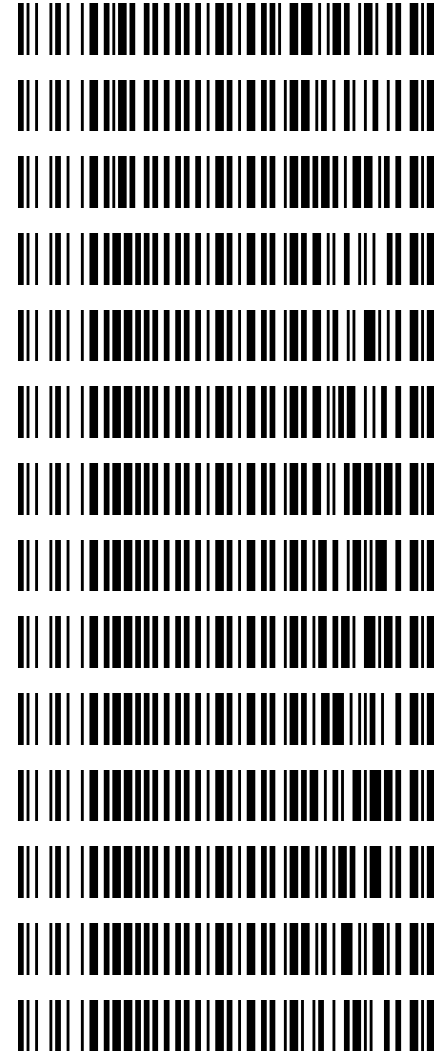


Worklist: 3729

<u>LAB_CASE</u>	<u>ITEM</u>	<u>TASK_ID</u>	<u>DESCRIPTION</u>
M2019-3972	2	165526	AM 27 Blood THC Quant by LC-QQQ
M2019-4002	2	165527	AM 27 Blood THC Quant by LC-QQQ
M2019-4097	2	165528	AM 27 Blood THC Quant by LC-QQQ
P2019-2798	1	165529	AM 27 Blood THC Quant by LC-QQQ
P2019-2799	1	165530	AM 27 Blood THC Quant by LC-QQQ
P2019-2800	1	165531	AM 27 Blood THC Quant by LC-QQQ
P2019-2801	1	165532	AM 27 Blood THC Quant by LC-QQQ
P2019-2808	1	165533	AM 27 Blood THC Quant by LC-QQQ
P2019-2809	1	165534	AM 27 Blood THC Quant by LC-QQQ
P2019-2814	1	165535	AM 27 Blood THC Quant by LC-QQQ
P2019-2815	1	165536	AM 27 Blood THC Quant by LC-QQQ
P2019-2861	1	165537	AM 27 Blood THC Quant by LC-QQQ
P2019-2872	1	165538	AM 27 Blood THC Quant by LC-QQQ
P2019-2886	1	165539	AM 27 Blood THC Quant by LC-QQQ



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

Extraction Date: 10/01/19

Analyst: Sarah Pickle

Plate lot#: Lot # 190716 Item # IDP-108

Plate Expiration: 1/16/20

Mobile phase A: 0.1% Formic Acid in LCMS Water
MTBE

Mobile phase B: 0.1% Formic acid in Acetonitrile
Hexane

Blank Blood Lot: 445283-3

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: #3**
- 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes. *Shaker ID: 067105*
- 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) Manifold ID: 067104
- 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.
SPE Dry ID: 067103
- 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.
Worklist path: D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP Batch Name: THCQ SP
- 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: 10ng/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 Ranges: THC: 3-100, THC-COOH 10-250, THC-OH 3-100*

Samples were extracted on 10/01/19. Due to the pressure maxing out and stopping the run, the samples were not ran until 10/02/19.

