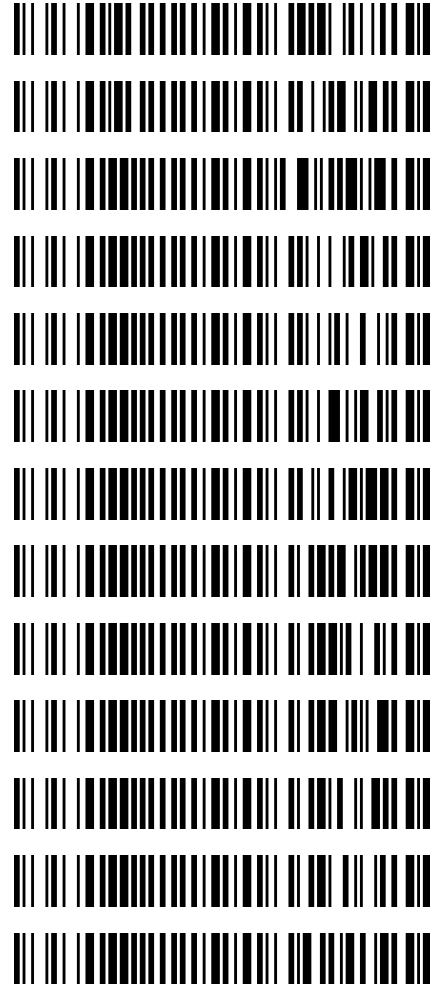


Worklist: 4871

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
M2021-1061	3	BCK	AM 27 Blood THC Quant by LC-QQQ
M2021-1132	3	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0609	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0784	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0785	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0788	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0791	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0833	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0834	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0835	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0839	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0840	1	BCK	AM 27 Blood THC Quant by LC-QQQ
P2021-0846	1	BCK	AM 27 Blood THC Quant by LC-QQQ



80

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

Extraction Date: 03/31/21
 Plate lot#: IDP-108-2-201206

Analyst: Sarah Collins
 Plate Expiration: 06/06/21

Mobile phase A: 0.1% Formic Acid in LCMS Water
 MTBE
Mobile phase B: 0.1% Formic acid in Acetonitrile
 Hexane
Blank Blood Lot: Lampire 20L20724
LCMS-QQQ ID: 069901
Column: UCT Selectra DA 100 x 2.1mm 3um
 LCMS Methanol

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 r² 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? Y / N
- 6. Enter QCs into control charting.
- 7. Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS: *Curve range limited: THC 3-100 THC-OH 3-100*

*Reinjected Calibrator 3 and M2021-1132-3 due low internal standard response in initial injection. Reinject data used.
 Case sample P2021-0438-1 was ran with this batch due to varying results between screen and quant in initial data.*

**Idaho State Police
Forensic Services
Toxicology Discipline**

Request for Departure from an Analytical Method

Date of Request

02/24/2021

Forensic Scientist

Anne Nord

Analytical Method

Toxicology AM #27: Quantitative Analysis of THC and Metabolites in Blood and Urine by LCMS-QQQ

Request

The method currently reads:

4.3.2.5 If any points are dropped from the approved quantitative range of the curve, the compound will be reported qualitatively. For calibrators and controls 10 ng and below, the accuracy must be within 30%, for calibrators and controls greater than 10 ng/mL the accuracy must be within 20%. If a control falls outside the accuracy range, at the analyst's discretion, the compound may be reported qualitatively.

I would like to add in the following exception:

If the 1ng/ml point is dropped for THC. If the 1 ng/ml point is dropped the quantitative range will be 3-50 ng/ml.

Discipline Leader Review

Departure approved

Comments: This deviation is approved and will remain in effect until it is changed in the actual method.

Departure Not Approved

Comments:



Celena Shrum

Toxicology Discipline Lead

Date: 02/24/2021

8C

	1	2	3	4	5	6
A	IS + Cal. 1	negative	p2021-0833-1*	p2021-0785-1		IS + QC_1
B	IS + Cal. 2	m2021-1061-3*	p2021-0834-1	p2021-0833-1		IS + Cal. 7
C	IS + Cal. 3	m2021-1132-3	p2021-0835-1	p2021-0839-1		IS + Cal. 6
D	IS + Cal. 4	p2021-0609-1	p2021-0839-1*			IS + Cal. 5
E	IS + Cal. 5	p2021-0784-1	p2021-0840-1			IS + Cal. 4
F	IS + Cal. 6	p2021-0785-1*	p2021-0846-1			IS + Cal. 3
G	IS + Cal. 7	p2021-0788-1	p2021-0438-1			IS + Cal. 2
H	IS + QC_1	p2021-0791-1	m2021-1061-3			IS + Cal. 1

