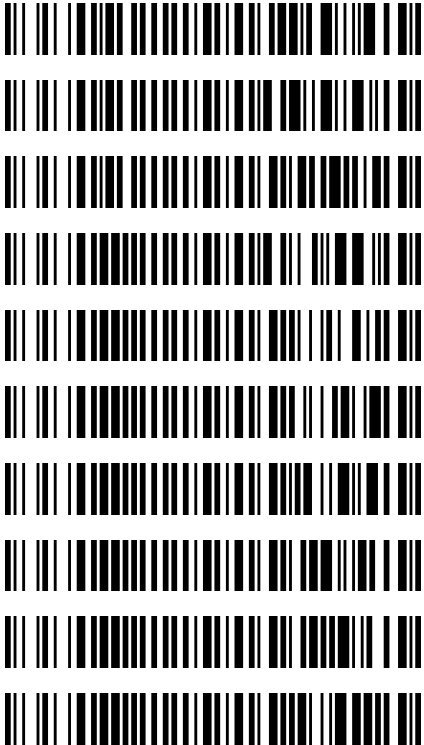


Worklist: 3386

<u>LAB_CASE</u>	<u>ITEM</u>	<u>TASK_ID</u>	<u>DESCRIPTION</u>
M2018-6230	1	151552	AM 27 Blood THC Quant by LC-QQQ
M2019-1530	1	151553	AM 27 Blood THC Quant by LC-QQQ
M2019-1579	1	151554	AM 27 Blood THC Quant by LC-QQQ
P2019-0890	1	151555	AM 27 Blood THC Quant by LC-QQQ
P2019-1179	1	151556	AM 27 Blood THC Quant by LC-QQQ
P2019-1185	1	151557	AM 27 Blood THC Quant by LC-QQQ
P2019-1200	1	151558	AM 27 Blood THC Quant by LC-QQQ
P2019-1215	1	151559	AM 27 Blood THC Quant by LC-QQQ
P2019-1216	1	151560	AM 27 Blood THC Quant by LC-QQQ
P2019-1266	1	151561	AM 27 Blood THC Quant by LC-QQQ



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

Extraction Date: 5/13/19
Plate lot#: 0539904

Analyst: Sarah Pickle
Plate Expiration: 9/10/19

Mobile phase A: 0.1% Formic Acid in LCMS Water
MTBE
Mobile phase B: 0.1% Formic acid in Acetonitrile
LCMS Methanol Hexane
Blank Blood Lot: 445283-1
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\051319 THCQ SP Batch Name: THCQ
- 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 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 Range limited: THC-COOH 5-100*



Idaho State Police Forensic Services

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

Methanol External Control Solution (Lot: WS041619)

10 ul of 1mg/mL THC, 100 ul of 100 ug/mL THC-OH, C-THC in 9790 ul MeOH
Approximate concentration 1ug/mL.

Component	Source	Source Lot Number	Expiration Date
Methanol (LCMS)	Fisher	184782	
THC	Cerilliant	FE09101501	11/30/2020
C-THC	Cerilliant	FE07171501	09/30/2019
THC-OH	Cerilliant	FE01121503	01/31/2020
Prepared:	04/16/2019		
Prepared By:	Tamara Salazar		
Expires:	01/31/2020		

Blood External Control Solution (Lot: 041619)

100 ul of methanol external control solution was added to 9900 ul of blood.
Approximately 10ng/mL of each compound.

Component	Source	Source Lot Number
Negative Blood	Hemostat	445283-1
Methanol External Control Solution	-	WS041619
Prepared:	04/16/19	
Prepared by:	Tamara Salazar	
Expires:	01/31/2020	

AM #27 Cannabinoids Quant. Results

**Batch results**

D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin

Calibration Last Update

5/13/2019 3:35:29 PM

Instrument

FALCO-LCMS (Property ID 069901)

