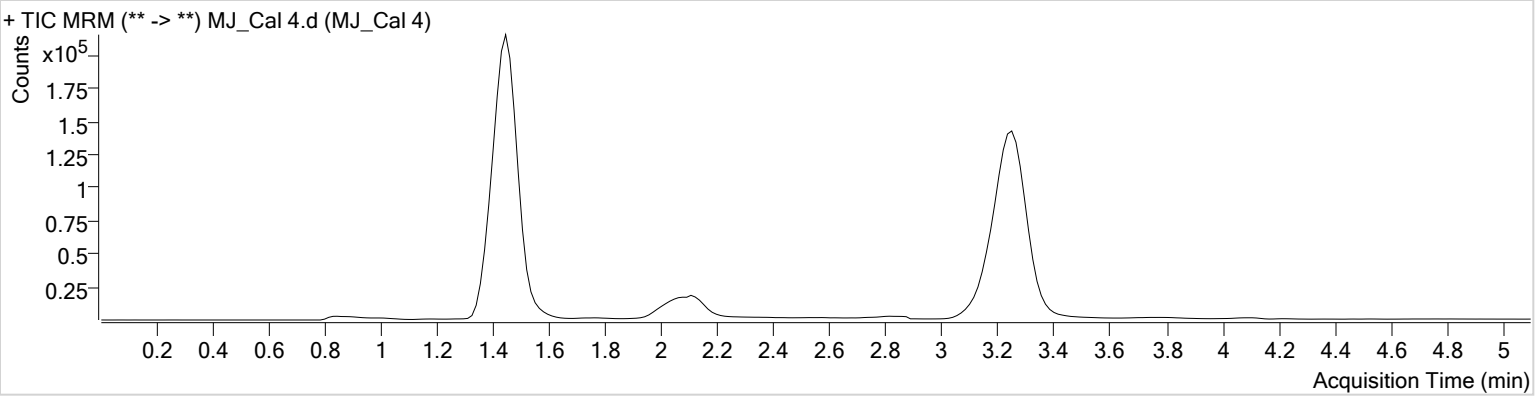
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.438	42293	254.80	13.9	150.20	592755	4.9745	ng/ml
THC-COOH	1.474	91531	∞	65.9	∞	167304	19.7440	ng/ml
THC	3.254	48917	438.23	34.4	∞	1036428	5.2292	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin
Calibration Last Update 11/20/2020 12:39:05 PM

Instrument	Instrument 1	**Data File**	MJ_Cal 4.d
Type	Cal	**Sample**	MJ_Cal 4
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Pickle
Sample Position	P1-D1	**Comment**	
Injection Volume	10		
Acq. Date-Time	11/19/2020 11:03:46 PM		
Sample Info.			