All wells to contain 100 µl of residual DMSO

*Samples moved during analytical step 6 due to blood clot

SC



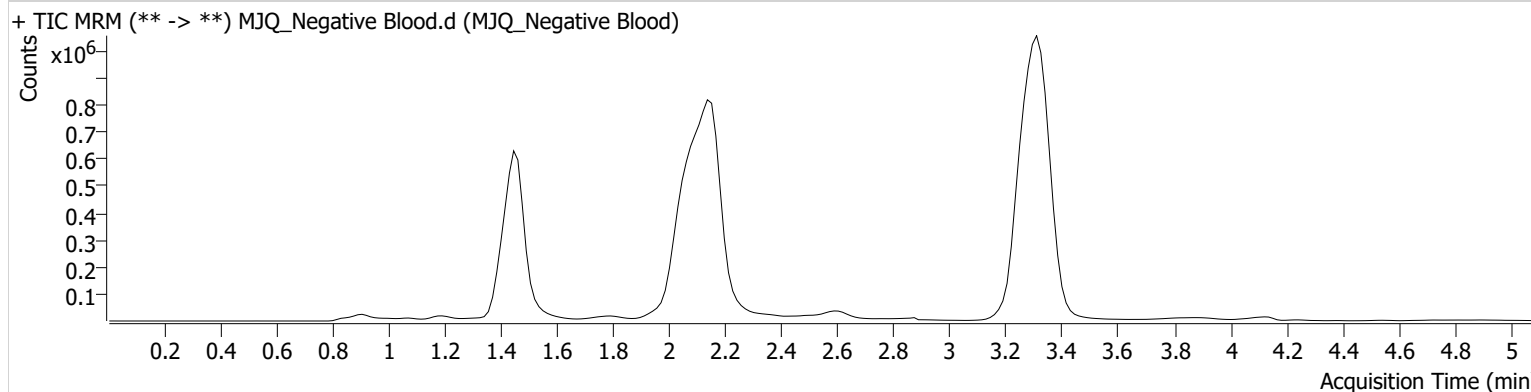
AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 4/1/2021 10:21:04 AM

Instrument	Instrument 1	Data File	MJQ_Negative Blood.d
Type	Sample	Sample	MJQ_Negative Blood
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-A2	Comment	
Injection Volume	10		
Acq. Date-Time	3/31/2021 12:04:29 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.384 Low	31650	∞			711618	1.0233 ng/ml Low

SC

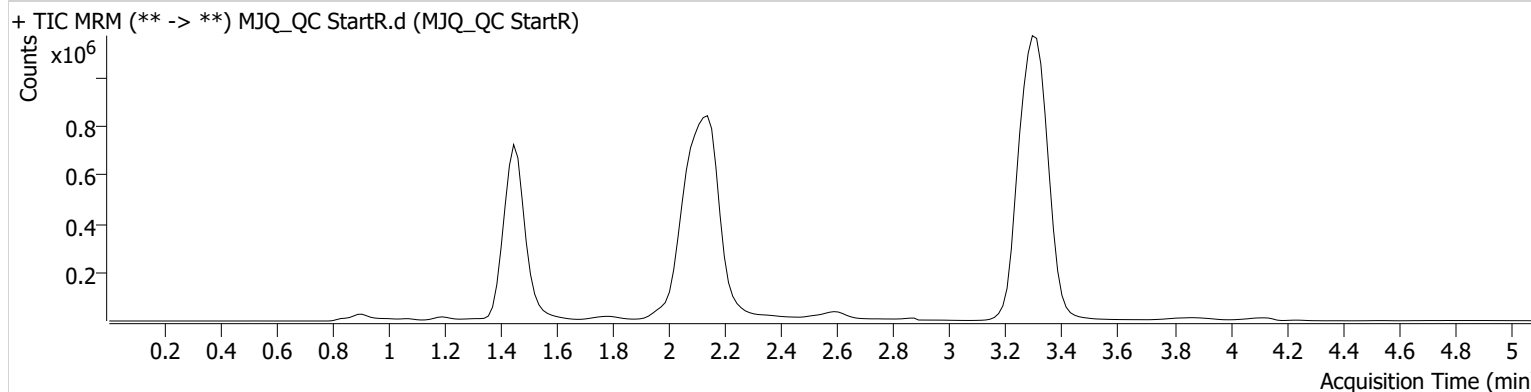


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 4/1/2021 10:21:04 AM

Instrument	Instrument 1	Data File	MJQ_QC StartR.d
Type	Sample	Sample	MJQ_QC StartR
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-H1	Comment	**Not a reinject. Original injection was the incorrect vial position
Injection Volume	10		
Acq. Date-Time	3/31/2021 1:05:19 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.453	183607	∞	9.1	102.59	2530194	3.9777 ng/ml
THC-COOH	1.489	256538	∞	51.4	∞	675128	14.7125 ng/ml
THC	3.315	293473	∞	30.3	194.29	8270204	4.1444 ng/ml