AM #27 Cannabinoids Quant. Results

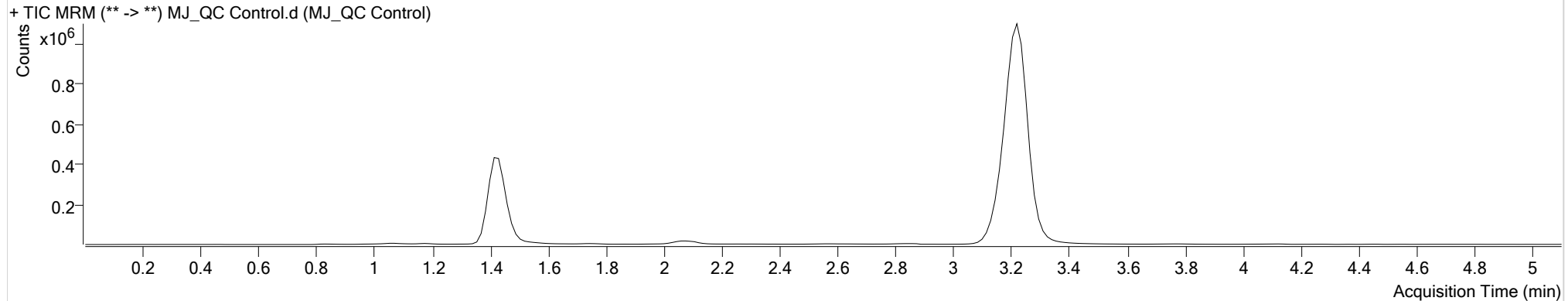


S

Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Calibration Last Update 10/4/2019 8:35:27 AM

Instrument	Falco	Data File	MJ_QC Control.d
Type	Sample	Sample	MJ_QC Control
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-A6		
Injection Volume	10		
Acq. Date-Time	10/2/2019 3:02:08 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.224	203926	1257.90	27.7	124.47	6140226	4.4410 ng/ml
THC-COOH	1.459	121093	1596.49	57.8	1209.02	380758	13.0215 ng/ml
THC-OH	1.423	81518	∞	12.3	109.79	1264193	4.2396 ng/ml

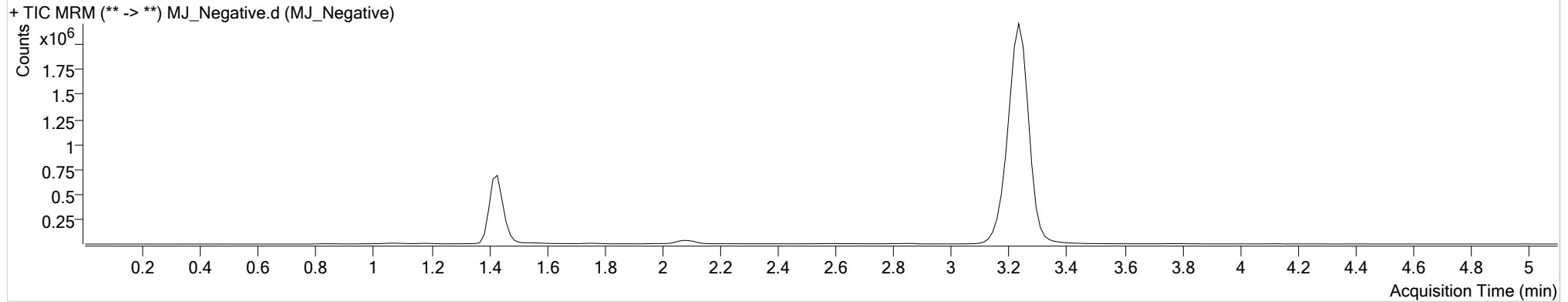
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Calibration Last Update 10/4/2019 8:35:27 AM

Instrument	Falco	Data File	MJ_Negative.d
Type	Sample	Sample	MJ_Negative
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-H5		
Injection Volume	10		
Acq. Date-Time	10/2/2019 3:17:21 PM		
Sample Info.			

