REVIEWED
By Sarah Pickle at 10:15 am, Apr 15, 2019

TS

4/6/2019

Worklist: 3237

LAB_CASE	ITEM	TASK_ID	DESCRIPTION
M2019-1297	1	147657	AM 27 Blood THC Quant by LC-QQQ
M2019-1311	1	147658	AM 27 Blood THC Quant by LC-QQQ
P2019-0785	2	147659	AM 27 Blood THC Quant by LC-QQQ
P2019-0873	3	147660	AM 27 Blood THC Quant by LC-QQQ
P2019-0959	1	147661	AM 27 Blood THC Quant by LC-QQQ
P2019-0980	1	147662	AM 27 Blood THC Quant by LC-QQQ
P2019-0986	1	147663	AM 27 Blood THC Quant by LC-QQQ
P2019-0987	1	147664	AM 27 Blood THC Quant by LC-QQQ
P2019-0998	1	147665	AM 27 Blood THC Quant by LC-QQQ

15

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

Extraction Date: 04/11/19 Analyst: Tamara Salazar
Plate lot#: 0539904 Plate Expiration: 09/10/19

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: Hemostat 445283-1 **Column**: UCT Selectra DA 100 x 2.1mm 3um
LCMS-QQQ ID: 069910

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.
☒ 3. Create worklist:

Analytic:

☒ 1. Remove standards, plate, controls, and samples from cold storage. Allow to reach room temperature.
☒ 2. Pipette **1000µL blood/urine (calibrated pipette) Pipette ID: 27** in wells of analytical (standards) plate.
☒ 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes. *Shaker ID: 067105*
☒ 4. Pipette **500µL 0.1% formic acid in water** in wells of analytical plate for blood samples.
☒ 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
☒ 6. Transfer **800µL 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 85-100 PSI- Selector to the right)*** Manifold ID: 067104
☒ 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. *SPE Dry ID: 067103*
☒ 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.
Worklist path: D:\MassHunter\Data\2019\AM 27\AM 27 urine validations 041019 CS\reinjections
Batch Name: AM 27 wklst 3237 TS
☒ 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: *Curves limited: THC-COOH 10-100,*
THC-OH reported qualitatively for case sample P2019-0987 due to possible interfering peak.

15

Idaho State Police Forensic Services

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

Methanol External Control Solution (Lot: WS020419)

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	FE04231406	04/30/2019
C-THC	Cerilliant	FE07171501	09/31/2020
THC-OH	Cerilliant	FE01121503	01/31/2020
Prepared:	02/04/19		
Prepared By:	Tamara Salazar		
Expires:	04/30/2019		

Blood External Control Solution (Lot: 020419)

100 ul of methanol external control solution was added to 9900 ul of blood.

Approximately 10ng/mL of each compound.

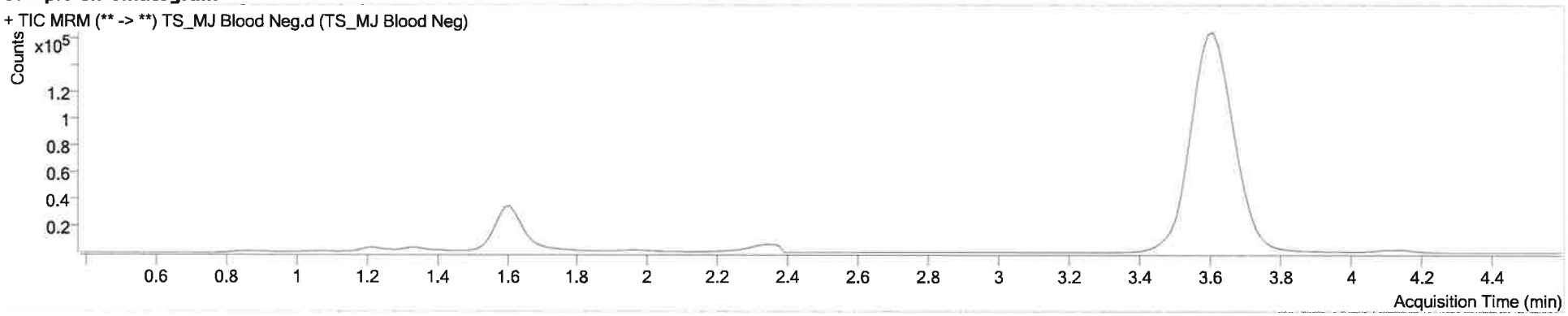
Component	*Source*	*Source Lot Number*
Negative Blood	Hemostat	445283-1
Methanol External Control Solution	-	WS020419
Prepared:	02/04/19	
Prepared by:	Tamara Salazar	
Expires:	04/30/2019	

AM #27 Cannabinoids Quant. Results

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Calibration Last Update	4/12/2019 2:23:48 PM

Instrument	FALCO-LCMS (Property ID 069901)	**Data File**	TS_MJ Blood Neg.d
Type	Sample	**Sample**	TS_MJ Blood Neg
Acq. Method	AM 27 THC quant.m		
Sample Position	P4-A2	**Comment**	
Injection Volume	10		
Acq. Date-Time	4/11/2019 5:29:30 PM		
Sample Info.			