Type

Sample

Data File

Negative.d

Acq. Method

AM 27 THC quant.m

Sample

Negative

Sample Position

P3-A2

Comment**Injection Volume**

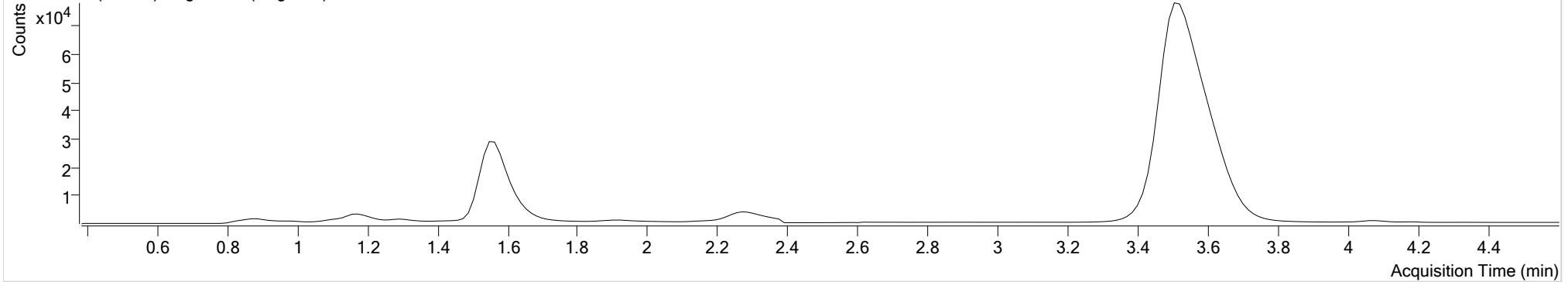
10

Acq. Date-Time

5/13/2019 12:39:48 PM

Sample Info.**Sample Chromatogram**

+ TIC MRM (** -> **) Negative.d (Negative)



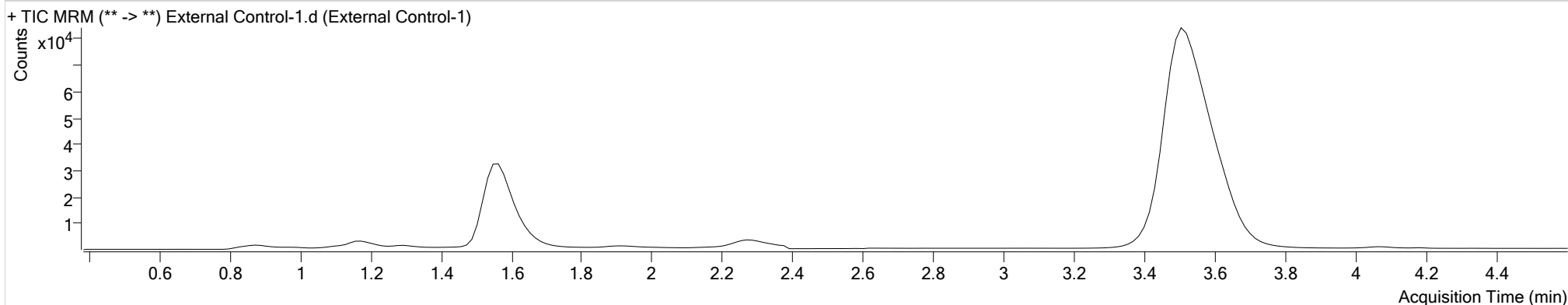
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin
Calibration Last Update 5/13/2019 3:35:29 PM

Instrument Type	FALCO-LCMS (Property ID 069901)	Data File	External Control-1.d
Acq. Method	AM 27 THC quant.m	Sample	External Control-1
Sample Position	P3-B2	Comment	
Injection Volume	10		
Acq. Date-Time	5/13/2019 12:54:59 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.521	51085	∞	28.2	∞	736596	7.9452 ng/ml
THC-COOH	1.595	10383	21.57	46.6	93.58	43657	7.9596 ng/ml
THC-OH	1.558	17038	∞	12.5	64.79	118128	8.0749 ng/ml

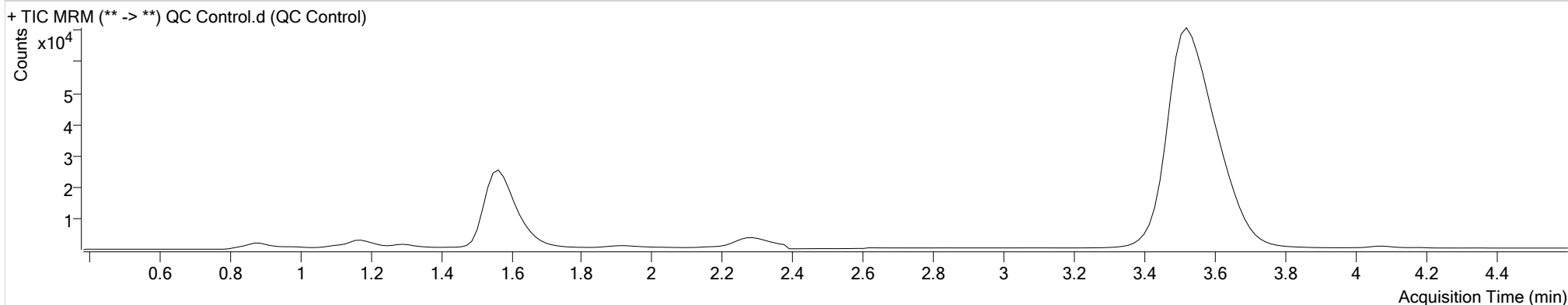
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin
Calibration Last Update 5/13/2019 3:35:29 PM

Instrument FALCO-LCMS (Property ID 069901) **Data File** QC Control.d
Type QC **Sample** QC Control
Acq. Method AM 27 THC quant.m
Sample Position P3-H1 **Comment**
Injection Volume 10
Acq. Date-Time 5/13/2019 12:24:37 PM
Sample Info.

