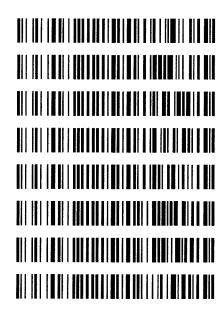
3/23/2018



Worklist: 2274

LAB CASE C2018-0234	ITEM 1	TASK ID 111055	DESCRIPTION AM 27 Blood THC Quant by LC-QQQ
C2018-0264	1	111054	AM 27 Blood THC Quant by LC-QQQ
C2018-0266	1	111053	AM 27 Blood THC Quant by LC-QQQ
C2018-0315	1	111052	AM 27 Blood THC Quant by LC-QQQ
C2018-0317	1	111051	AM 27 Blood THC Quant by LC-QQQ
C2018-0497	2	111050	AM 27 Blood THC Quant by LC-QQQ
C2018-0505	1	111049	AM 27 Blood THC Quant by LC-QQQ
C2018-0525	1	111048	AM 27 Blood THC Quant by LC-QQQ



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

Extraction Date: 3-22-18 Analyst: Anno North

Plate lot#: 0515037 Plate Expiration: 9/28/2018

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: 17J0718 Column: UCT Selectra DA 100 x 2.1mm 3um

LCMS-QQQ ID: 62340

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.
- 4 2. Pipette 1000μL blood (calibrated pipette) Pipette ID: k52558g in wells of analytical (standards) plate.
- 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes. Shaker ID: 66759
- 4. Pipette 500μL 0.1% formic acid in water in wells of analytical plate.
- ☑ 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- Σ 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: 66792
- 🖾 8. Wait 5 minutes.
- ☑ 9. Add 2.25mL MTBE. (Add in 3 increments of 750uL)
- \square 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)
- 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: 66819
- 16. Reconstitute in 100μL 100% MeOH and heat seal plate with foil. Place in autosampler and run worklist.

Post-Analytic

 \square 1. Create batch and process data.

Worklist path: 03222018 can quant Batch Name: 32218 can quant

- \triangle 2. Make any necessary integration changes, Curve weighting of Linear 1/x with r^2 values ≥ 0.98 for each analyte
- △ 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
- 27 Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS:

A

Request for Departure from an Analytical Method

Date of Request 03/23/18

Forensic Scientist

Anne Nord

Analytical Method

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

4.3.2.4 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 the control over 10 ng falls outside the accuracy range at the analyst's discretion the compound may be reported qualitatively.

Request

I am requesting a deviation to report the results for THC and THC-OH from worklist 2274 qualitatively due to the external control concentrations of THC being outside the accuracy range of 30% with a value of 6.16 ng/ml.

Discipline Leader Review		
☑ Departure approved Comments:		
Departure Not Approved Comments:		

Date: 03/23/2018

Celena Shrum Toxicology Discipline Lead

aleron Shrum





Stock solution 1mg/ml 10 ul THC, 100 ug/ml 100 ul each THC-OH, C-THC in 9880 ul meOH working solution 1 ug/ml in meoh C-THC, THC-OH, THC by AMN Toxicology AM method 27 external prep information Ppd 3/19/18 Exp: 9/19/18 lot 91918

 Drug
 lot (cerilliant)
 expiration

 C-THC
 FE07171501
 9/1/2020

 THC-OH
 FE07221601
 7/1/2021

 THC
 FE01041701
 3/1/2022

by AMN Concentration 10 ng/ml each AM 27 control 100 ul working solution lot (91918) in 9900 ul blood lot (17J20718) lot 61918 ppd 3/19/18 Exp 6/19/18