Sample Chromatogram



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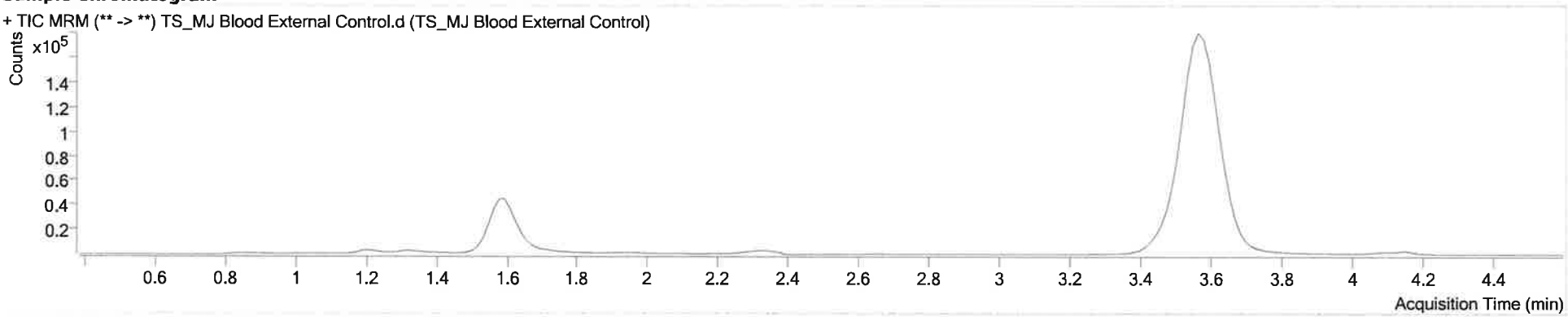
AM #27 Cannabinoids Quant. Results

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Calibration Last Update	4/15/2019 9:48:52 AM

Instrument	FALCO-LCMS (Property ID 069901)	**Data File**	TS_MJ Blood External Control.d
Type	Sample	**Sample**	TS_MJ Blood External Control
Acq. Method	AM 27 THC quant.m		
Sample Position	P4-B2	**Comment**	
Injection Volume	10		
Acq. Date-Time	4/11/2019 5:44:44 PM		
Sample Info.			

Sample Chromatogram



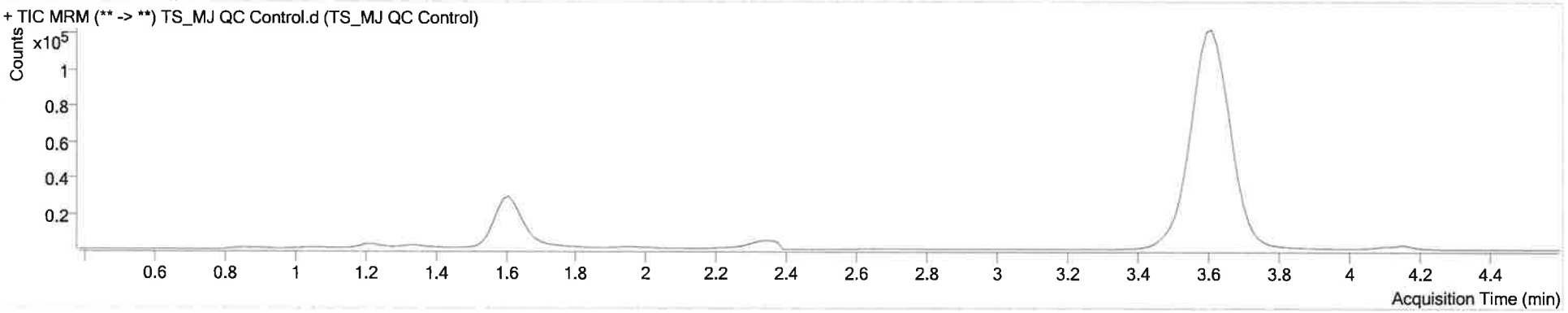
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.582	80828	∞	26.7	196.71	1248568	7.0630 ng/ml
THC-COOH	1.625	13176	92.02	48.7	133.19	52433	8.3908 ng/ml
THC-OH	1.603	21916	202.57	13.3	33.35	153027	7.9250 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Calibration Last Update	4/12/2019 2:23:48 PM

Instrument	FALCO-LCMS (Property ID 069901)	**Data File**	TS_MJ QC Control.d
Type	Sample	**Sample**	TS_MJ QC Control
Acq. Method	AM 27 THC quant.m		
Sample Position	P4-H1	**Comment**	
Injection Volume	10		
Acq. Date-Time	4/11/2019 5:14:20 PM		
Sample Info.			