Sample Chromatogram



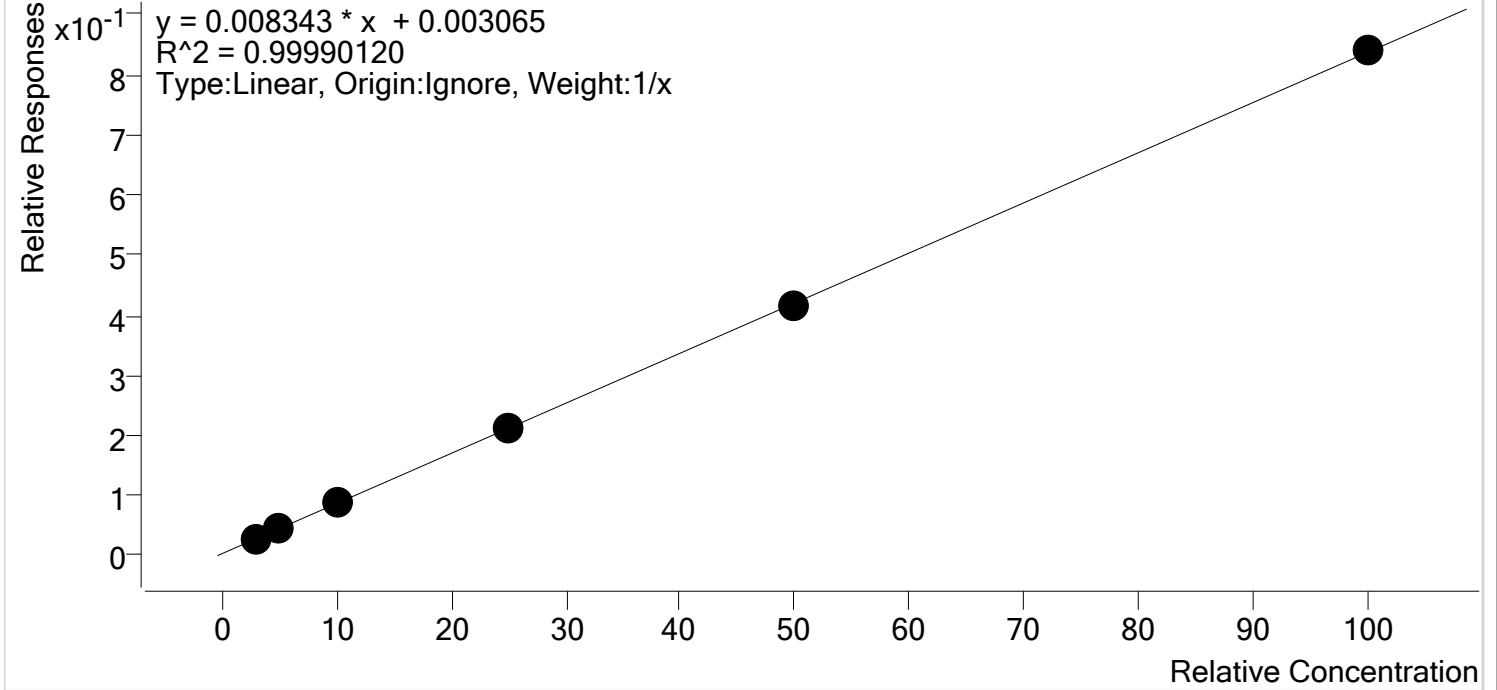
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.536	28209	256.47	28.2	75.05	638860	4.9251 ng/ml
THC-COOH	1.595	10143	21.68	44.3	71.42	35583	10.2703 ng/ml
THC-OH	1.573	9974	∞	10.8	17.90	94344	5.5243 ng/ml



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin
Last Cal. Update 5/13/2019 3:35 PM
Analyst Name ISP\Datastor
Analyte THC **Internal Standard** THC-D3