SC

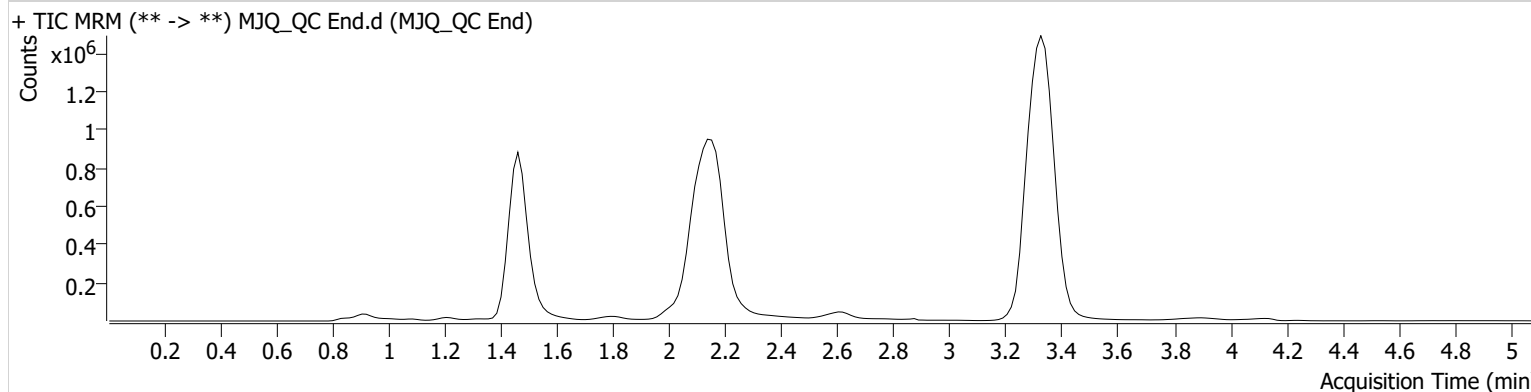


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 4/1/2021 10:21:04 AM

Instrument	Instrument 1	Data File	MJQ_QC End.d
Type	Sample	Sample	MJQ_QC End
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-H1	Comment	
Injection Volume	10		
Acq. Date-Time	3/31/2021 4:38:22 PM		

Sample Chromatogram



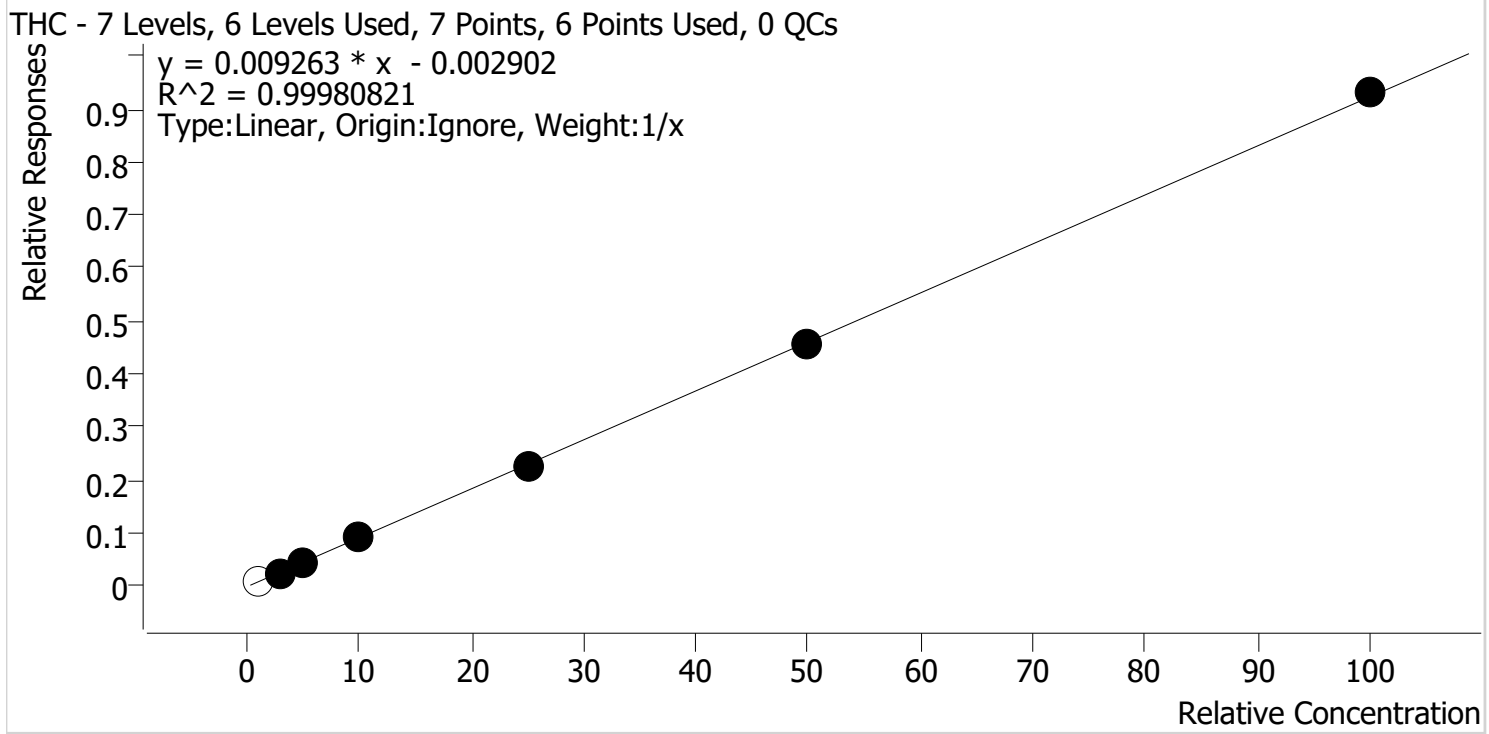
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.468	198361	∞	9.4	∞	2882590	3.6954 ng/ml
THC-COOH	1.489	245401	∞	56.4	∞	726687	12.9871 ng/ml
THC	3.345	354742	1631.99	27.4	177.79	9762838	4.2362 ng/ml

SC



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update 4/1/2021 10:21 AM
Analyst Name ISP\Datastor
Analyte THC **Internal Standard** THC-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJQ_Cal 1	1	x	1.0	1.2	124.2
MJQ_Cal 2	2	✓	3.0	3.0	99.1
MJQ_Cal 3R	3	✓	5.0	5.1	102.2
MJQ_Cal 4	4	✓	10.0	10.1	101.0
MJQ_Cal 5	5	✓	25.0	24.5	97.9
MJQ_Cal 6	6	✓	50.0	49.5	99.0
MJQ_Cal 7	7	✓	100.0	100.9	100.9

Calibrator 1 dropped due to not meeting ratio requirement.