Sample Chromatogram

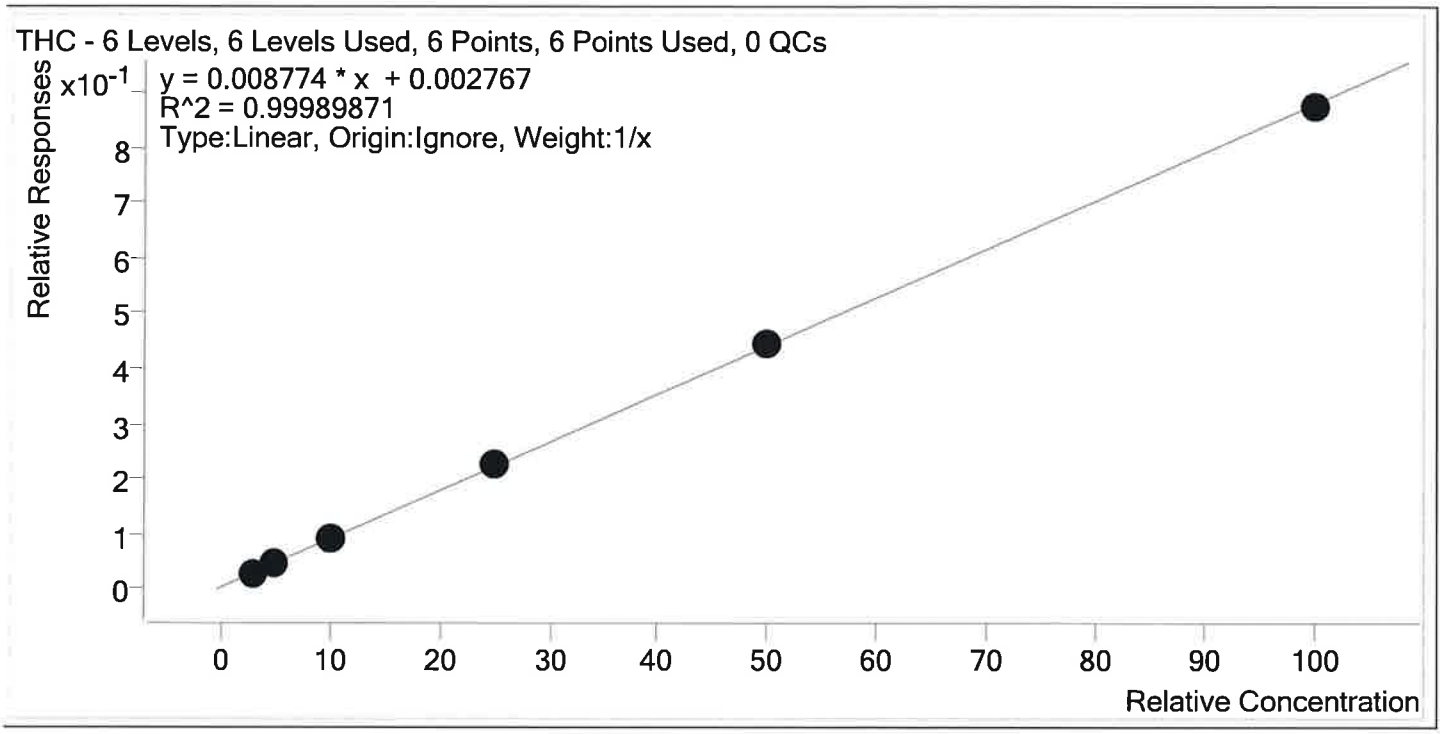


Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.612	39982	384.73	27.8	122.22	879013	4.8688 ng/ml
THC-COOH	1.640	10967	84.85	45.1	83.32	38071	10.1710 ng/ml
THC-OH	1.618	9998	∞	12.2	16.87	101190	4.9444 ng/ml