Batch Data Path

D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Analysis Time

3/23/2018 9:09 AM

Analyst Name ISP Tox

Report Time Last Calib Update 3/23/2018 9:11 AM

Reporter Name ISP Tox

3/23/2018 9:09 AM

Batch State Processed

Analysis Info

Acq Time

2018-03-22 15:12

Data File

Negative Control.d

Sample Type

Sample

Sample Name

Negative Control

Dilution

Acq Method

AM 27 Quant THC 7-2017.m

Position

P1-A2

Sample Info

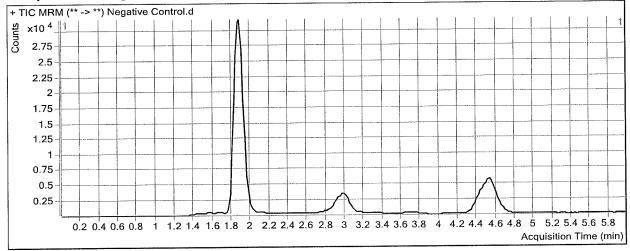
Inj Vol

-1

Comment

AM 27 Cannabinoid Confirmation

Sample Chromatogram



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Compound THC-OH

ISTD Compound

THC-OH-d3

RT 2.075 Response 389

ISTD Resp 170596

Resp Ratio 0.0023

Final Conc 0.1075



Batch Data Path

D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Analysis Time

3/23/2018 9:09 AM

Analyst Name ISP Tox

Report Time Last Calib Update 3/23/2018 9:11 AM 3/23/2018 9:09 AM Reporter Name ISP Tox

Batch State Processed

Analysis Info

Acq Time Sample Type 2018-03-22 15:36 QC

Data File Sample Name External Control lot 61918.d External Control lot 61918

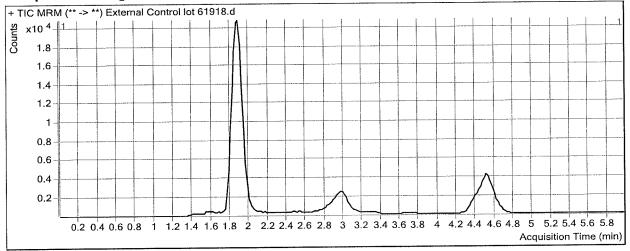
Dilution **Position** 1 P1-B2 **Acq Method**

AM 27 Quant THC 7-2017.m

Sample Info

Inj Vol -1 Comment

AM 27 Cannabinoid Confirmation



Results				TCTD Does	Resp Ratio	Final Conc
Compound	ISTD Compound	RT	Response	ISTD Resp		
THC-OH	THC-OH-d3	1.895	8061	109805	0.0734	7.0242
THC-COOH	THC-COOH-d9	1.945	6182	35787	0.1727	8.4059
THC	THC-d3	4.531	3866	46302	0.0835	6.1635



Batch Data Path D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Analysis Time 3/23/2018 9:09 AM Analyst Name ISP Tox
Report Time 3/23/2018 9:11 AM Reporter Name ISP Tox
Last Calib Update 3/23/2018 9:09 AM Batch State Processed

Analysis Info

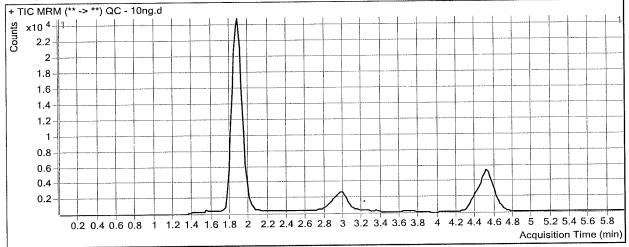
 Acq Time
 2018-03-22 15:24
 Data File
 QC - 10ng.d

 Sample Type
 QC
 Sample Name
 QC - 10ng

Dilution 1 **Acq Method** AM 27 Quant THC 7-2017.m

Position P1-H1 Sample Info

Inj Vol -1 Comment AM 27 Cannabinoid Confirmation



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	1.895	13195	124423	0.1061	10.1982
THC-COOH	THC-COOH-d9	1.945	8659	39939	0.2168	10.5216
,			7729	54478	0.1419	10.6629
THC	THC-d3	4.531	1123	37770	0.1 113	1010023