THC - 6 Levels, 6 Levels Used, 6 Points, 6 Points Used, 1 QCs

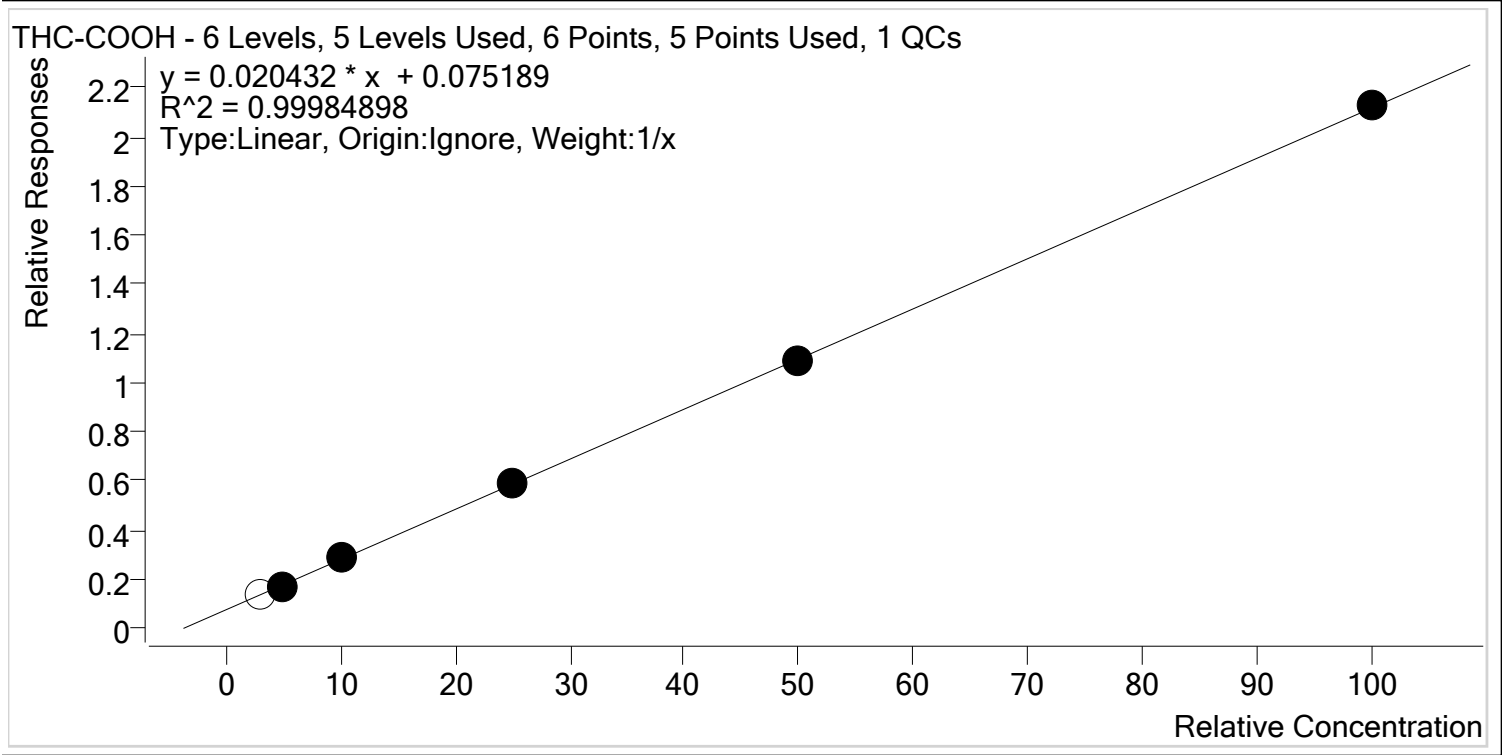


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1-3ng	1	✓	3.0	3.0	100.5
Cal 2- 5ng	2	✓	5.0	5.0	99.4
Cal 3 -10ng	3	✓	10.0	10.0	99.7
Cal 4-25ng	4	✓	25.0	25.4	101.4
Cal 5-50ng	5	✓	50.0	49.4	98.8
Cal 6-100ng	6	✓	100.0	100.3	100.3



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin
Last Cal. Update 5/13/2019 3:35 PM
Analyst Name ISP\Datastor
Analyte THC-COOH **Internal Standard** THC-COOH-D9