TS

AM #28 Multi-Drug Quant. Calibration Curve Report

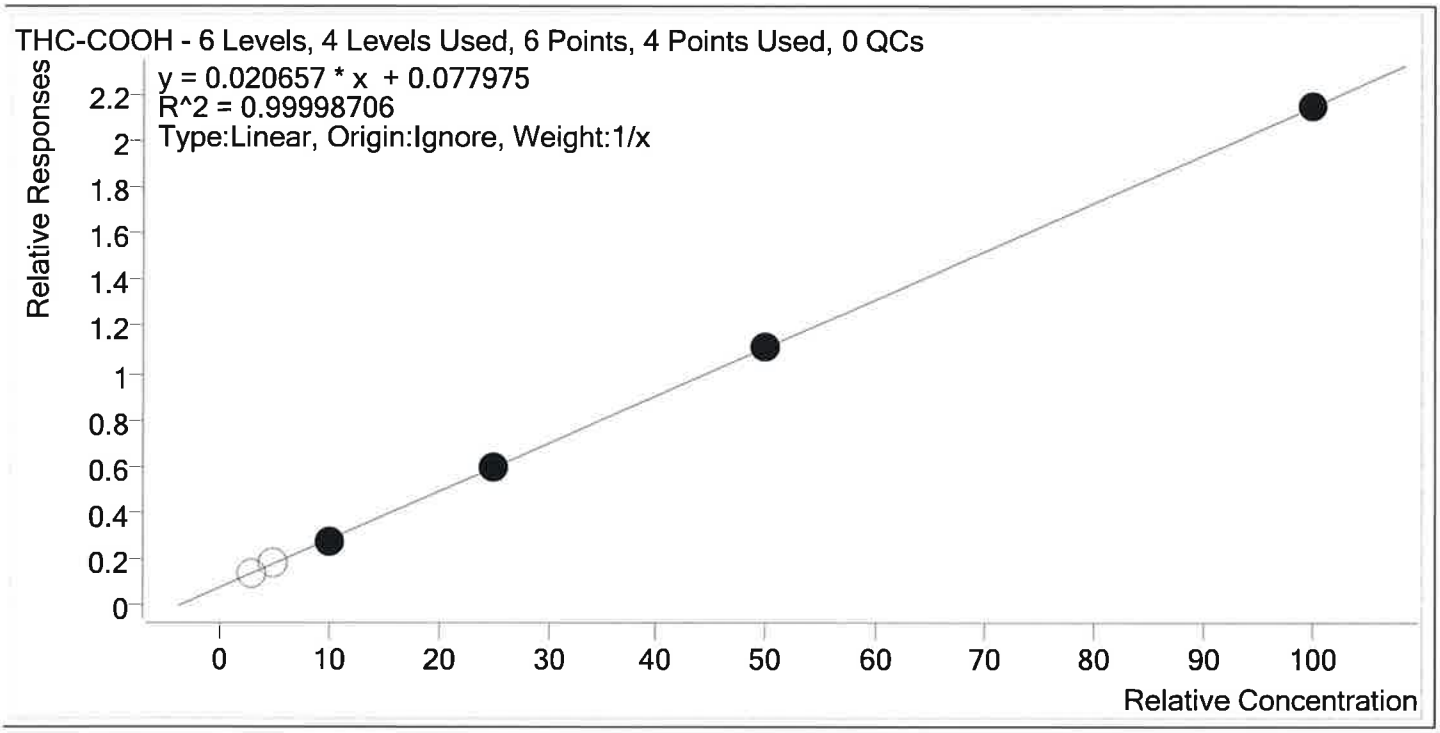
Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Last Cal. Update	4/12/2019 2:23 PM
Analyst Name	ISP\Datastor
Analyte	THC
Internal Standard	THC-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
TS_MJ Cal 1-3ng	1	✓	3.0	3.0	99.4
TS_MJ Cal 2- 5ng	2	✓	5.0	5.0	99.4
TS_MJ Cal 3 -10ng	3	✓	10.0	9.9	99.5
TS_MJ Cal 4-25ng	4	✓	25.0	25.4	101.8
TS_MJ Cal 5-50ng	5	✓	50.0	50.3	100.6
TS_MJ Cal 6-100ng	6	✓	100.0	99.4	99.4

AM #28 Multi-Drug Quant. Calibration Curve Report

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Last Cal. Update	4/12/2019 2:23 PM
Analyst Name	ISP\Datastor
Analyte	THC-COOH
Internal Standard	THC-COOH-D9

