

**REVIEWED**

By Celena Shrum at 3:04 pm, Jul 12, 2019

**REVIEWED**

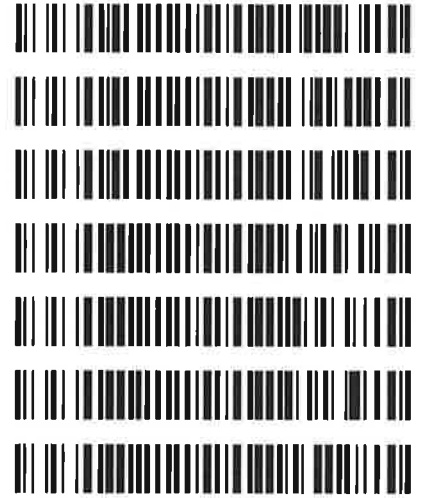
By Sarah Pickle at 12:42 pm, Jul 12, 2019

7/8/2019

TS

**Worklist: 3530**

<u>LAB CASE</u>	<u>ITEM</u>	<u>TASK ID</u>	<u>DESCRIPTION</u>
M2019-2723	1	156635	AM 27 Blood THC Quant by LC-QQQ
M2019-2982	3	156636	AM 27 Blood THC Quant by LC-QQQ
M2019-2991	5	156637	AM 27 Blood THC Quant by LC-QQQ
P2019-1712	1	156641	AM 27 Blood THC Quant by LC-QQQ
P2019-1860	1	156638	AM 27 Blood THC Quant by LC-QQQ
P2019-1897	1	156639	AM 27 Blood THC Quant by LC-QQQ
P2019-1991	1	156640	AM 27 Blood THC Quant by LC-QQQ



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

Extraction Date: 07/09/19  
Plate lot#: 0539904

Analyst: Tamara Salazar  
Plate Expiration: 09/10/19

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

**Mobile phase B:** 0.1% Formic acid in Acetonitrile  
Hexane

**Blank Blood Lot:** Hemostat 445283-2  
**LCMS-QQQ ID:** 069901

**Column:** UCT Selectra DA 100 x 2.1mm 3um

## 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: 3** 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\070919 THCQ TS  
Batch Name: wklst 3530 THCQ TS
- 2. Make any necessary integration changes, Curve weighting of Linear 1/x with  $r^2$  values  $\geq 0.98$  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: *THC-COOH: 5-100*



# Idaho State Police Forensic Services

TS

## 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.

<i>Component</i>	<i>Source</i>	<i>Source Lot Number</i>	<i>Expiration Date</i>
Methanol (LCMS)	Fisher	184782	
THC	Cerilliant	FE09101501	11/30/2020
C-THC	Cerilliant	FE07171501	09/30/2020
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.

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



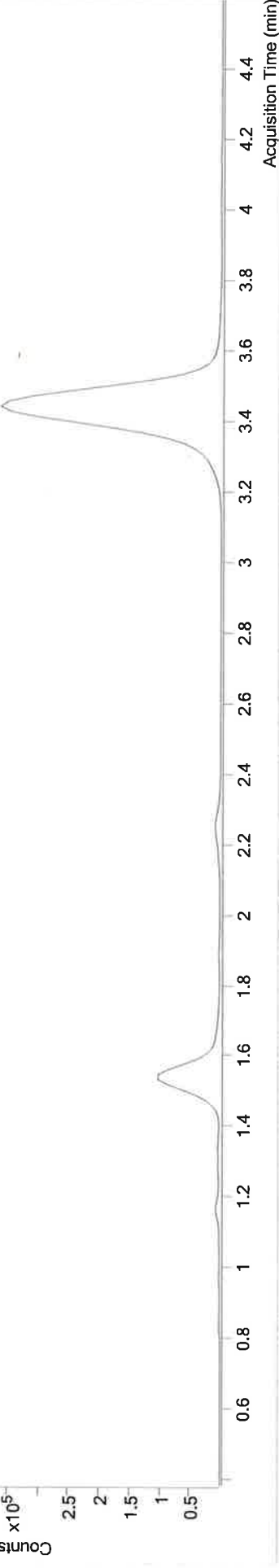
# AM #27 Cannabinoids Quant. Results

Batch results Calibration Last Update D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklist 3530 THCQ TS.batch.bin  
7/11/2019 10:56:46 AM

<b>Instrument Type</b>	Falco Sample	Negative.d
<b>Acq. Method</b>	AM 27 THC quant.m	Negative
<b>Sample Position</b>	P3-A2	
<b>Injection Volume</b>	10	
<b>Acq. Date-Time</b>	7/9/2019 1:24:21 PM	
<b>Sample Info.</b>		