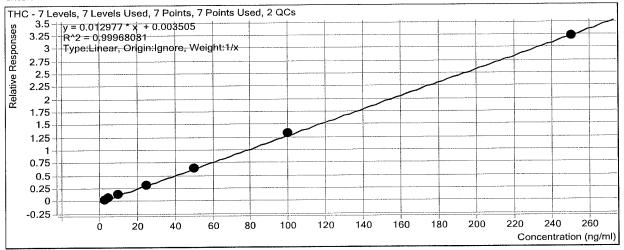


ISP Forensics Calibration Curve Report

Batch Data Path D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Last Calib Update 3/23/2018 9:09 AM Analyst Name ISP TOX

Target CompoundTHCInternal StandardTHC-d3



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1 - 3ng	1	\square	3	2.9	96.0
Cal 2 - 5ng	2	\square	5	5.2	103.2
Cal 3 - 10ng	3	☑	10	10.1	100.9
QC - 10ng	3	☑	10	10.7	106.6
External Control lot 61918	3	☑	10	6.2	61.6
Cal 4 - 25ng	4	\square	25	24.8	99.2
Cal 5 - 50ng	5	☑	50	49.4	98.8
Cal 6 - 100ng	6		100	102.8	102.8
Cal 7 - 250ng	7	\square	250	247.8	99.1



ISP Forensics Calibration Curve Report

Batch Data Path

D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Last Calib Update

3/23/2018 9:09 AM

Analyst Name

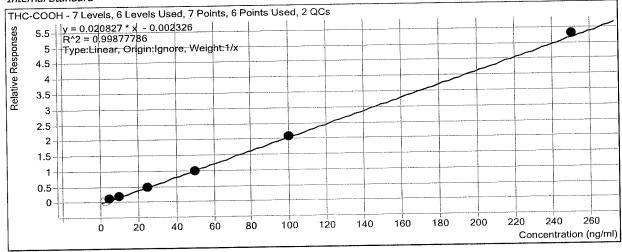
ISP TOX

Target Compound

THC-COOH

Internal Standard

THC-COOH-d9



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1 - 3ng	1		3	3.4	112.5
Cal 2 - 5ng	2	☑	5	5.7	114.8
Cal 3 - 10ng	3	☑	10	9.4	93.8
OC - 10ng	3	☑	10	10.5	105.2
External Control lot 61918	3	☑	10	8.4	84.1
Cal 4 - 25ng	4	☑	25	24.0	95.8
Cal 5 - 50ng	5	◩	50	47.3	94.5
Cal 6 - 100ng	6	☑	100	99.3	99.3
Cal 7 - 250ng	7	☑	250	254.3	101.7

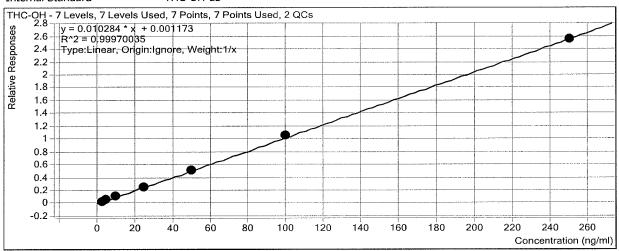


ISP Forensics Calibration Curve Report

Batch Data Path D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Last Calib Update 3/23/2018 9:09 AM Analyst Name ISP TOX

Target CompoundTHC-OHInternal StandardTHC-OH-d3



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1 - 3ng	1	\square	3	2.9	97.6
Cal 2 - 5ng	2	\square	5	5.2	104.1
Cal 3 - 10ng	3	\square	10	9.9	98.9
QC - 10ng	3	\square	10	10.2	102.0
External Control lot 61918	3	\square	10	7.0	70.2
Cal 4 - 25ng	4	\square	25	24.6	98.6
Cal 5 - 50ng	5	\square	50	49.4	98.8
Cal 6 - 100ng	6	☑	100	102.7	102.7
Cal 7 - 250ng	7	Ø	250	248.2	99.3