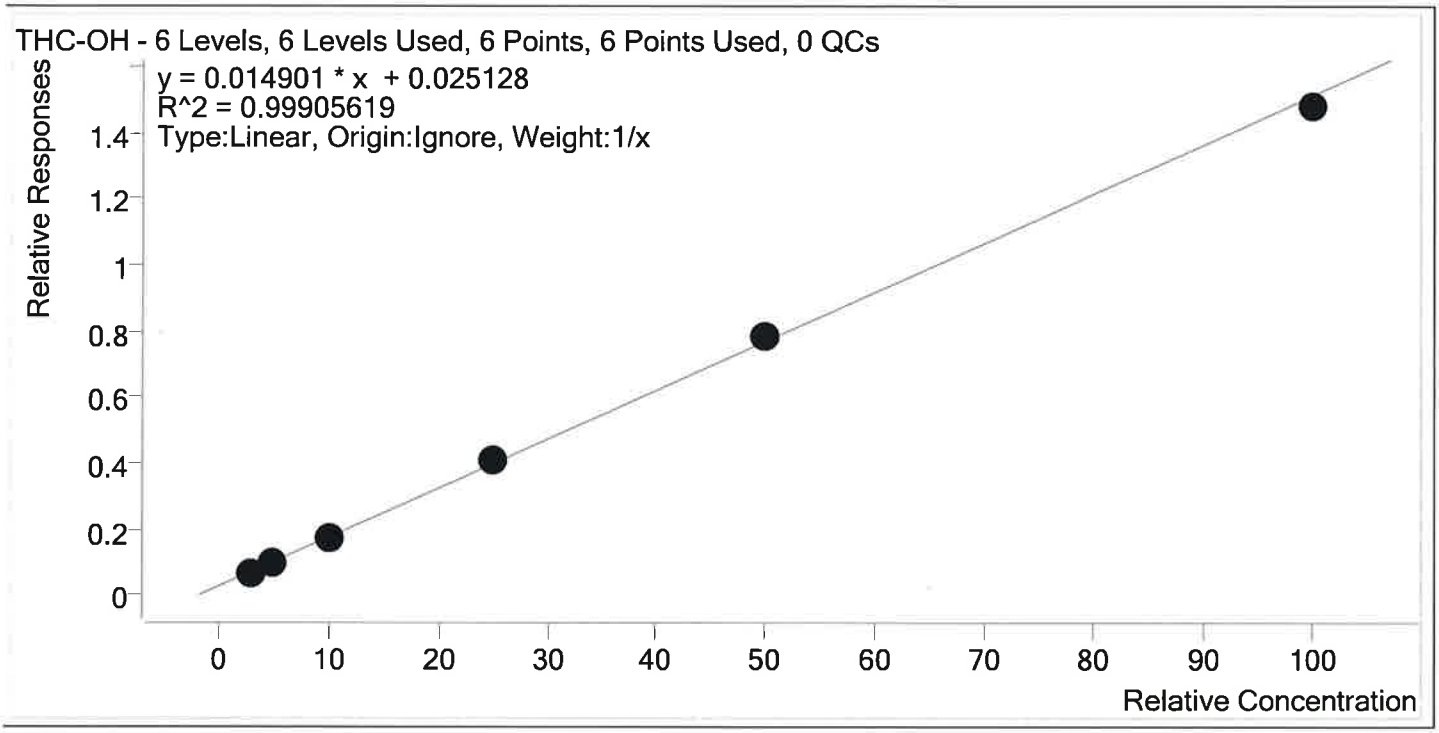


Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
TS_MJ Cal 1-3ng	1	×	3.0	3.2	105.7
TS_MJ Cal 2- 5ng	2	×	5.0	5.2	103.5
TS_MJ Cal 3 -10ng	3	✓	10.0	10.0	99.8
TS_MJ Cal 4-25ng	4	✓	25.0	25.1	100.4
TS_MJ Cal 5-50ng	5	✓	50.0	49.8	99.6
TS_MJ Cal 6-100ng_	6	✓	100.0	100.1	100.1

TS

AM #28 Multi-Drug Quant. Calibration Curve Report

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Last Cal. Update	4/12/2019 2:23 PM
Analyst Name	ISP\Datastor
Analyte	THC-OH
Internal Standard	THC-OH-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
TS_MJ Cal 1-3ng	1	✓	3.0	2.9	95.2
TS_MJ Cal 2- 5ng	2	✓	5.0	5.0	100.7
TS_MJ Cal 3 -10ng	3	✓	10.0	10.0	99.7
TS_MJ Cal 4-25ng	4	✓	25.0	26.0	103.9
TS_MJ Cal 5-50ng	5	✓	50.0	51.4	102.8
TS_MJ Cal 6-100ng_	6	✓	100.0	97.8	97.8