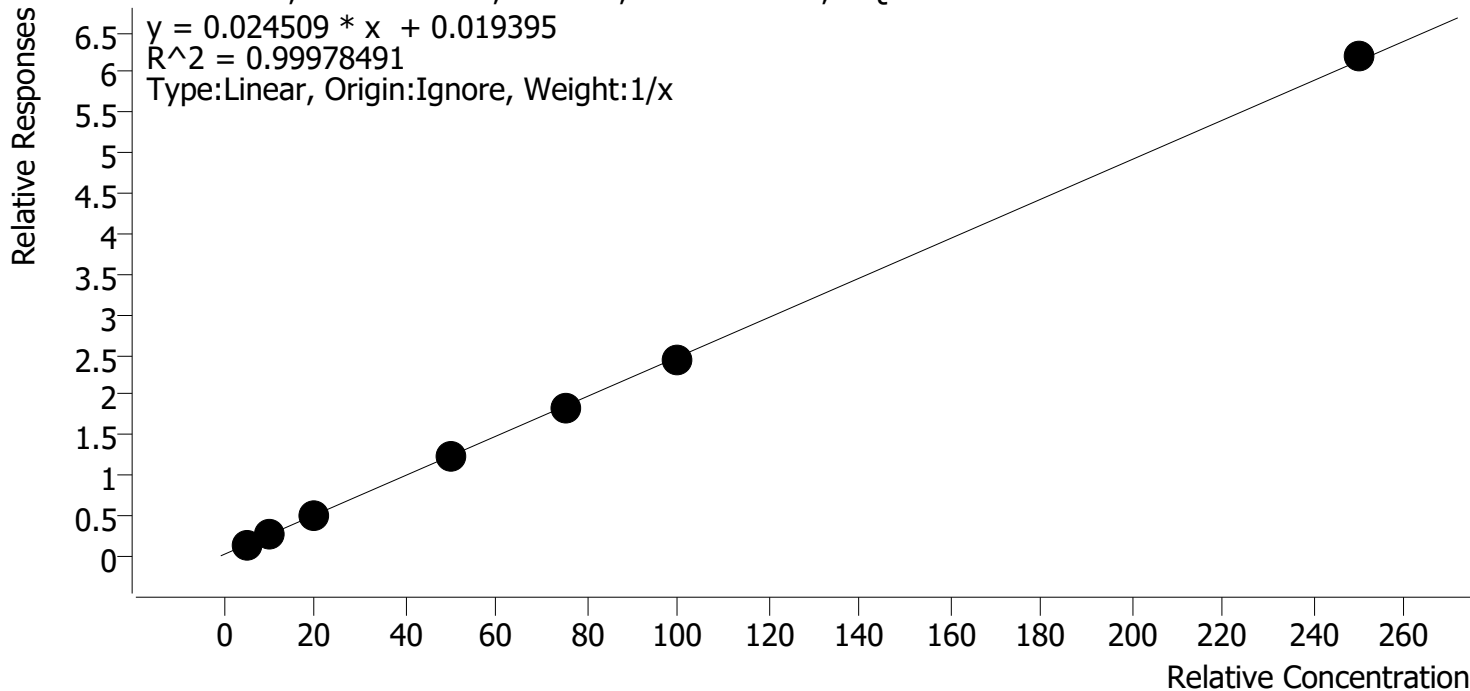
SC



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update 4/1/2021 10:21 AM
Analyst Name ISP\Datastor
Analyte THC-COOH **Internal Standard** THC-COOH-D9

THC-COOH - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJQ_Cal 1	1	✓	5.0	4.9	97.1
MJQ_Cal 2	2	✓	10.0	10.4	104.3
MJQ_Cal 3R	3	✓	20.0	20.3	101.6
MJQ_Cal 4	4	✓	50.0	49.3	98.6
MJQ_Cal 5	5	✓	75.0	73.7	98.2
MJQ_Cal 6	6	✓	100.0	99.3	99.3
MJQ_Cal 7	7	✓	250.0	252.1	100.8