## Sample Chromatogram

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



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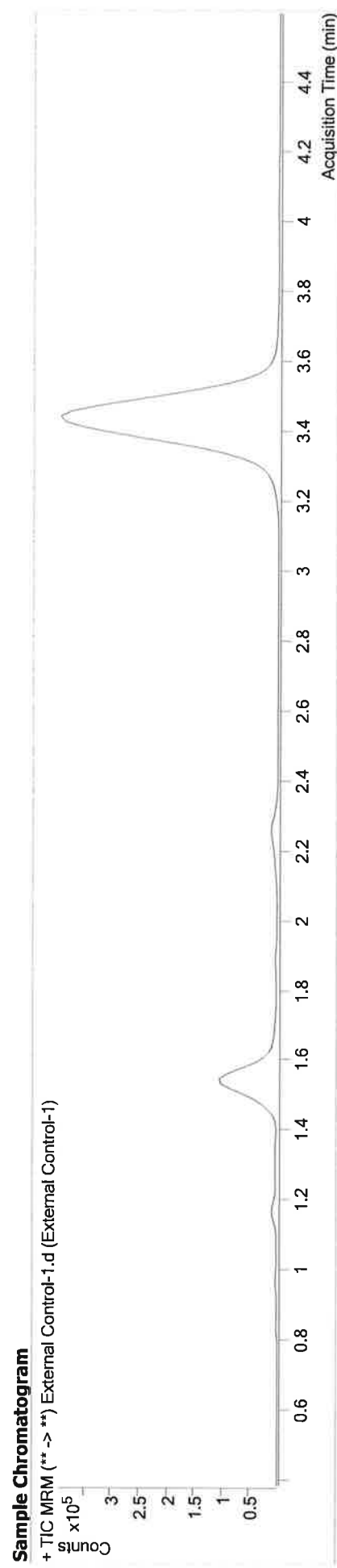
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# AM #27 Cannabinoids Quant. Results

Batch results Calibration Last Update D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklist 3530 THCQ TS.batch.bin  
7/11/2019 10:56:46 AM

<b>Instrument Type</b>	Falco	<b>Data File Sample</b>	External Control-1.d
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Sample</b>	External Control-1
<b>Sample Position</b>	P3-B2	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/9/2019 1:39:32 PM		
<b>Sample Info.</b>			



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.461	203475	1448.38	26.7	322.06	3273370	7.0994 ng/ml
THC-COOH	1.580	31774	∞	43.0	153.03	138687	9.4568 ng/ml
THC-OH	1.558	56277	∞	11.1	104.26	382616	8.1690 ng/ml

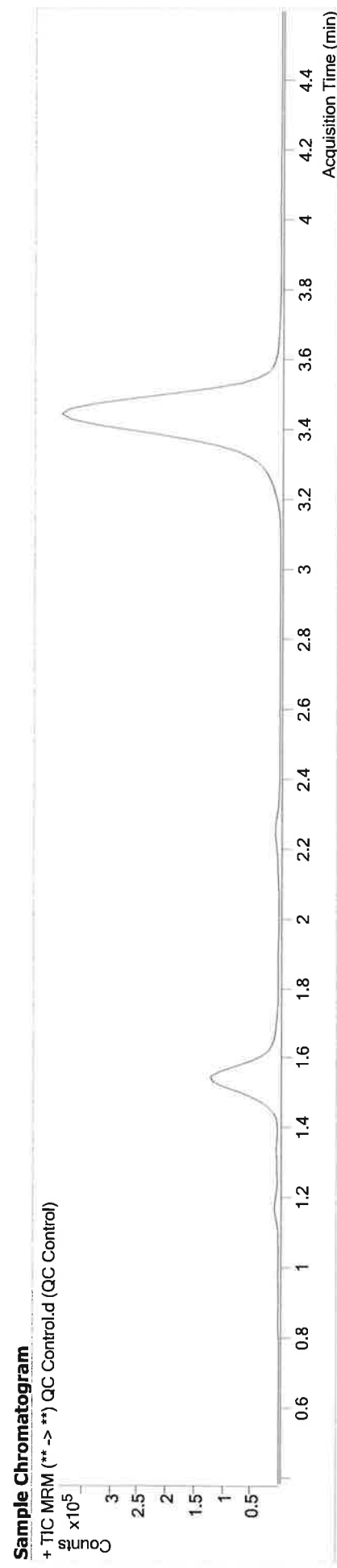
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# AM #27 Cannabinoids Quant. Results