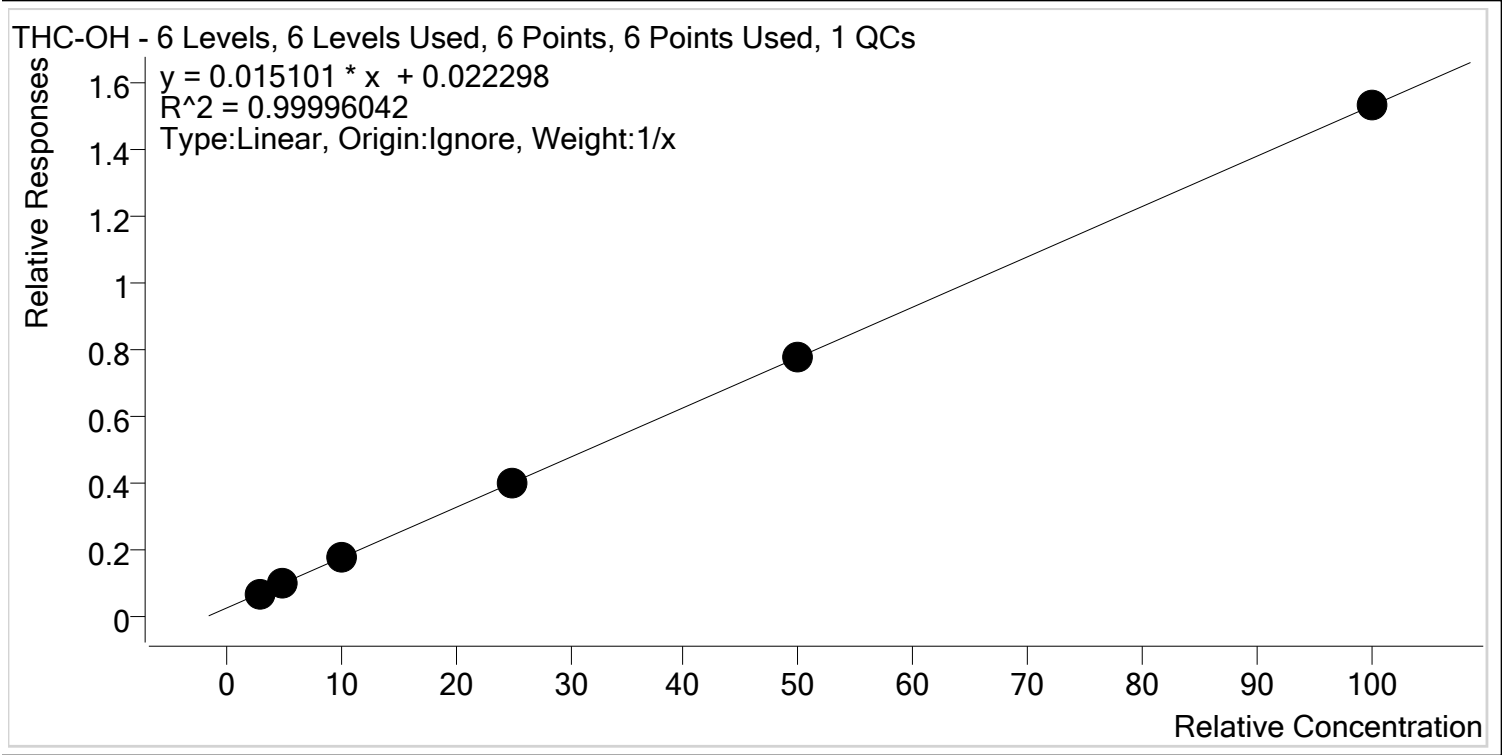


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1-3ng	1	x	3.0	3.2	107.9
Cal 2- 5ng	2	✓	5.0	4.9	98.1
Cal 3 -10ng	3	✓	10.0	10.3	102.6
Cal 4-25ng	4	✓	25.0	25.0	100.0
Cal 5-50ng	5	✓	50.0	49.4	98.8
Cal 6-100ng	6	✓	100.0	100.4	100.4



AM #27 Cannabinoids Quant. Calibration Curve Report

Batch results D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin
Last Cal. Update 5/13/2019 3:35 PM
Analyst Name ISP\Datastor
Analyte THC-OH **Internal Standard** THC-OH-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1-3ng	1	✓	3.0	2.9	97.7
Cal 2- 5ng	2	✓	5.0	5.0	101.0
Cal 3 -10ng	3	✓	10.0	10.2	101.8
Cal 4-25ng	4	✓	25.0	24.9	99.8
Cal 5-50ng	5	✓	50.0	49.9	99.8
Cal 6-100ng	6	✓	100.0	100.0	100.0