Batch Data Path D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

 Analysis Time
 3/23/2018 9:09 AM
 Analyst Name
 ISP Tox

 Report Time
 3/23/2018 9:11 AM
 Reporter Name
 ISP Tox

 Last Calib Update
 3/23/2018 9:09 AM
 Batch State
 Processed

Analysis Info

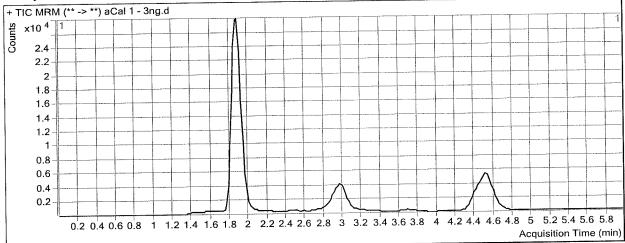
Acq Time2018-03-22 13:38Data FileaCal 1 - 3ng.dSample TypeCalibrationSample NameCal 1 - 3ng

Dilution 1 Acq Method AM 27 Quant THC 7-2017.m

Position P1-A1 Sample Info

Inj Vol -1 Comment AM 27 Cannabinoid Confirmation

Sample Chromatogram



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
- " •	THC-OH-d3	1.875	4605	147187	0.0313	2.9279
THC-OH			3251	47824	0.0680	3.3756
THC-COOH	THC-COOH-d9	1.945				
THC	THC-d3	4.531	2624	64190	0.0409	2.8794



Printed at: 9:13 AM on: 3/23/2018

Batch Data Path D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Analysis Time 3/23/2018 9:09 AM Analyst Name ISP Tox
Report Time 3/23/2018 9:11 AM Reporter Name ISP Tox
Last Calib Update 3/23/2018 9:09 AM Batch State Processed

Analysis Info

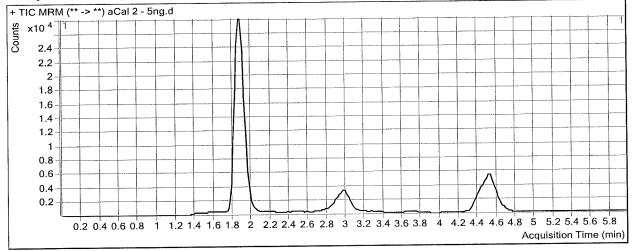
Acq Time2018-03-22 13:49Data FileaCal 2 - 5ng.dSample TypeCalibrationSample NameCal 2 - 5ng

Dilution 1 Acq Method AM 27 Quant THC 7-2017.m

Position P1-B1 Sample Info

Inj Vol -1 Comment AM 27 Cannabinoid Confirmation

Sample Chromatogram



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	1.875	7920	144788	0.0547	5.2051
THC-COOH	THC-COOH-d9	1.945	5486	46813	0.1172	5.7386
THC	THC-d3	4.511	4358	61863	0.0704	5.1579
1110	1110 00					



Printed at: 9:13 AM on: 3/23/2018

Batch Data Path D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Analysis Time 3/23/2018 9:09 AM Analyst Name ISP Tox Report Time 3/23/2018 9:11 AM Reporter Name ISP Tox Last Calib Update 3/23/2018 9:09 AM Batch State Processed

Analysis Info

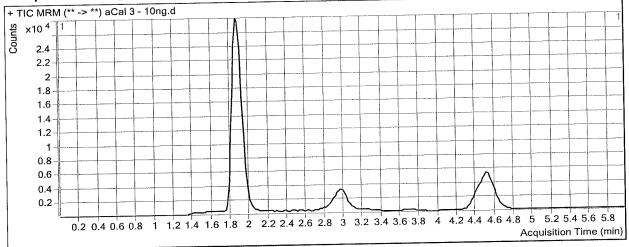
 Acq Time
 2018-03-22 14:01
 Data File
 aCal 3 - 10ng.d