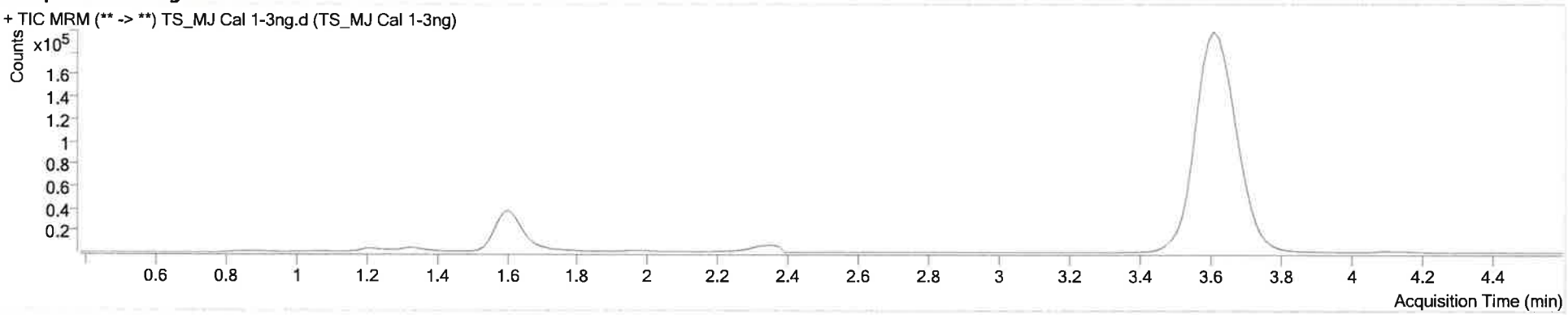
15

AM #27 Cannabinoids Quant. Results

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Calibration Last Update	4/12/2019 2:23:48 PM

Instrument	FALCO-LCMS (Property ID 069901)	**Data File**	TS_MJ Cal 1-3ng.d
Type	Cal	**Sample**	TS_MJ Cal 1-3ng
Acq. Method	AM 27 THC quant.m		
Sample Position	P4-B1	**Comment**	
Injection Volume	10		
Acq. Date-Time	4/11/2019 4:21:18 PM		
Sample Info.			

Sample Chromatogram



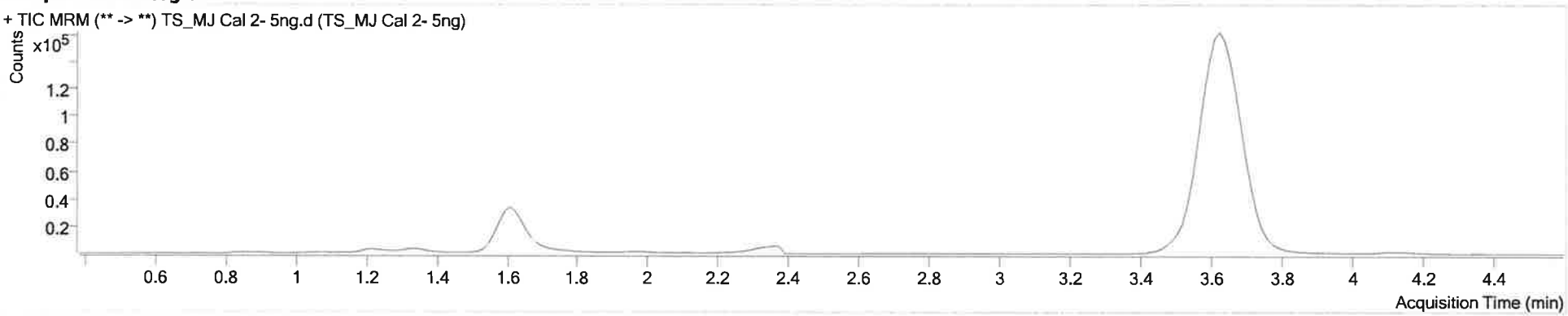
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.627	43590	528.92	28.1	61.39	1506632	2.9822 ng/ml
THC-COOH	1.640	7197	22.44	31.2 Low	37.54	50160	3.1708 ng/ml
THC-OH	1.618	9682	∞	11.0	13.32	143072	2.8551 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Calibration Last Update	4/12/2019 2:23:48 PM

Instrument	FALCO-LCMS (Property ID 069901)	**Data File**	TS_MJ Cal 2- 5ng.d
Type	Cal	**Sample**	TS_MJ Cal 2- 5ng
Acq. Method	AM 27 THC quant.m		
Sample Position	P4-C1	**Comment**	
Injection Volume	10		
Acq. Date-Time	4/11/2019 4:28:52 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.642	56671	339.69	27.9	77.61	1222000	4.9704 ng/ml
THC-COOH	1.640	8280	70.94	36.0 Low	40.64	44783	5.1756 ng/ml
THC-OH	1.618	12252	106.18	12.2	29.38	122344	5.0344 ng/ml