AM #27 Cannabinoids Quant. Results

**Batch results**

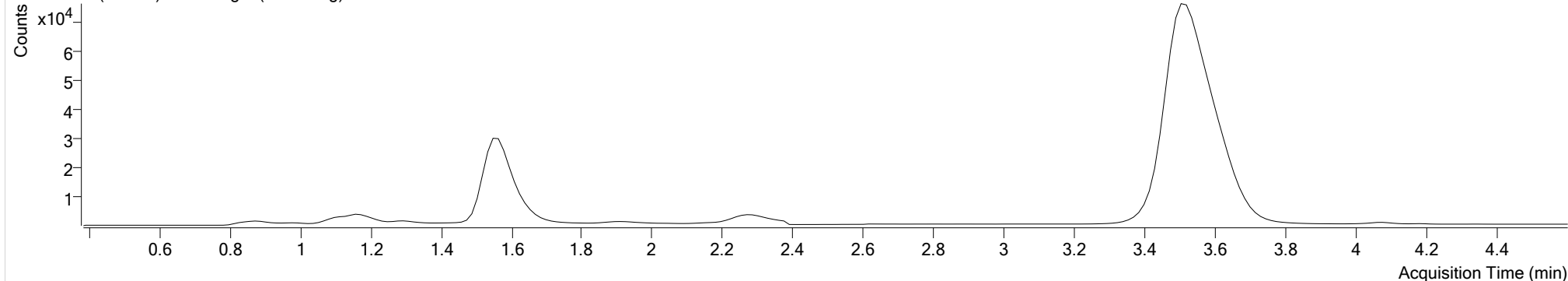
D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin

Calibration Last Update

5/13/2019 3:35:29 PM

Instrument FALCO-LCMS (Property ID 069901)**Type** Cal**Acq. Method** AM 27 THC quant.m**Sample Position** P3-B1**Injection Volume** 10**Acq. Date-Time** 5/13/2019 11:39:10 AM**Sample Info.****Data File** Cal 1-3ng.d**Sample** Cal 1-3ng**Comment****Sample Chromatogram**

+ TIC MRM (** -> **) Cal 1-3ng.d (Cal 1-3ng)



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.521	20175	∞	28.5	66.61	715175	3.0138 ng/ml
THC-COOH	1.595	6103	13.56	29.4 Low	28.85	43192	3.2360 ng/ml
THC-OH	1.573	7933	43.87	10.3	16.20	119178	2.9311 ng/ml

AM #27 Cannabinoids Quant. Results

**Batch results**

D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin

Calibration Last Update

5/13/2019 3:35:29 PM

Instrument

FALCO-LCMS (Property ID 069901)

Type

Cal

Acq. Method

AM 27 THC quant.m

Sample Position

P3-C1

Injection Volume

10

Acq. Date-Time

5/13/2019 11:46:44 AM

Sample Info.**Data File**

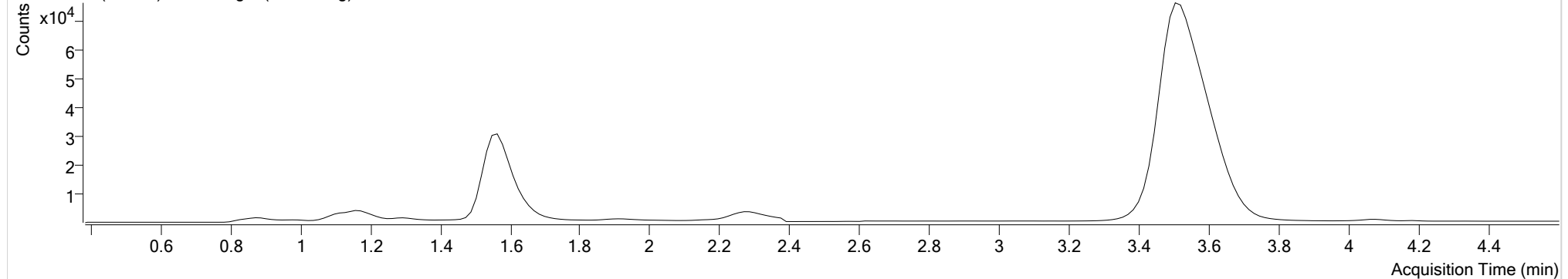
Cal 2- 5ng.d

Sample

Cal 2- 5ng

Comment**Sample Chromatogram**

+ TIC MRM (** -> **) Cal 2- 5ng.d (Cal 2- 5ng)



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.521	31206	242.93	29.0	∞	701106	4.9676 ng/ml
THC-COOH	1.595	7535	14.72	42.9	31.42	42947	4.9065 ng/ml
THC-OH	1.573	11491	154.26	11.1	14.00	116638	5.0476 ng/ml