Sample Chromatogram



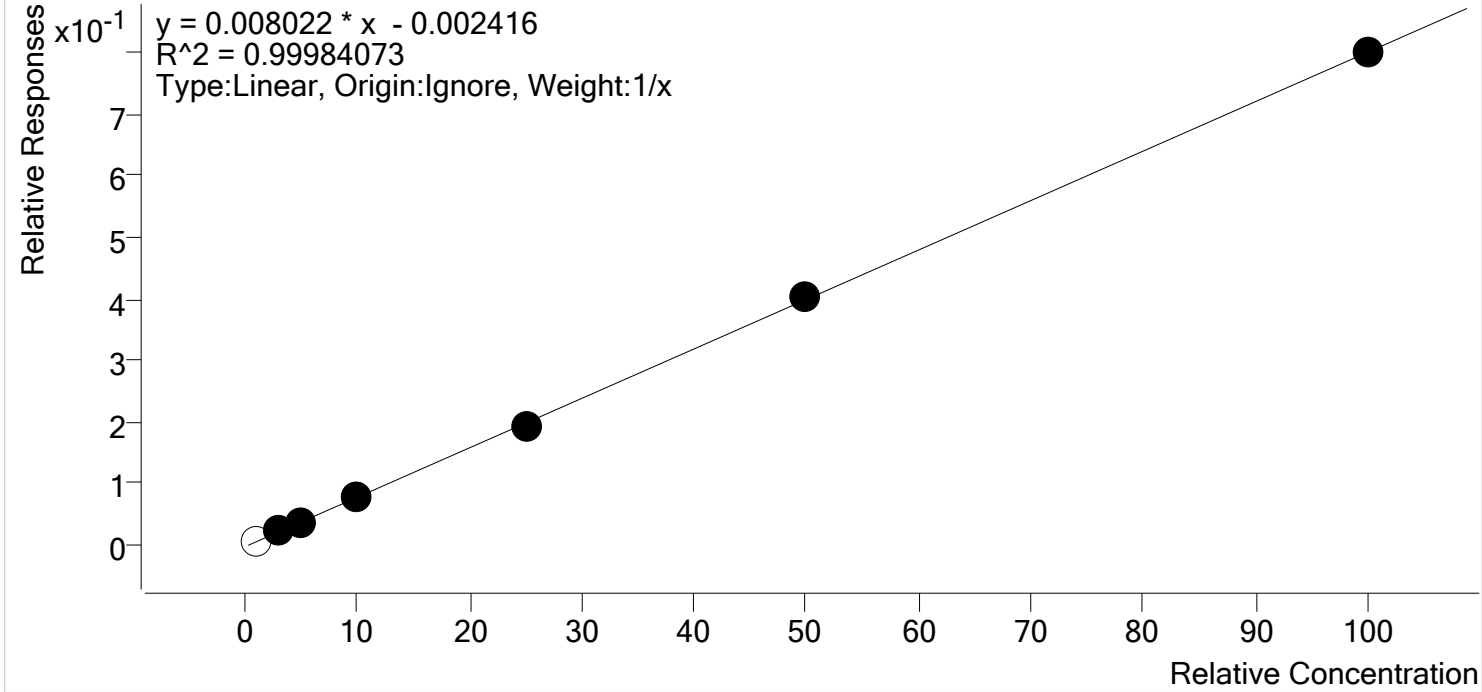
2



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Last Cal. Update 10/4/2019 8:35 AM
Analyst Name ISP\Datastor
Analyte THC **Internal Standard** THC-D3