Batch results Calibration Last Update D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklist 3530 THCQ TS.batch.bin 7/11/2019 10:56:46 AM

<b>Instrument Type</b>	Falco	<b>Data File</b>	QC Control.d
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Sample</b>	QC Control
<b>Sample Position</b>	P3-H1	<b>Comment</b>	
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/9/2019 1:09:08 PM		
<b>Sample Info.</b>			

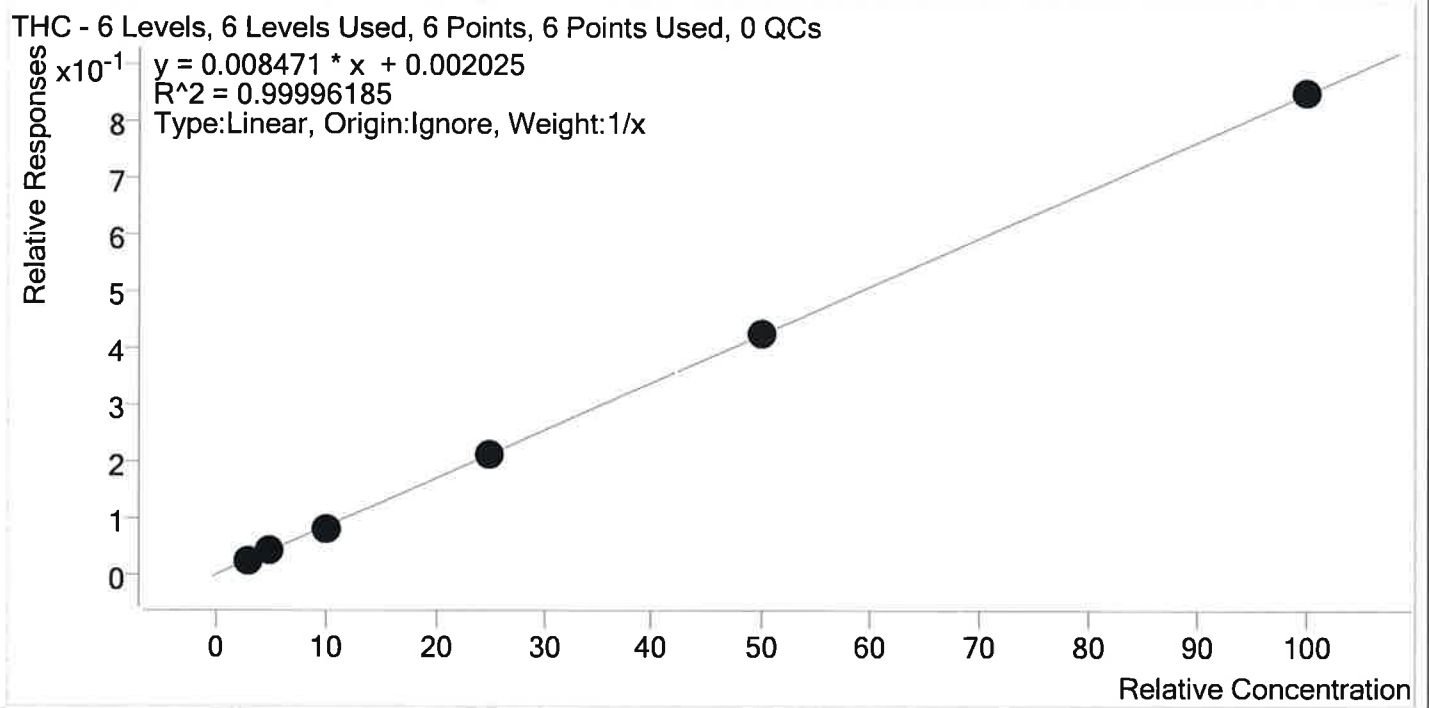


Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.461	130405	1015.03	27.5	132.77	3052887	4.8038 ng/ml
THC-COOH	1.580	32342	∞	56.5	270.39	155524	8.3340 ng/ml
THC-OH	1.558	46795	∞	11.3	25.26	473629	4.9610 ng/ml



# AM #27 Cannabinoids Quant. Calibration Curve Report

**Batch results** D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklst 3530 THCQ TS.batch.bin  
**Last Cal. Update** 7/11/2019 10:56 AM  
**Analyst Name** ISP\datastor  
**Analyte** THC **Internal Standard** THC-D3