Sample Chromatogram



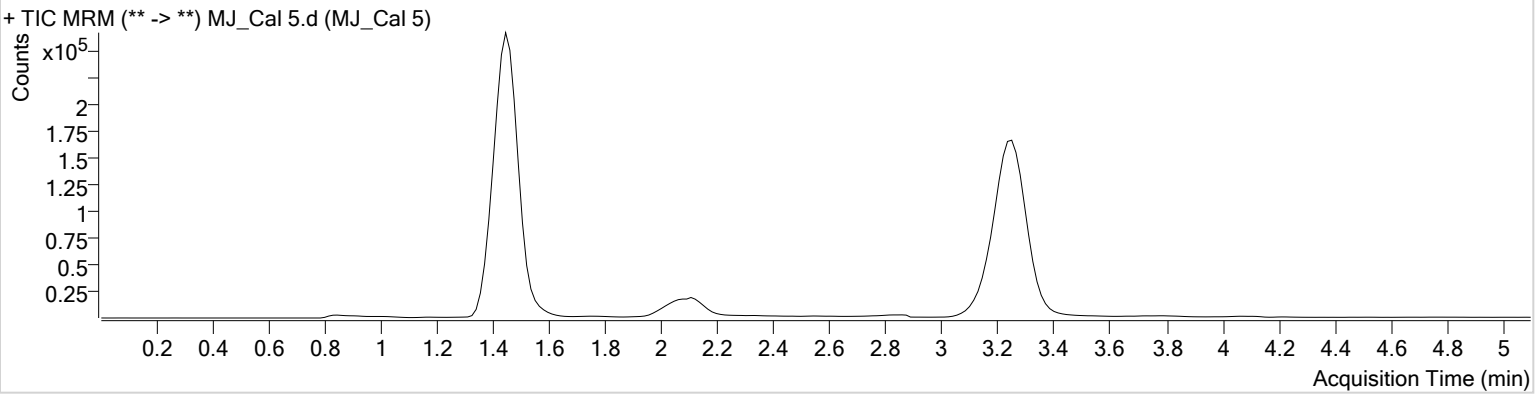
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.438	86136	∞	13.8	128.63	626251	9.7774	ng/ml
THC-COOH	1.474	235944	∞	67.4	3157.80	174475	48.8931	ng/ml
THC	3.254	94849	6953.54	29.8	58.53	1103883	9.4937	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin
Calibration Last Update 11/20/2020 12:39:05 PM

Instrument	Instrument 1	**Data File**	MJ_Cal 5.d
Type	Cal	**Sample**	MJ_Cal 5
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Pickle
Sample Position	P1-E1	**Comment**	
Injection Volume	10		
Acq. Date-Time	11/19/2020 11:11:21 PM		
Sample Info.			

Sample Chromatogram



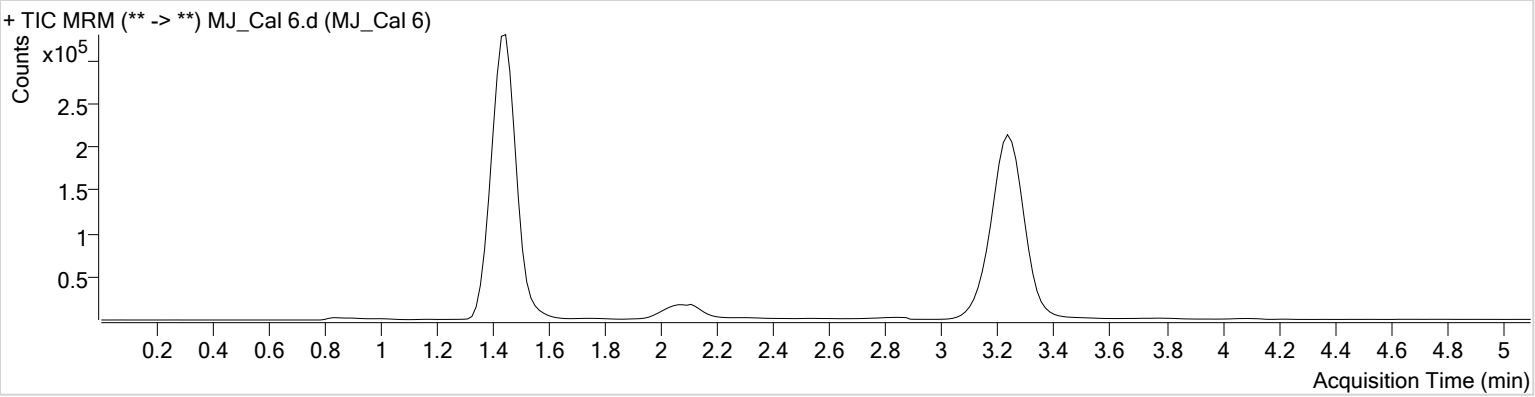
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.438	205346	1393.80	13.9	781.01	593181	24.9160	ng/ml
THC-COOH	1.474	334211	∞	67.7	∞	161573	74.8190	ng/ml
THC	3.254	240740	1140.82	29.2	111.36	1091563	24.3188	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin
Calibration Last Update 11/20/2020 12:39:05 PM

Instrument	Instrument 1	**Data File**	MJ_Cal 6.d
Type	Cal	**Sample**	MJ_Cal 6
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Pickle
Sample Position	P1-F1	**Comment**	
Injection Volume	10		
Acq. Date-Time	11/19/2020 11:18:57 PM		
Sample Info.			