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

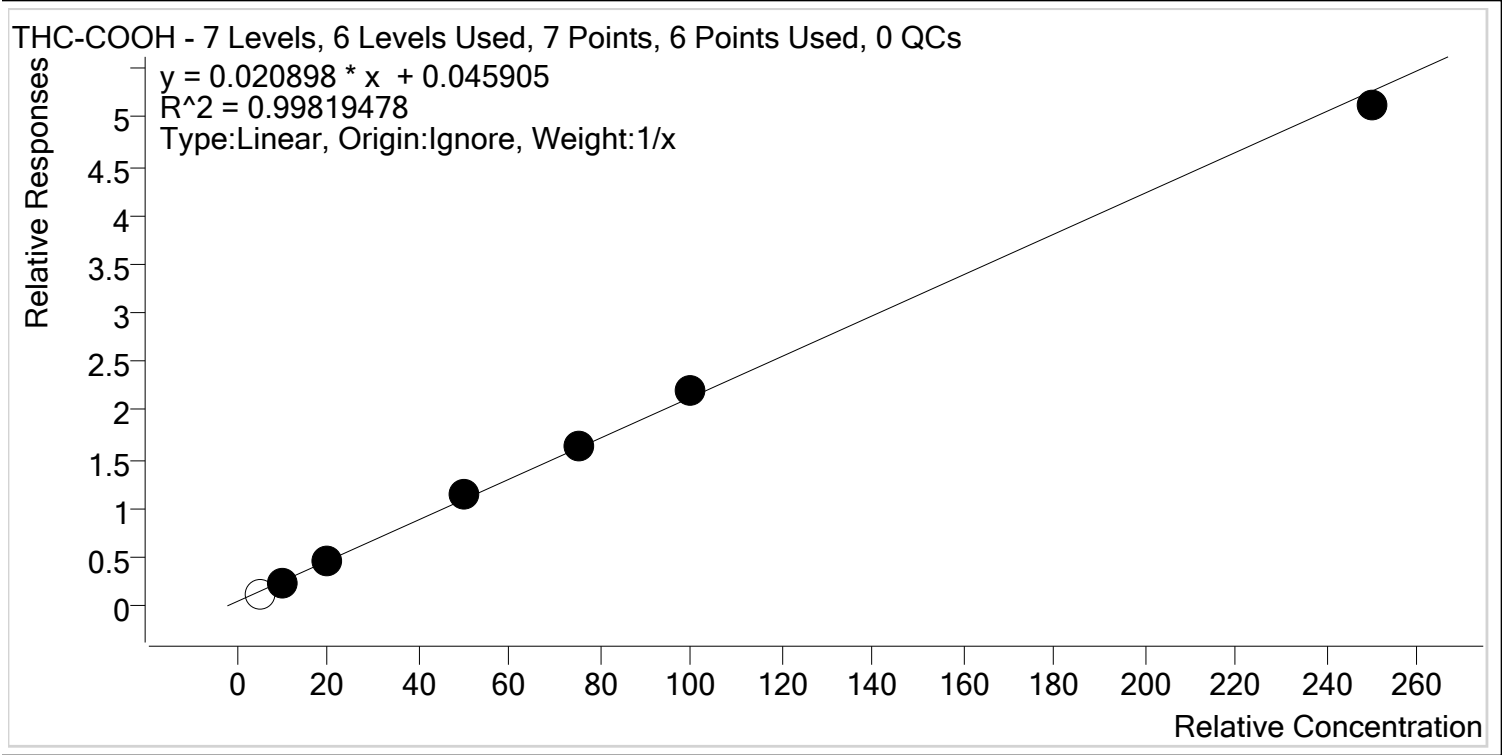


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	x	1.0	1.2	123.7
MJ_Cal 2	2	✓	3.0	3.1	102.2
MJ_Cal 3	3	✓	5.0	4.9	97.0
MJ_Cal 4	4	✓	10.0	10.2	101.7
MJ_Cal 5	5	✓	25.0	24.5	98.1
MJ_Cal 6	6	✓	50.0	50.5	101.0
MJ_Cal 7	7	✓	100.0	99.9	99.9



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Last Cal. Update 10/4/2019 8:35 AM
Analyst Name ISP\Datastor
Analyte THC-COOH **Internal Standard** THC-COOH-D9