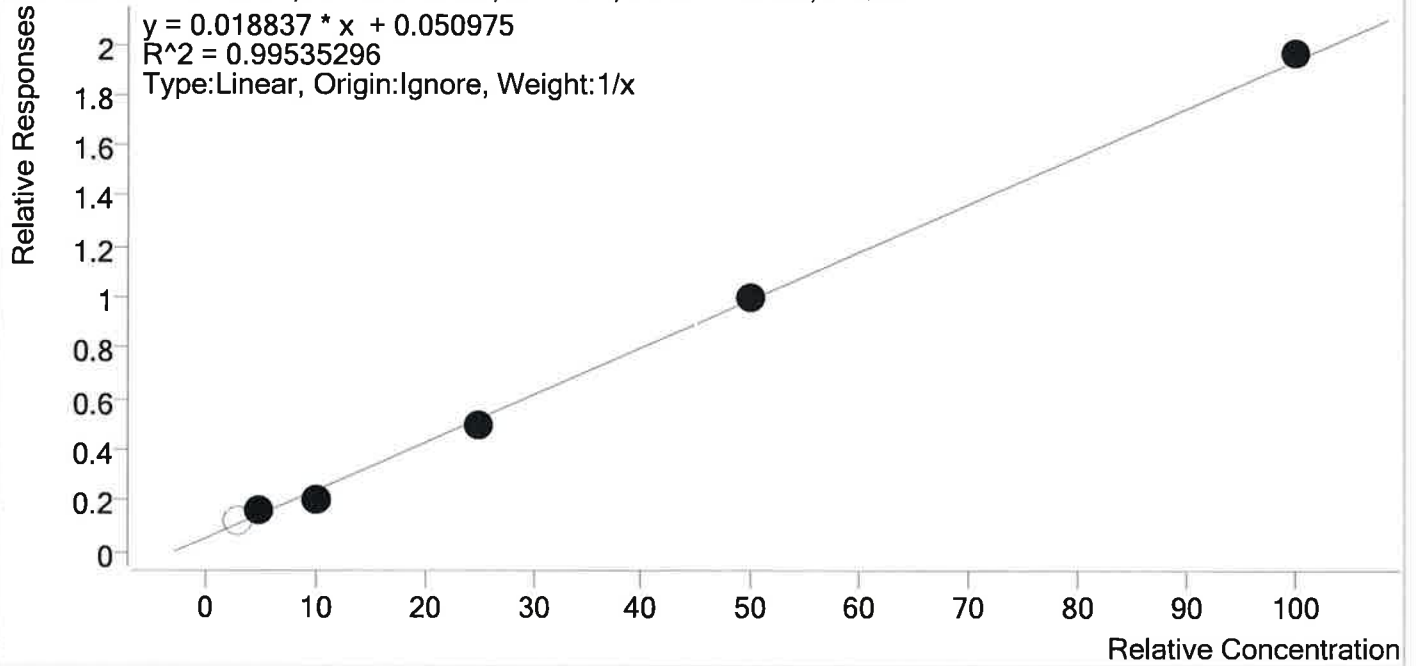
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1-3ng	1	✓	3.0	3.1	102.1
Cal 2- 5ng	2	✓	5.0	5.0	99.1
Cal 3 -10ng	3	✓	10.0	9.8	98.5
Cal 4-25ng	4	✓	25.0	25.1	100.5
Cal 5-50ng	5	✓	50.0	49.9	99.7
Cal 6-100ng	6	✓	100.0	100.2	100.2



# AM #27 Cannabinoids Quant. Calibration Curve Report

**Batch results** D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklst 3530 THCQ TS.batch.bin  
**Last Cal. Update** 7/11/2019 10:56 AM  
**Analyst Name** ISP\datastor  
**Analyte** THC-COOH **Internal Standard** THC-COOH-D9