AM #27 Cannabinoids Quant. Results

**Batch results**

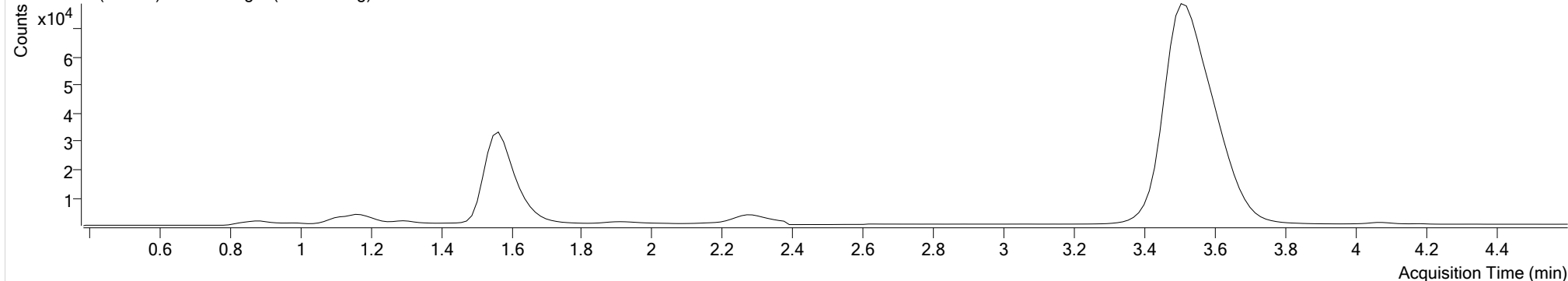
D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin

Calibration Last Update

5/13/2019 3:35:29 PM

Instrument FALCO-LCMS (Property ID 069901)**Type** Cal**Acq. Method** AM 27 THC quant.m**Sample Position** P3-D1**Injection Volume** 10**Acq. Date-Time** 5/13/2019 11:54:19 AM**Sample Info.****Data File** Cal 3 -10ng.d**Sample** Cal 3 -10ng**Comment****Sample Chromatogram**

+ TIC MRM (** -> **) Cal 3 -10ng.d (Cal 3 -10ng)



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.521	59655	∞	27.7	∞	691773	9.9686 ng/ml
THC-COOH	1.595	11941	47.66	45.6	63.71	41912	10.2639 ng/ml
THC-OH	1.573	20347	154.95	12.3	58.25	115603	10.1789 ng/ml

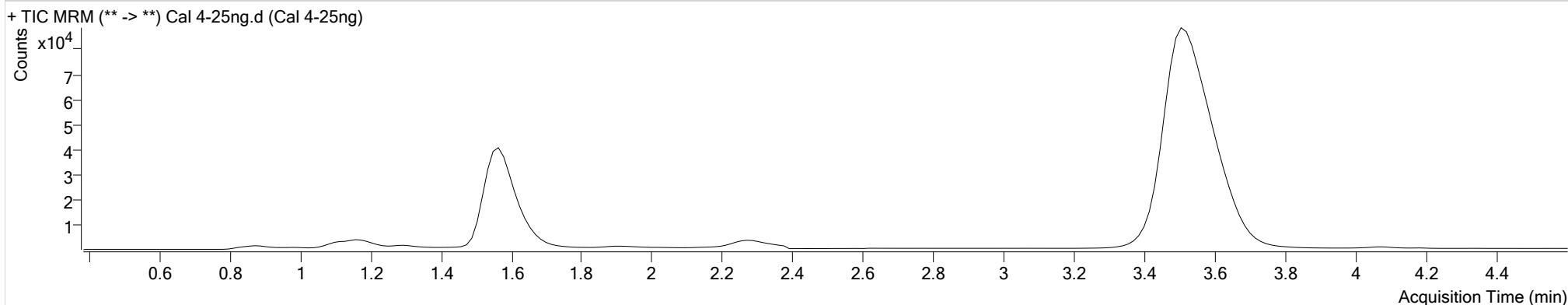
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin
Calibration Last Update 5/13/2019 3:35:29 PM

Instrument Type	FALCO-LCMS (Property ID 069901)	Data File	Cal 4-25ng.d
Acq. Method	AM 27 THC quant.m	Sample	Cal 4-25ng
Sample Position	P3-E1	Comment	
Injection Volume	10		
Acq. Date-Time	5/13/2019 12:01:53 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.521	144437	∞	27.7	∞	673002	25.3563 ng/ml
THC-COOH	1.595	24617	61.15	53.6	552.25	41996	25.0086 ng/ml
THC-OH	1.558	45756	261.58	13.8	224.75	114702	24.9397 ng/ml