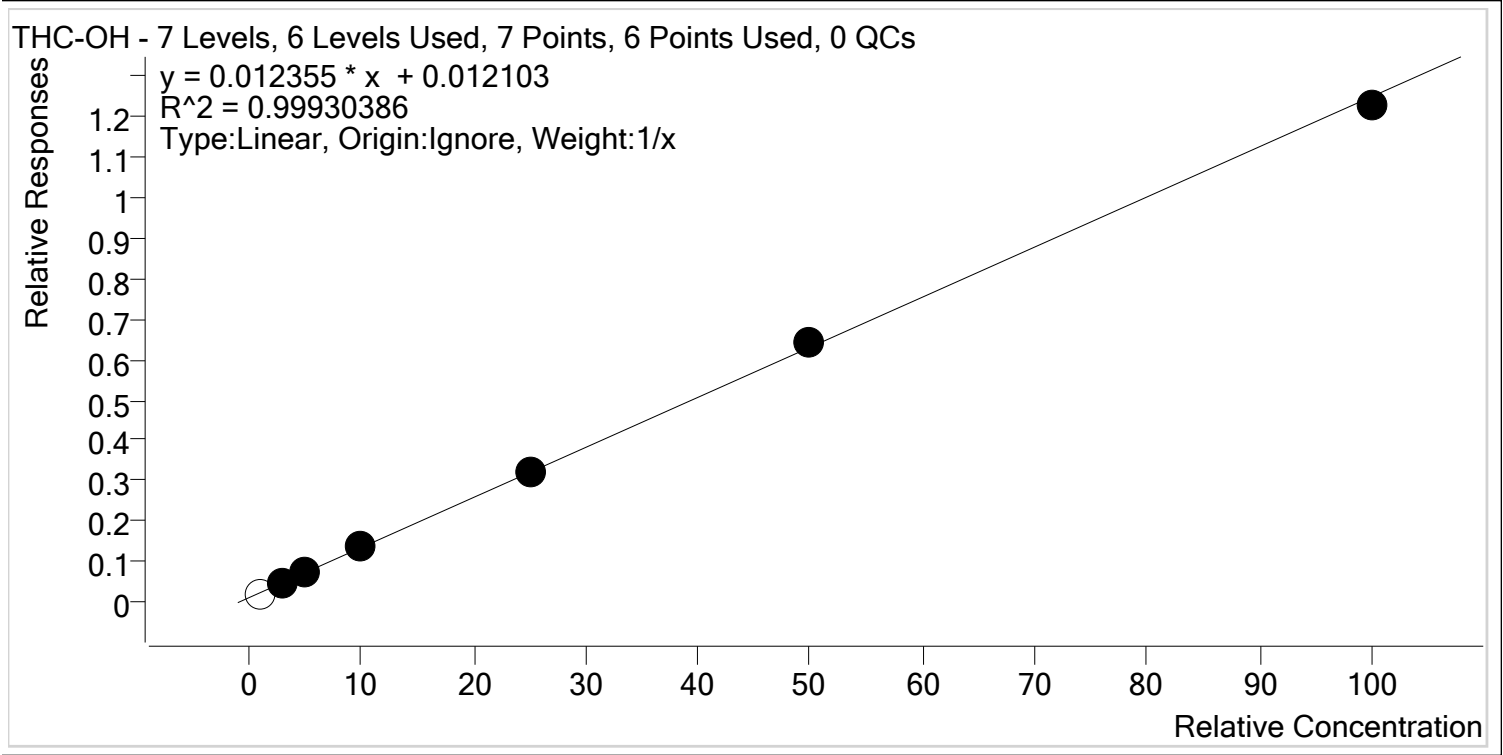


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	x	5.0	3.8	75.6
MJ_Cal 2	2	✓	10.0	9.4	93.8
MJ_Cal 3	3	✓	20.0	19.5	97.6
MJ_Cal 4	4	✓	50.0	52.6	105.2
MJ_Cal 5	5	✓	75.0	76.7	102.3
MJ_Cal 6	6	✓	100.0	103.9	103.9
MJ_Cal 7	7	✓	250.0	242.8	97.1



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Last Cal. Update 10/4/2019 8:35 AM
Analyst Name ISP\Datastor
Analyte THC-OH **Internal Standard** THC-OH-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
MJ_Cal 1	1	x	1.0	0.8	81.0
MJ_Cal 2	2	✓	3.0	2.9	97.0
MJ_Cal 3	3	✓	5.0	4.8	96.4
MJ_Cal 4	4	✓	10.0	10.4	104.3
MJ_Cal 5	5	✓	25.0	25.4	101.8
MJ_Cal 6	6	✓	50.0	51.2	102.5
MJ_Cal 7	7	✓	100.0	98.2	98.2

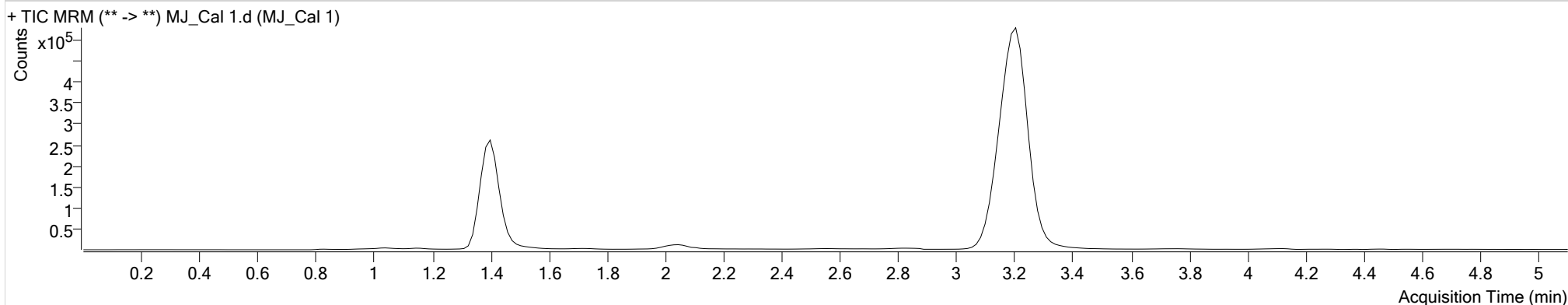
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Calibration Last Update 10/4/2019 8:35:27 AM

Instrument Falco **Data File** MJ_Cal 1.d
Type Cal **Sample** MJ_Cal 1
Acq. Method AM 27 THC quant.m
Sample Position P3-B6 **Comment**
Injection Volume 10
Acq. Date-Time 10/2/2019 2:08:59 PM
Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.209	27029	149.59	31.9	16.84	3602023	1.2365 ng/ml
THC-COOH	1.429	35224	113.29	51.0	189.76	282088	3.7785 ng/ml
THC-OH	1.423	19363	∞	8.5 Low	15.21	875622	0.8103 ng/ml