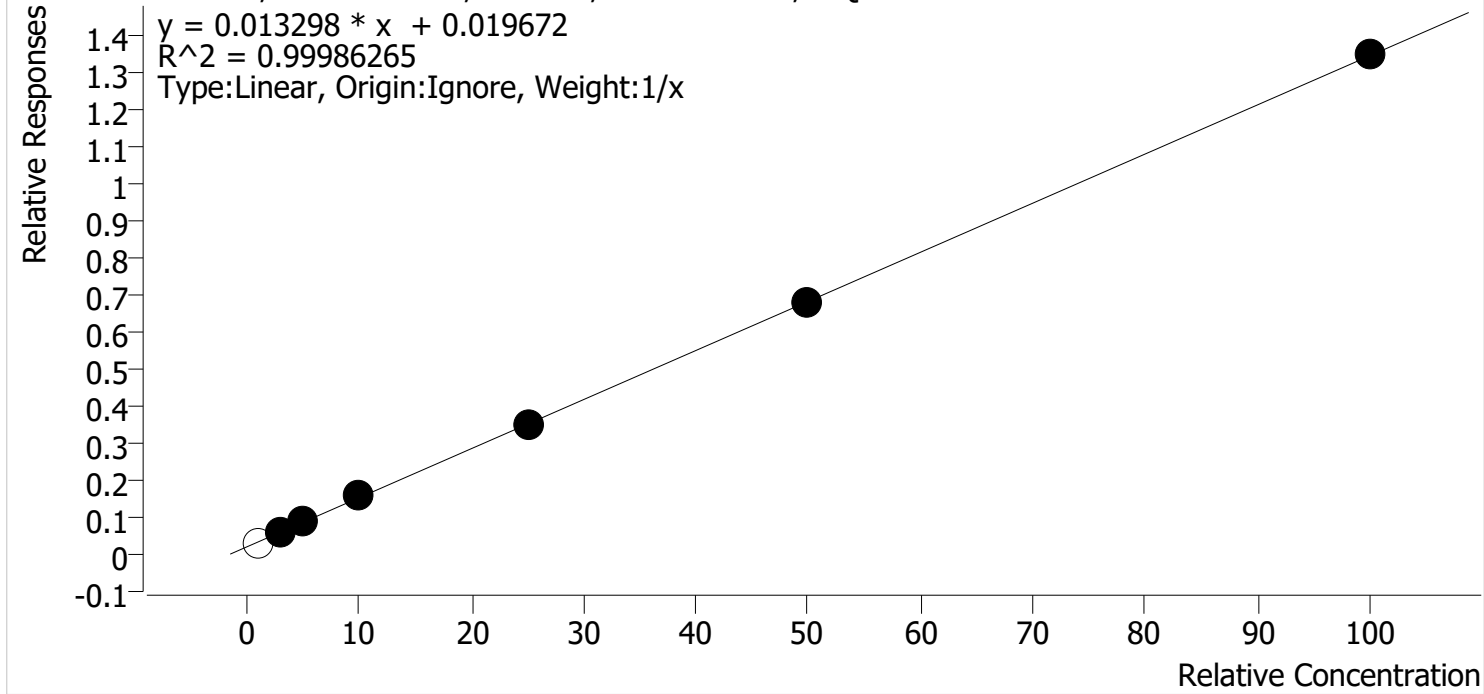
SC



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Last Cal. Update 4/1/2021 10:21 AM
Analyst Name ISP\Datastor
Analyte THC-OH **Internal Standard** THC-OH-D3

THC-OH - 7 Levels, 6 Levels Used, 7 Points, 6 Points Used, 0 QCs



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJQ_Cal 1	1	x	1.0	0.8	84.8
MJQ_Cal 2	2	✓	3.0	2.9	95.9
MJQ_Cal 3R	3	✓	5.0	5.2	103.4
MJQ_Cal 4	4	✓	10.0	10.2	102.0
MJQ_Cal 5	5	✓	25.0	24.7	98.8
MJQ_Cal 6	6	✓	50.0	49.9	99.7
MJQ_Cal 7	7	✓	100.0	100.2	100.2

Calibrator 1 dropped due to not meeting ratio requirement.

SC

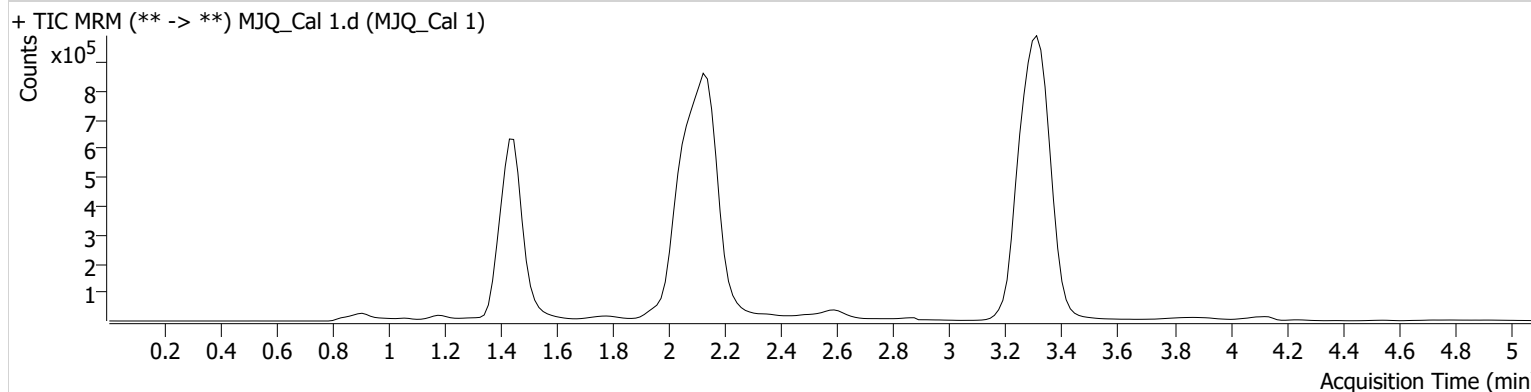


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 4/1/2021 10:21:04 AM

Instrument	Instrument 1	Data File	MJQ_Cal 1.d
Type	Cal	Sample	MJQ_Cal 1
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-A1	Comment	
Injection Volume	10		
Acq. Date-Time	3/31/2021 10:55:56 AM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.498 High	81449	∞	6.6 Low	49.23	2631450	0.8483 ng/ml Low
THC-COOH	1.474	92615	∞	48.6	∞	669177	4.8556 ng/ml Low
THC	3.330	66787	∞	59.6 High	54.87	7761574	1.2423 ng/ml