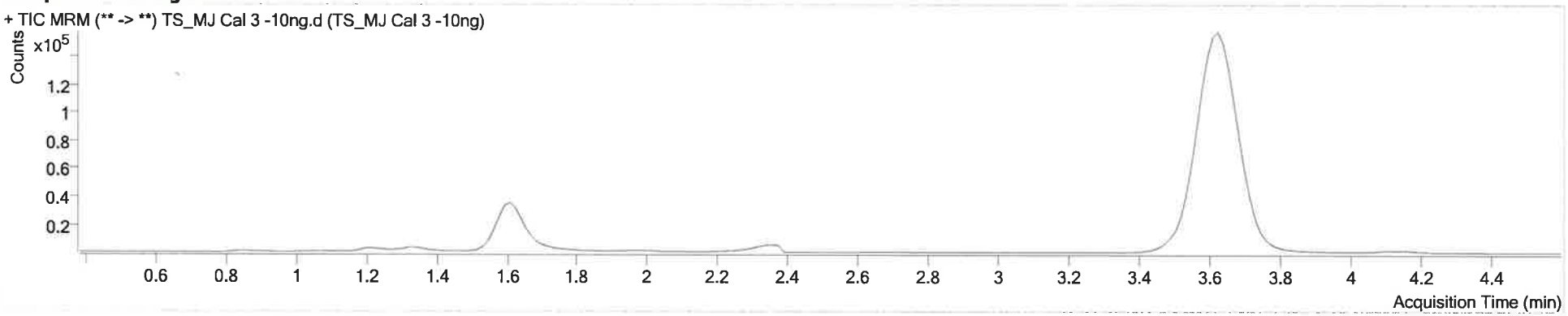
P1

AM #27 Cannabinoids Quant. Results

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Calibration Last Update	4/12/2019 2:23:48 PM

Instrument	FALCO-LCMS (Property ID 069901)	**Data File**	TS_MJ Cal 3 -10ng.d
Type	Cal	**Sample**	TS_MJ Cal 3 -10ng
Acq. Method	AM 27 THC quant.m		
Sample Position	P4-D1	**Comment**	
Injection Volume	10		
Acq. Date-Time	4/11/2019 4:36:26 PM		
Sample Info.			

Sample Chromatogram



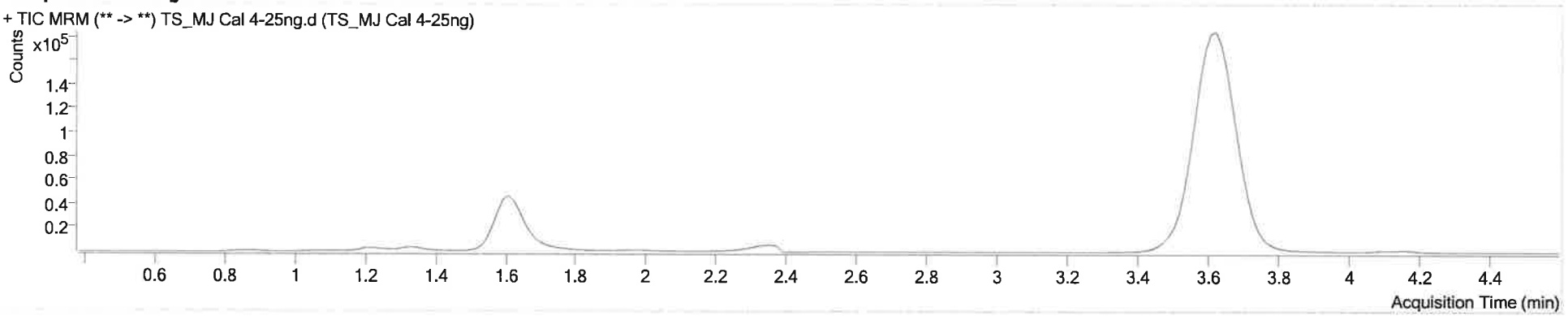
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.627	101132	3437.25	27.5	247.11	1123375	9.9453 ng/ml
THC-COOH	1.640	12018	85.93	45.0	52.97	42284	9.9846 ng/ml
THC-OH	1.618	21047	∞	12.9	59.89	121214	9.9667 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Calibration Last Update	4/12/2019 2:23:48 PM

Instrument	FALCO-LCMS (Property ID 069901)	**Data File**	TS_MJ Cal 4-25ng.d
Type	Cal	**Sample**	TS_MJ Cal 4-25ng
Acq. Method	AM 27 THC quant.m		
Sample Position	P4-E1	**Comment**	
Injection Volume	10		
Acq. Date-Time	4/11/2019 4:44:00 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.627	261092	∞	27.6	907.62	1155319	25.4421 ng/ml
THC-COOH	1.640	26010	430.76	54.1	240.06	43600	25.1054 ng/ml
THC-OH	1.618	50329	224.10	14.2	212.53	122084	25.9803 ng/ml