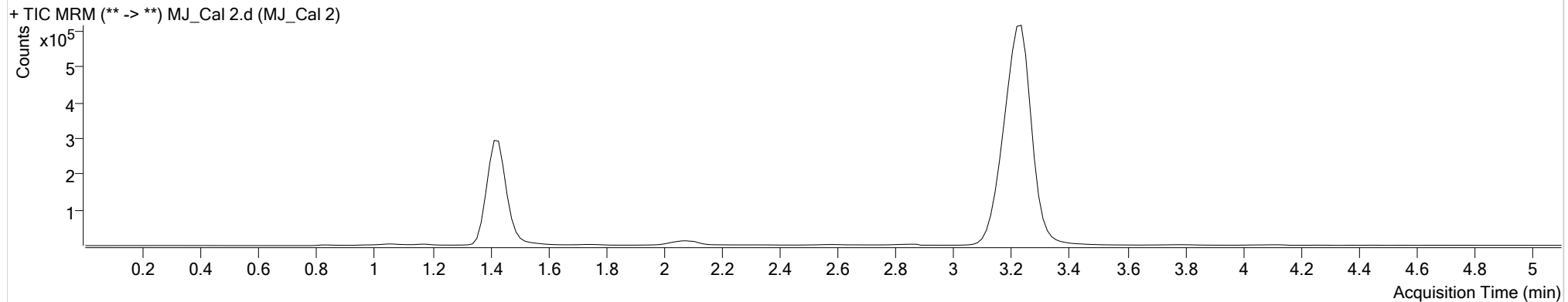
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Calibration Last Update 10/4/2019 8:35:27 AM

Instrument Falco **Data File** MJ_Cal 2.d
Type Cal **Sample** MJ_Cal 2
Acq. Method AM 27 THC quant.m
Sample Position P3-C6 **Comment**
Injection Volume 10
Acq. Date-Time 10/2/2019 2:16:43 PM
Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.239	88985	352.74	29.7	85.58	4009785	3.0674 ng/ml
THC-COOH	1.459	72207	∞	53.2	244.83	298386	9.3830 ng/ml
THC-OH	1.438	45443	∞	11.1	279.01	945737	2.9096 ng/ml

AM #27 Cannabinoids Quant. Results

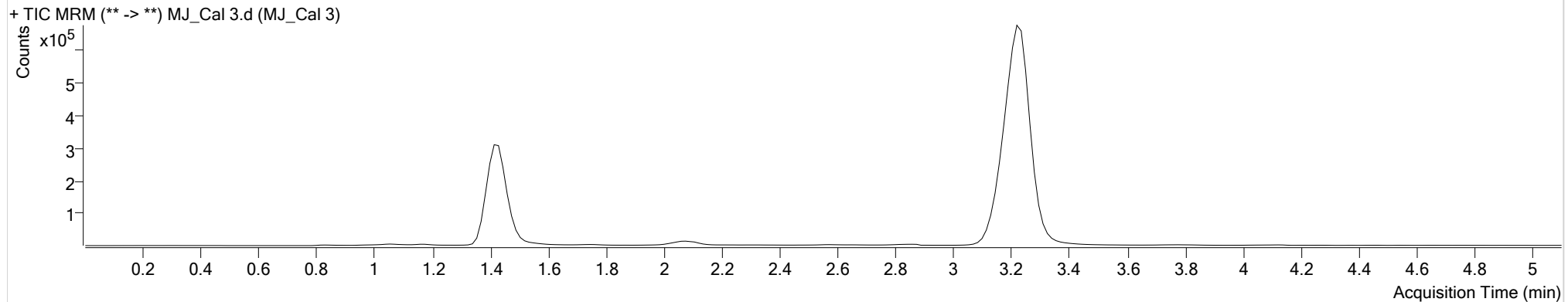


5

Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Calibration Last Update 10/4/2019 8:35:27 AM

Instrument Falco **Data File** MJ_Cal 3.d
Type Cal **Sample** MJ_Cal 3
Acq. Method AM 27 THC quant.m
Sample Position P3-D6 **Comment**
Injection Volume 10
Acq. Date-Time 10/2/2019 2:24:17 PM
Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.239	151838	372.21	28.4	132.00	4159419	4.8515 ng/ml
THC-COOH	1.459	134998	∞	56.9	954.51	297431	19.5221 ng/ml
THC-OH	1.423	67657	∞	11.7	131.60	944535	4.8181 ng/ml