SC

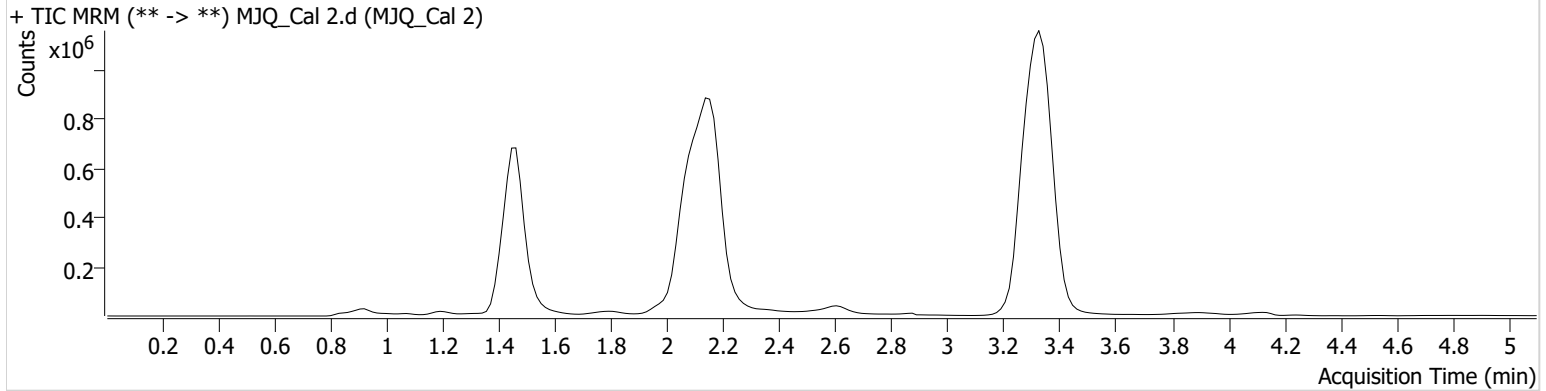


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 4/1/2021 10:21:04 AM

Instrument	Instrument 1	Data File	MJQ_Cal 2.d
Type	Cal	Sample	MJQ_Cal 2
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-B1	Comment	
Injection Volume	10		
Acq. Date-Time	3/31/2021 11:03:42 AM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.468	149419	∞	8.6	∞	2579024	2.8775 ng/ml Low
THC-COOH	1.489	191429	∞	48.7	∞	696043	10.4300 ng/ml
THC	3.345	205893	220.63	27.0	96.98	8362220	2.9715 ng/ml

SC

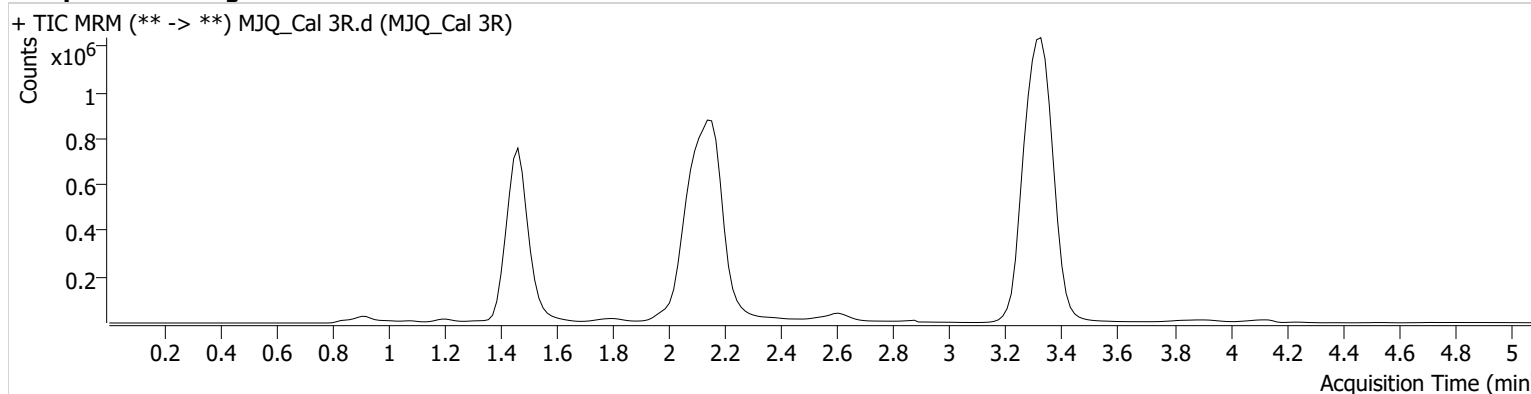


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 4/1/2021 10:21:04 AM

Instrument	Instrument 1	Data File	MJQ_Cal 3R.d
Type	Cal	Sample	MJQ_Cal 3R
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-C1	Comment	
Injection Volume	10		
Acq. Date-Time	3/31/2021 11:34:05 AM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.468	225504	∞	8.8	∞	2550741	5.1689 ng/ml
THC-COOH	1.489	357222	∞	52.5	∞	690476	20.3174 ng/ml
THC	3.330	383283	3654.20	30.9	644.27	8626068	5.1104 ng/ml