THC-COOH - 6 Levels, 5 Levels Used, 6 Points, 5 Points Used, 0 QCs



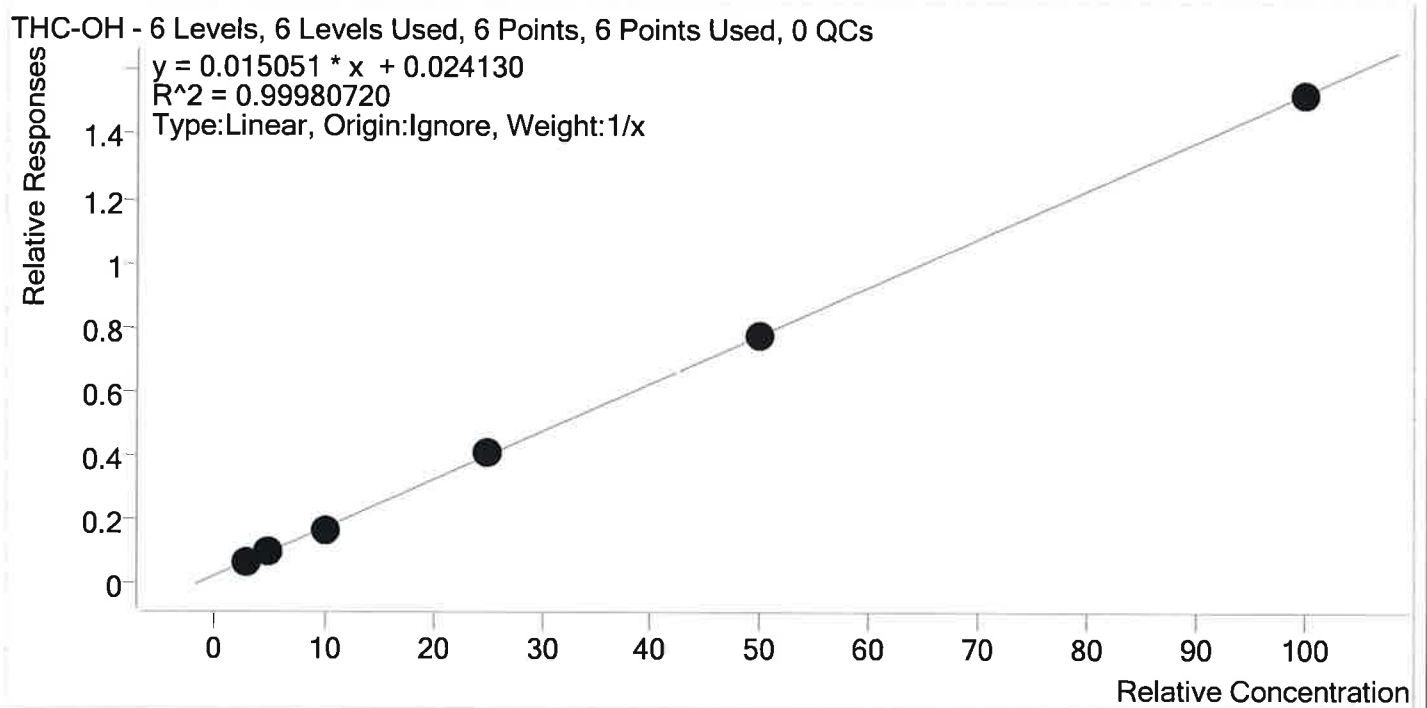
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
Cal 1-3ng	1	×	3.0	3.9	128.6
Cal 2- 5ng	2	✓	5.0	6.0	119.6
Cal 3 -10ng	3	✓	10.0	8.3	82.9
Cal 4-25ng	4	✓	25.0	23.8	95.1
Cal 5-50ng	5	✓	50.0	50.4	100.9
Cal 6-100ng	6	✓	100.0	101.5	101.5





# AM #27 Cannabinoids Quant. Calibration Curve Report

**Batch results** D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklst 3530 THCQ TS.batch.bin  
**Last Cal. Update** 7/11/2019 10:56 AM  
**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	3.1	101.9
Cal 2- 5ng	2	✓	5.0	4.9	97.3
Cal 3 -10ng	3	✓	10.0	9.9	98.6
Cal 4-25ng	4	✓	25.0	25.7	102.8
Cal 5-50ng	5	✓	50.0	49.9	99.7
Cal 6-100ng	6	✓	100.0	99.7	99.7