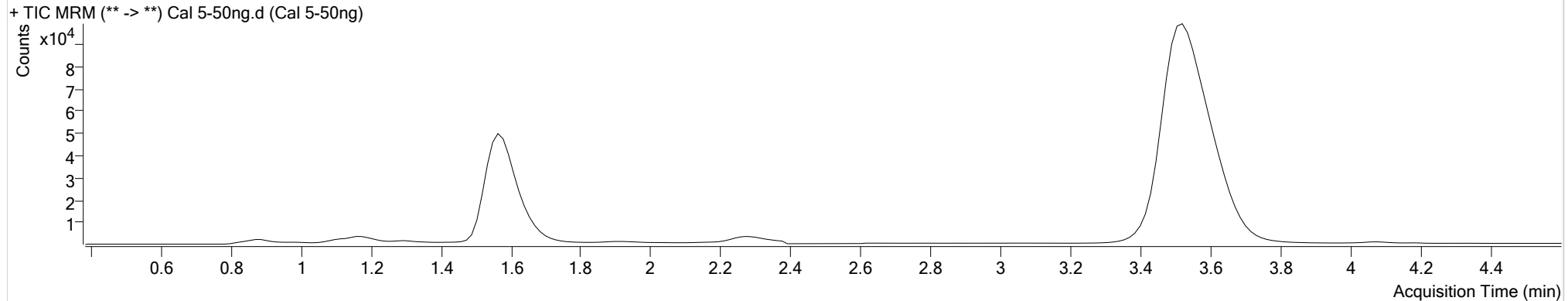
AM #27 Cannabinoids Quant. Results



Batch results D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin
Calibration Last Update 5/13/2019 3:35:29 PM

Instrument FALCO-LCMS (Property ID 069901) **Data File** Cal 5-50ng.d
Type Cal **Sample** Cal 5-50ng
Acq. Method AM 27 THC quant.m
Sample Position P3-F1 **Comment**
Injection Volume 10
Acq. Date-Time 5/13/2019 12:09:28 PM
Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.521	260736	∞	27.8	∞	628161	49.3835 ng/ml
THC-COOH	1.595	41860	71.59	56.6	113.03	38614	49.3757 ng/ml
THC-OH	1.573	81889	3940.41	13.9	314.23	105564	49.8929 ng/ml

AM #27 Cannabinoids Quant. Results

**Batch results**

D:\MassHunter\Data\2019\AM 27\051319 THCQ SP\QuantResults\THCQ.batch.bin

Calibration Last Update

5/13/2019 3:35:29 PM

Instrument

FALCO-LCMS (Property ID 069901)

Type

Cal

Data File

Cal 6-100ng.d

Acq. Method

AM 27 THC quant.m

Sample

Cal 6-100ng

Sample Position

P3-G1

Comment**Injection Volume**

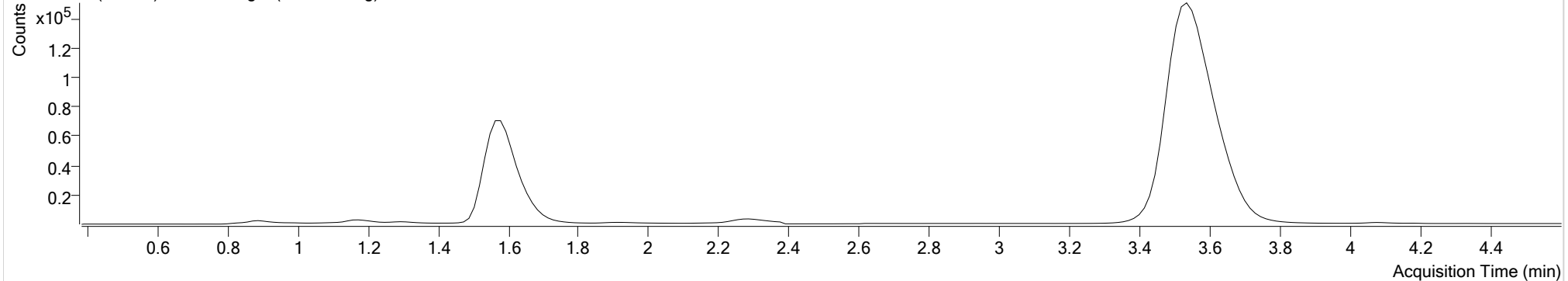
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Acq. Date-Time

5/13/2019 12:17:02 PM

Sample Info.**Sample Chromatogram**

+ TIC MRM (** -> **) Cal 6-100ng.d (Cal 6-100ng)



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.536	583529	∞	27.6	∞	694702	100.3102 ng/ml
THC-COOH	1.595	78295	115.42	57.8	690.33	36801	100.4453 ng/ml
THC-OH	1.573	157632	1919.84	14.0	500.29	102857	100.0098 ng/ml