SC

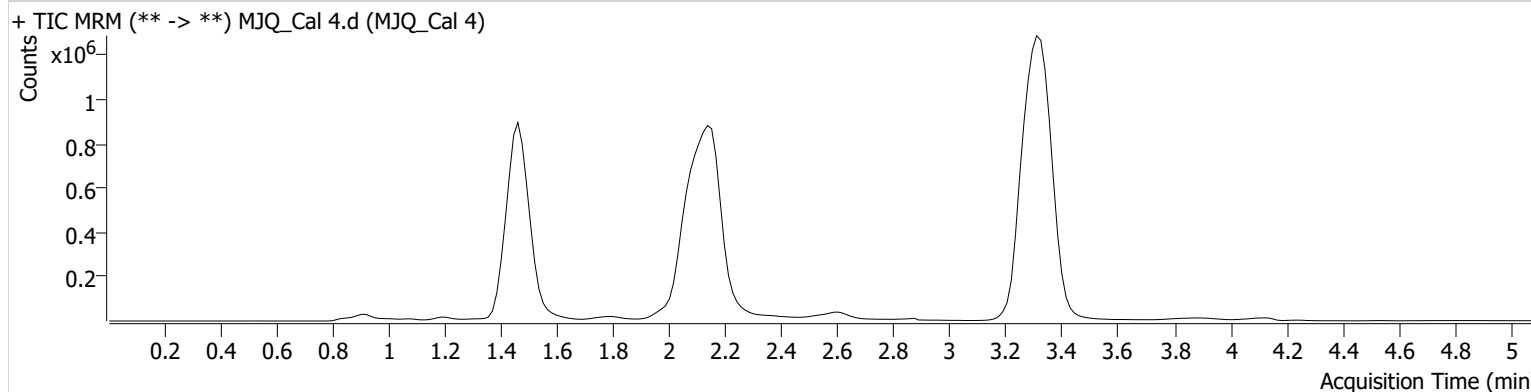


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 4/1/2021 10:21:04 AM

Instrument	Instrument 1	Data File	MJQ_Cal 4.d
Type	Cal	Sample	MJQ_Cal 4
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-D1	Comment	
Injection Volume	10		
Acq. Date-Time	3/31/2021 11:18:53 AM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.468	390122	∞	9.9	213.98	2511299	10.2027 ng/ml
THC-COOH	1.489	843598	∞	56.1	∞	687015	49.3091 ng/ml
THC	3.330	783151	3392.11	25.1	269.84	8638273	10.1012 ng/ml

SC

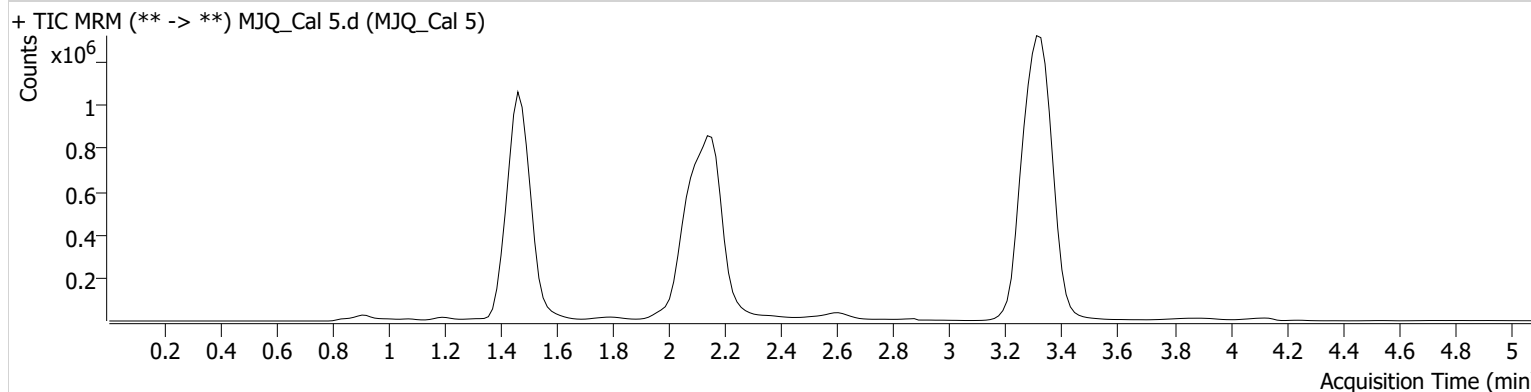


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 4/1/2021 10:21:04 AM

Instrument	Instrument 1	Data File	MJQ_Cal 5.d
Type	Cal	Sample	MJQ_Cal 5
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-E1	Comment	
Injection Volume	10		
Acq. Date-Time	3/31/2021 11:26:29 AM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.468	858351	∞	10.9	∞	2466377	24.6918 ng/ml
THC-COOH	1.489	1195087	∞	57.7	∞	654891	73.6652 ng/ml
THC	3.330	1752662	2063.31	24.7	301.88	7831774	24.4739 ng/ml