AM #27 Cannabinoids Quant. Results

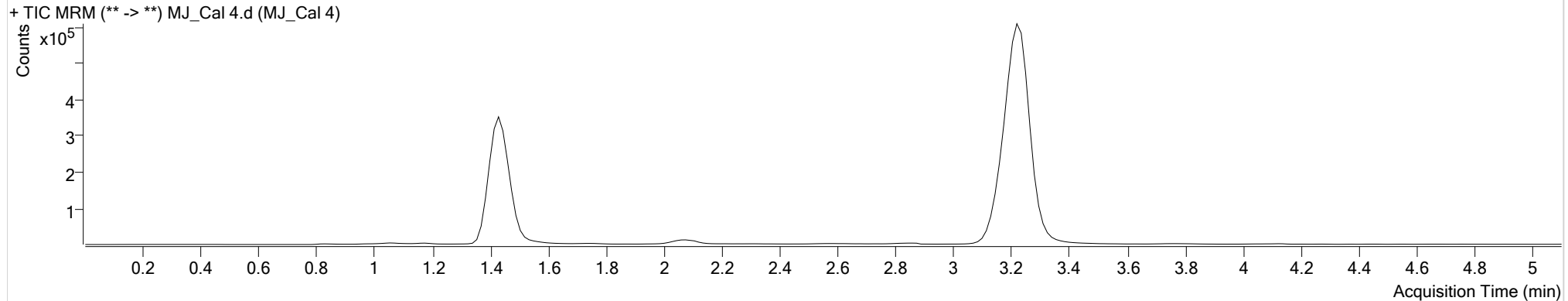


Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Calibration Last Update 10/4/2019 8:35:27 AM

Instrument Falco
Type Cal
Acq. Method AM 27 THC quant.m
Sample Position P3-E6
Injection Volume 10
Acq. Date-Time 10/2/2019 2:31:52 PM
Sample Info.

Data File MJ_Cal 4.d
Sample MJ_Cal 4
Comment

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.239	277338	839.30	27.1	247.40	3502786	10.1705 ng/ml
THC-COOH	1.459	295929	∞	59.3	1534.00	258430	52.5977 ng/ml
THC-OH	1.423	117258	∞	12.2	221.41	832167	10.4254 ng/ml

AM #27 Cannabinoids Quant. Results

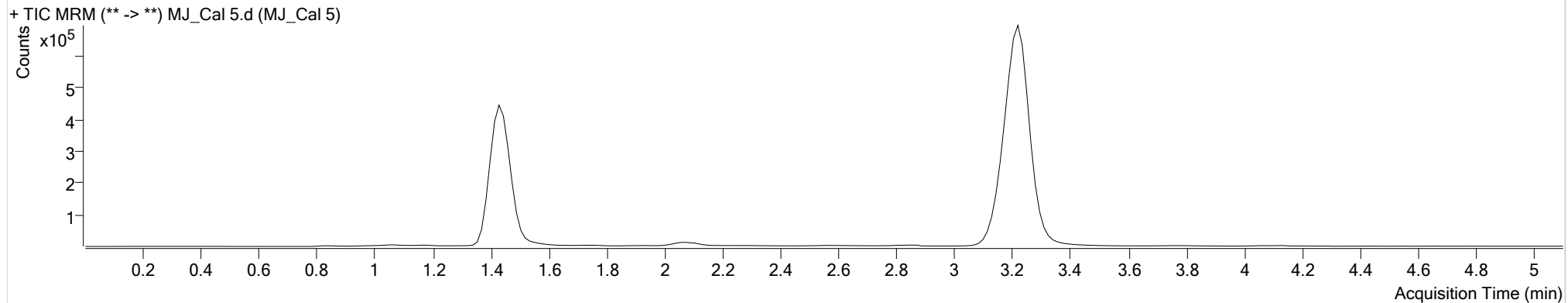


S

Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Calibration Last Update 10/4/2019 8:35:27 AM

Instrument	Falco	Data File	MJ_Cal 5.d
Type	Cal	Sample	MJ_Cal 5
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-F6		
Injection Volume	10		
Acq. Date-Time	10/2/2019 2:39:26 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.239	673641	1807.52	27.5	494.87	3464834	24.5360 ng/ml
THC-COOH	1.459	429128	1589.23	59.7	5515.36	260202	76.7199 ng/ml
THC-OH	1.423	279484	∞	13.5	∞	856241	25.4398 ng/ml