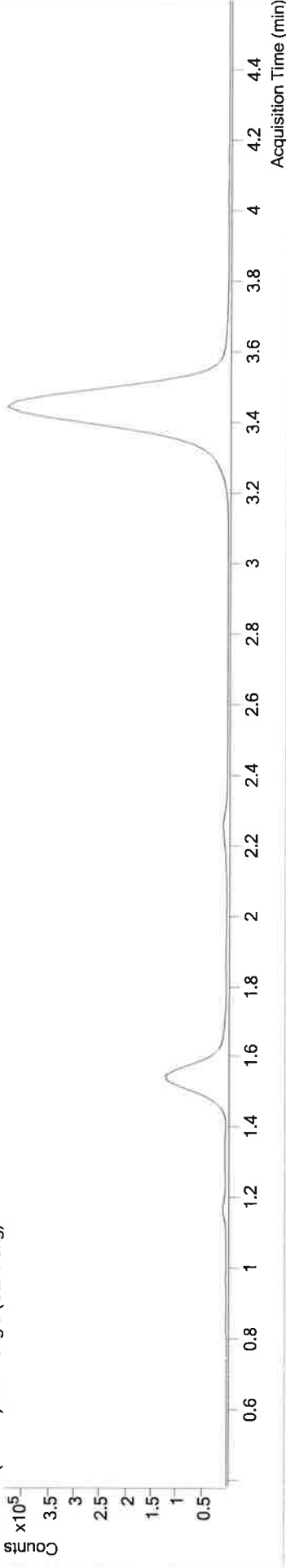
# AM #27 Cannabinoids Quant. Results

Batch results Calibration Last Update D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklist 3530 THCQ TS.batch.bin  
7/11/2019 10:56:46 AM

<b>Instrument</b>	Falco	<b>Data File</b>	Cal 1-3ng.d
<b>Type</b>	Cal	<b>Sample</b>	Cal 1-3ng
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Comment</b>	
<b>Sample Position</b>	P3-B1		
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/9/2019 12:23:43 PM		

### Sample Chromatogram

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



Name	RT	Resp.	S/N	Ratio	ISTD Resp.	Final Conc.
THC	3.461	92584	803.85	28.4	3309767	3.0633 ng/ml
THC-COOH	1.580	19882	∞	32.8 <b>Low</b>	160764	3.8594 ng/ml
THC-OH	1.558	33633	∞	9.8	479447	3.0575 ng/ml

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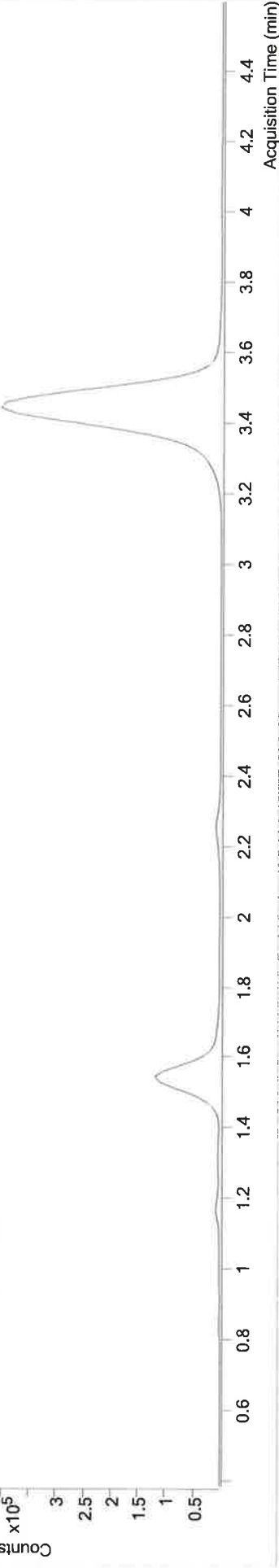
# AM #27 Cannabinoids Quant. Results

Batch results D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklist 3530 THCQ TS.batch.bin  
Calibration Last Update 7/11/2019 10:56:46 AM

Instrument	Falco	Data File	Cal 2- 5ng.d
Type	Cal	Sample	Cal 2- 5ng
Acq. Method	AM 27 THC quant.m	Comment	
Sample Position	P3-C1		
Injection Volume	10		
Acq. Date-Time	7/9/2019 12:31:17 PM		
Sample Info.			

## Sample Chromatogram

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



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.461	133416	953.55	28.1	185.65	3033645	4.9529 ng/ml
THC-COOH	1.580	24908	∞	40.8	97.77	152227	5.9803 ng/ml
THC-OH	1.558	44462	∞	10.9	37.93	456559	4.8670 ng/ml