SC

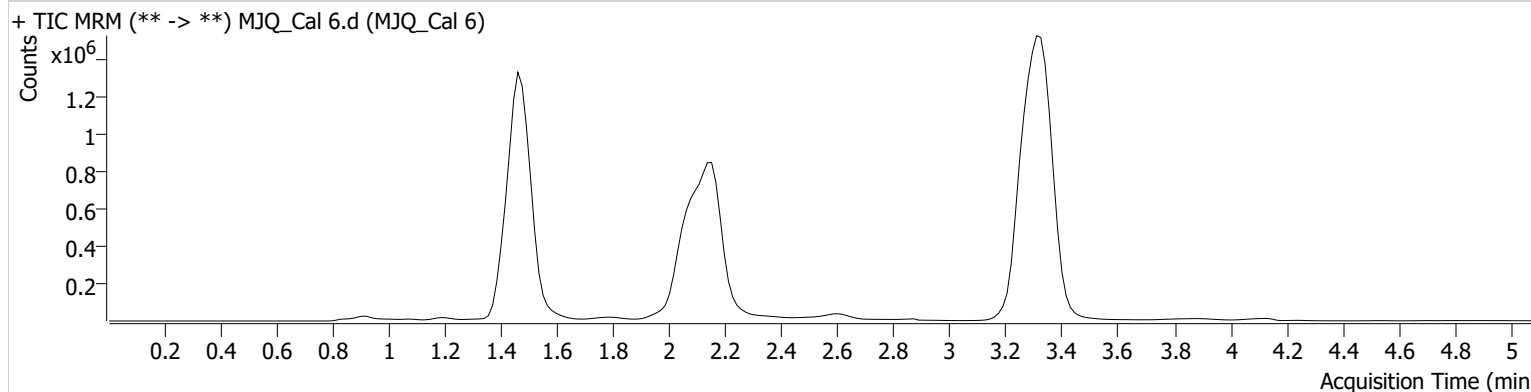


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 4/1/2021 10:21:04 AM

Instrument	Instrument 1	Data File	MJQ_Cal 6.d
Type	Cal	Sample	MJQ_Cal 6
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-F1	Comment	
Injection Volume	10		
Acq. Date-Time	3/31/2021 11:41:40 AM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.468	1705605	∞	11.4	3807.09	2498670	49.8525 ng/ml
THC-COOH	1.489	1588681	∞	58.1	∞	647452	99.3240 ng/ml
THC	3.330	3464311	4124.33	24.9	∞	7605178	49.4920 ng/ml

SC

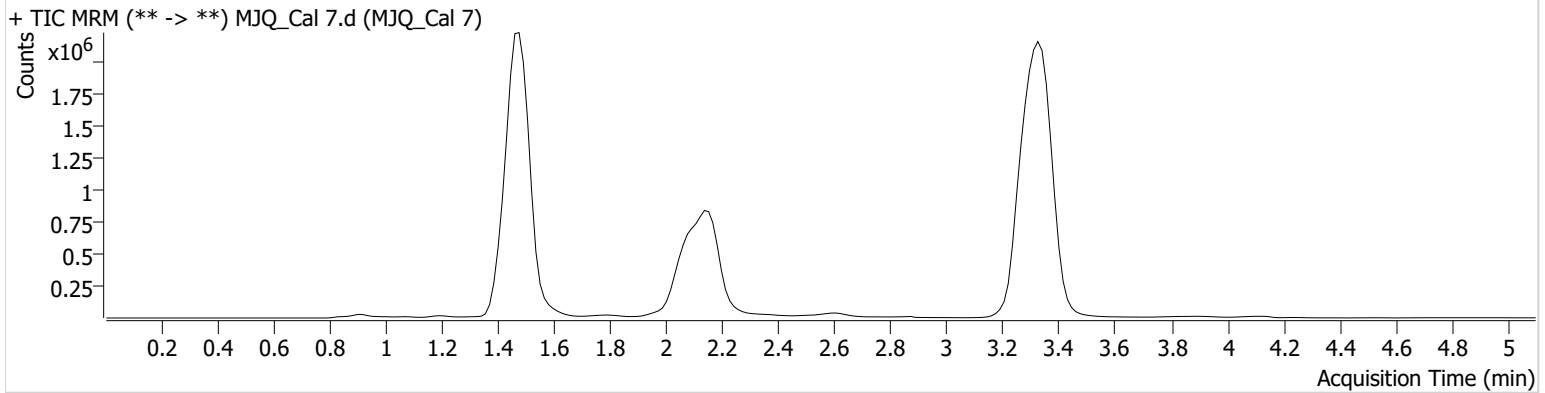


AM #27 Cannabinoid Quant. Results

Batch results D:\MassHunter\Data\2021\AM 27-28\033121 AM 27 28 SC\QuantResults\AM 27.batch.bin
Calibration Last Update 4/1/2021 10:21:04 AM

Instrument	Instrument 1	Data File	MJQ_Cal 7.d
Type	Cal	Sample	MJQ_Cal 7
Acq. Method	AM 27 THCQ.m	Operator	Sarah Collins
Sample Position	P1-G1	Comment	
Injection Volume	10		
Acq. Date-Time	3/31/2021 11:49:15 AM		

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-OH	1.468	3353946	∞	11.5	∞	2480351	100.2065 ng/ml
THC-COOH	1.489	3814878	∞	58.3	∞	615490	252.0988 ng/ml
THC	3.345	7262105	∞	25.2	3092.67	7798362	100.8510 ng/ml