Sample Chromatogram



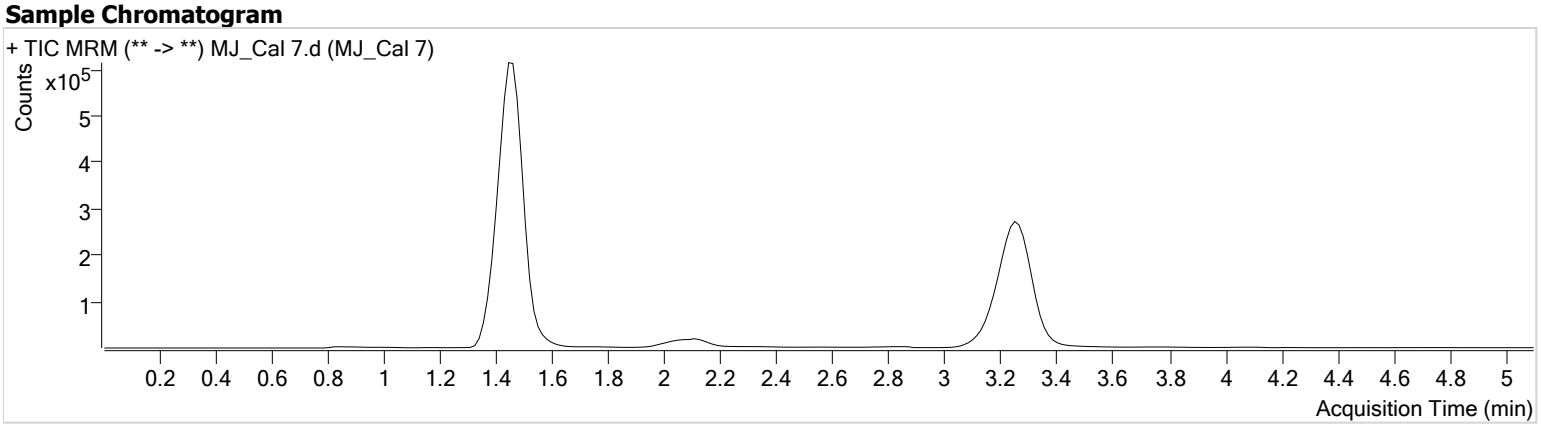
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.438	397800	5015.67	14.0	1004.97	576856	49.8350	ng/ml
THC-COOH	1.459	430010	∞	68.1	12078.61	158308	98.2693	ng/ml
THC	3.254	499744	1576.61	29.6	∞	1124642	48.9656	ng/ml

AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2020\AM 27-28\111920 AM 28 P2 TS SP SJ_AM 28 27 SP\QuantResults\AM 27 SP.batch.bin
Calibration Last Update 11/20/2020 12:39:05 PM

Instrument	Instrument 1	**Data File**	MJ_Cal 7.d
Type	Cal	**Sample**	MJ_Cal 7
Acq. Method	AM 27 THCQ.m	**Operator**	Sarah Pickle
Sample Position	P1-G1	**Comment**	
Injection Volume	10		
Acq. Date-Time	11/19/2020 11:26:31 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-OH	1.438	859006	∞	14.0	2706.83	618434	100.5840	ng/ml
THC-COOH	1.474	1119553	∞	67.5	∞	160096	253.0877	ng/ml
THC	3.254	1003781	1960.48	27.8	1191.70	1085048	101.9064	ng/ml