TS



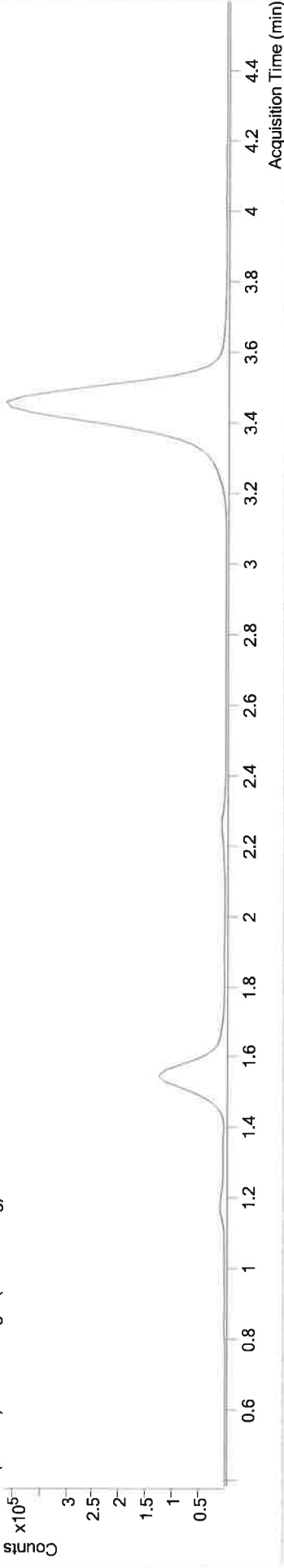
# AM #27 Cannabinoids Quant. Results

Batch results Calibration Last Update D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklist 3530 THCQ TS.batch.bin  
7/11/2019 10:56:46 AM

<b>Instrument</b>	Falco	<b>Data File</b>	Cal 3 -10ng.d
<b>Type</b>	Cal	<b>Sample</b>	Cal 3 -10ng
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Comment</b>	
<b>Sample Position</b>	P3-D1		
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/9/2019 12:38:51 PM		

### Sample Chromatogram

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



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.461	259009	1038.05	27.4	464.91	3031580	9.8473 ng/ml
THC-COOH	1.580	31871	230.37	56.8	271.54	153879	8.2893 ng/ml
THC-OH	1.558	78786	∞	12.9	318.72	456725	9.8577 ng/ml

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# AM #27 Cannabinoids Quant. Results

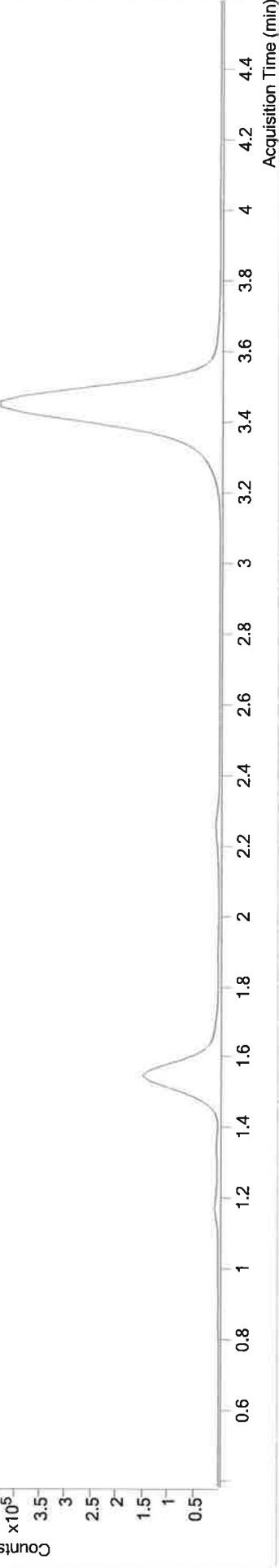
Batch results D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklist 3530 THCQ TS.batch.bin  
 Calibration Last Update 7/11/2019 10:56:46 AM

**Instrument** Falco  
**Type** Cal  
**Acq. Method** AM 27 THC quant.m  
**Sample Position** P3-E1  
**Injection Volume** 10  
**Acq. Date-Time** 7/9/2019 12:46:25 PM  
**Sample Info.**

**Data File** Cal 4-25ng.d  
**Sample** Cal 4-25ng  
**Comment**

**Sample Chromatogram**

+ TIC MRM (\*\* -> \*\*) Cal 4-25ng.d (Cal 4-25ng)



Name	RT	Resp.	S/N	Ratio	ISTD Resp.	Final Conc.
THC	3.461	579160	3433.59	27.4	2695888	25.1229 ng/ml
THC-COOH	1.580	70335	∞	57.9	141004	23.7751 ng/ml
THC-OH	1.558	175532	∞	12.9	427263	25.6920 ng/ml