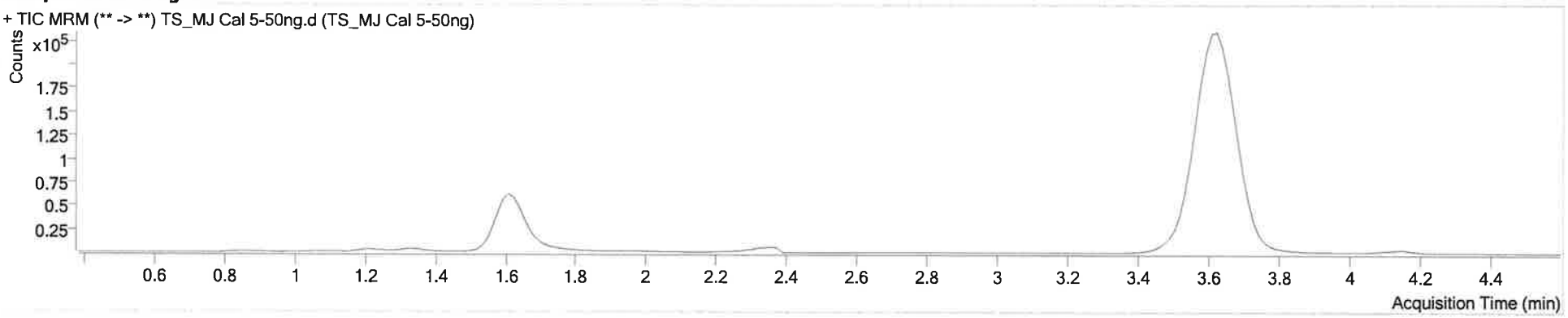
15

AM #27 Cannabinoids Quant. Results

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Calibration Last Update	4/12/2019 2:23:48 PM

Instrument	FALCO-LCMS (Property ID 069901)	**Data File**	TS_MJ Cal 5-50ng.d
Type	Cal	**Sample**	TS_MJ Cal 5-50ng
Acq. Method	AM 27 THC quant.m		
Sample Position	P4-F1	**Comment**	
Injection Volume	10		
Acq. Date-Time	4/11/2019 4:51:34 PM		
Sample Info.			

Sample Chromatogram



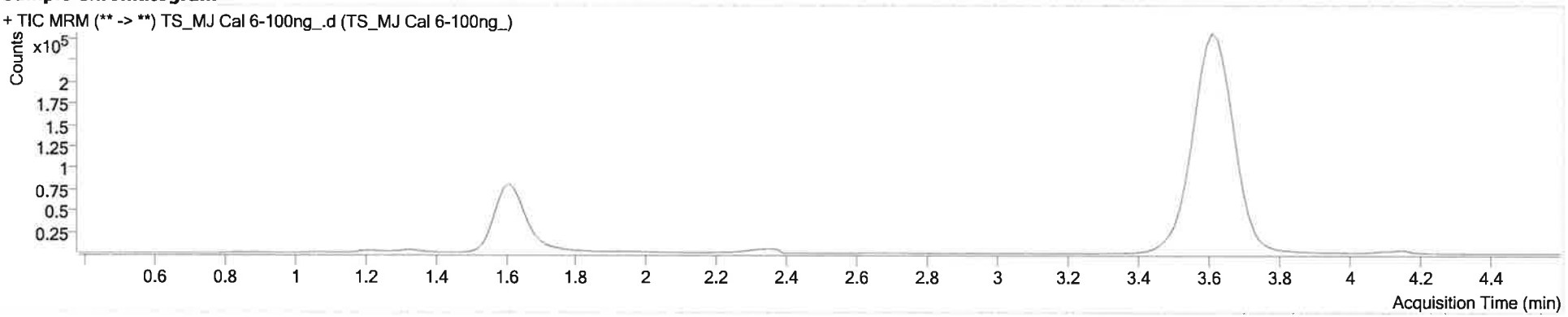
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.627	531046	∞	27.2	1721.58	1195739	50.3029 ng/ml
THC-COOH	1.640	49053	519.99	56.8	245.42	44306	49.8224 ng/ml
THC-OH	1.618	98728	478.25	14.3	310.22	124836	51.3897 ng/ml

AM #27 Cannabinoids Quant. Results

Batch results	D:\MassHunter\Data\2019\Urine Validations\AM 27 urine validations 041019 CS\reinjections\QuantResults\AM 27 wklst 3237 TS.batch.bin
Calibration Last Update	4/12/2019 2:23:48 PM

Instrument	FALCO-LCMS (Property ID 069901)	**Data File**	TS_MJ Cal 6-100ng_.d
Type	Cal	**Sample**	TS_MJ Cal 6-100ng_
Acq. Method	AM 27 THC quant.m		
Sample Position	P4-G1	**Comment**	
Injection Volume	10		
Acq. Date-Time	4/11/2019 4:59:10 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.627	833831	∞	27.2	∞	953485	99.3571 ng/ml
THC-COOH	1.640	81612	110.47	57.4	345.76	38040	100.0876 ng/ml
THC-OH	1.603	171840	1139.10	14.3	734.07	115950	97.7738 ng/ml