AM #27 Cannabinoids Quant. Results

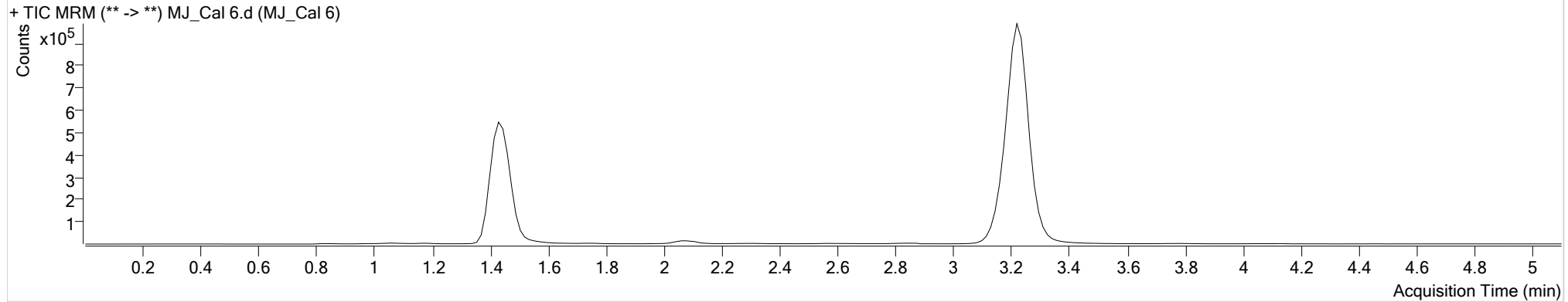


Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Calibration Last Update 10/4/2019 8:35:27 AM

Instrument Falco
Type Cal
Acq. Method AM 27 THC quant.m
Sample Position P3-G6
Injection Volume 10
Acq. Date-Time 10/2/2019 2:47:00 PM
Sample Info.

Data File MJ_Cal 6.d
Sample MJ_Cal 6
Comment

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.239	1484786	3921.03	27.0	893.31	3686964	50.4994 ng/ml
THC-COOH	1.459	544686	1684.60	59.0	4932.75	245585	103.9328 ng/ml
THC-OH	1.423	527390	832.07	14.1	1976.62	817583	51.2315 ng/ml

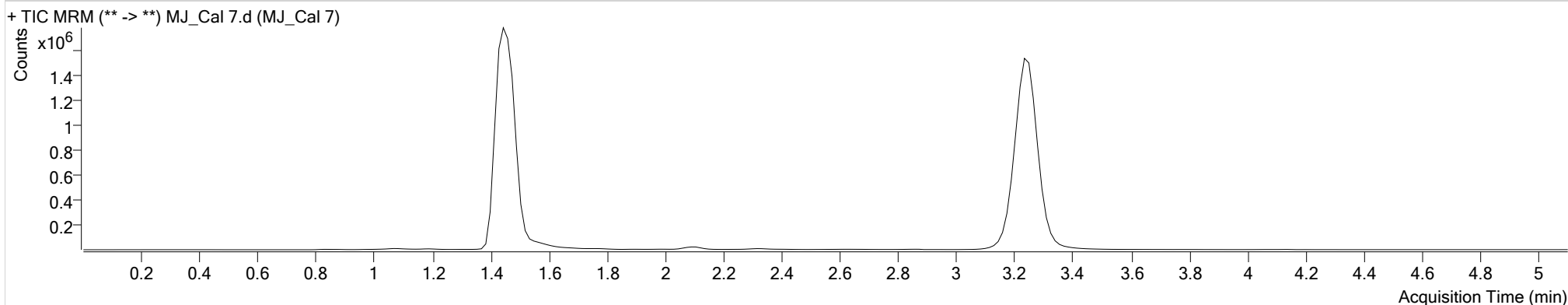
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 27\100119 MDS CS THCQ SP\QuantResults\THCQ SP.batch.bin
Calibration Last Update 10/4/2019 8:35:27 AM

Instrument Falco **Data File** MJ_Cal 7.d
Type Cal **Sample** MJ_Cal 7
Acq. Method AM 27 THC quant.m
Sample Position P3-H6 **Comment**
Injection Volume 10
Acq. Date-Time 10/2/2019 2:54:34 PM
Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.254	3387258	21390.38	26.9	1356.31	4240288	99.8752 ng/ml
THC-COOH	1.474	2007531	∞	60.2	8561.01	392025	242.8445 ng/ml
THC-OH	1.438	2209076	∞	14.3	∞	1803260	98.1756 ng/ml