TS



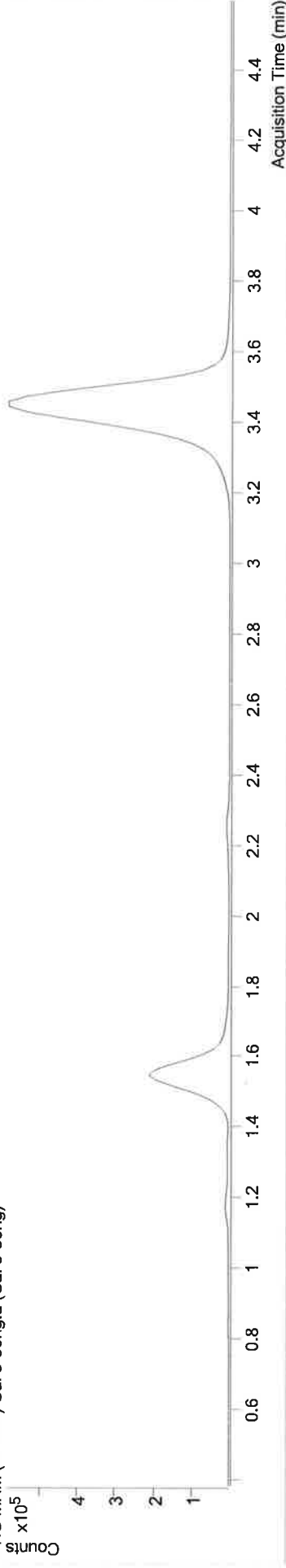
# AM #27 Cannabinoids Quant. Results

Batch results Calibration Last Update D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklist 3530 THCQ TS.batch.bin  
7/11/2019 10:56:46 AM

Instrument Type Falco  
Data File Sample Cal 5-50ng.d  
Acq. Method AM 27 THC quant.m  
Sample Position P3-F1  
Injection Volume 10  
Acq. Date-Time 7/9/2019 12:53:59 PM  
Sample Info. Comment

### Sample Chromatogram

+ TIC MRM (\*\* -> \*\*) Cal 5-50ng.d (Cal 5-50ng)



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.461	1316334	49928.74	27.1	3337.02	3102323	49.8526 ng/ml
THC-COOH	1.580	149714	∞	56.5	1435.49	149537	50.4451 ng/ml
THC-OH	1.558	364751	∞	13.1	833.60	470787	49.8718 ng/ml

B



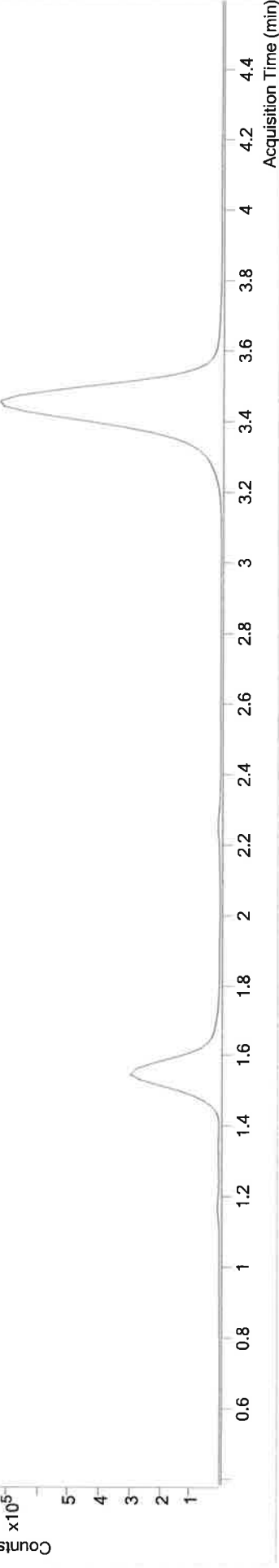
# AM #27 Cannabinoids Quant. Results

Batch results D:\MassHunter\Data\2019\AM 27\070919 THCQ TS\QuantResults\wklist 3530 THCQ TS.batch.bin  
Calibration Last Update 7/11/2019 10:56:46 AM

<b>Instrument Type</b>	Falco Cal	<b>Data File Sample</b>	Cal 6-100ng.d
<b>Acq. Method</b>	AM 27 THC quant.m	<b>Comment</b>	Cal 6-100ng
<b>Sample Position</b>	P3-G1		
<b>Injection Volume</b>	10		
<b>Acq. Date-Time</b>	7/9/2019 1:01:34 PM		

### Sample Chromatogram

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



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC	3.461	2366206	23040.72	27.4	2931.18	2782305	100.1610 ng/ml
THC-COOH	1.580	279717	∞	58.0	∞	142489	101.5101 ng/ml
THC-OH	1.558	671603	∞	13.5	1694.72	440668	99.6540 ng/ml

TS