 Sample Type
 Calibration
 Sample Name
 Cal 3 - 10ng

Dilution 1 Acq Method AM 27 Quant THC 7-2017.m

Position P1-C1 Sample Info

Inj Vol -1 Comment AM 27 Cannabinoid Confirmation



Results					n n-41-	Final Conc
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	9,8902
THC-OH	THC-OH-d3	1.875	14072	136770	0.1029	
THC-COOH	THC-COOH-d9	1.945	8545	44259	0.1931	9.3819
THC	THC-d3	4.531	7686	57167	0.1344	10.0897



Batch Data Path D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Analysis Time 3/23/2018 9:09 AM Analyst Name ISP Tox Report Time 3/23/2018 9:11 AM Reporter Name ISP Tox Last Calib Update 3/23/2018 9:09 AM Batch State Processed

Analysis Info

 Acq Time
 2018-03-22 14:13
 Data File
 aCal 4 - 25ng.d

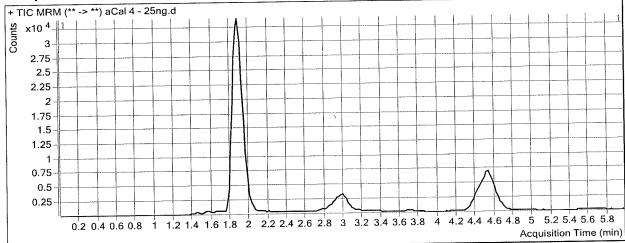
 Sample Type
 Calibration
 Sample Name
 Cal 4 - 25ng

Dilution 1 Acq Method AM 27 Quant THC 7-2017.m

Position P1-D1 Sample Info

Inj Vol -1 Comment AM 27 Cannabinoid Confirmation

Sample Chromatogram



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
•	THC-OH-d3	1.875	36156	141998	0.2546	24.6445
THC-OH	***=	1.945	22676	45661	0.4966	23,9564
THC-COOH	THC-COOH-d9	_,			0.3254	24.8018
THC	THC-d3	4.531	19526	60015	0.3234	24.0010



Printed at: 9:13 AM on: 3/23/2018

Samples Report Cannabinoids.xlsx

Batch Data Path

D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Analysis Time

3/23/2018 9:09 AM

Analyst Name ISP Tox

Report Time Last Calib Update 3/23/2018 9:11 AM 3/23/2018 9:09 AM Reporter Name ISP Tox
Batch State Processed

Analysis Info

Acq Time Sample Type Dilution 2018-03-22 14:25

Data File Sample Name aCal 5 - 50ng.d

Calibration 1 Sample Name Cal 5 - 50ng
Acq Method AM 27 Quant THC 7-2017.m

Sample Info

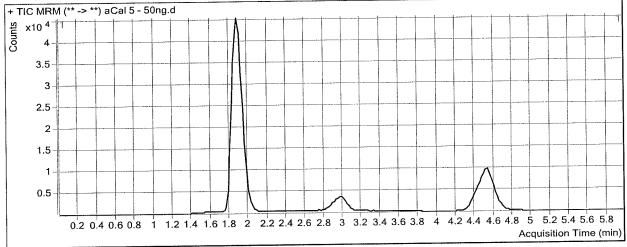
Position P1-E1

Comment

Inj Vol -1

AM 27 Cannabinoid Confirmation

Sample Chromatogram



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	1.895	78768	154682	0.5092	49.4021
	THC-COOH-d9	1.945	47462	48331	0.9820	47.2617
THC-COOH	,,, , =======			64643	0.6444	49,3892
THC	THC-d3	4.531	41658	04043	דדדטיט	13,3032



Printed at: 9:13 AM on: 3/23/2018

Batch Data Path

D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

Analysis Time Report Time

3/23/2018 9:09 AM 3/23/2018 9:11 AM **Analyst Name** ISP Tox

Reporter Name ISP Tox

Last Calib Update

3/23/2018 9:09 AM

Batch State

Processed

Analysis Info

Acq Time

2018-03-22 14:37

Data File

aCal 6 - 100ng.d

Sample Type

Calibration

Sample Name

Cal 6 - 100ng

Dilution

1

Acq Method

AM 27 Quant THC 7-2017.m

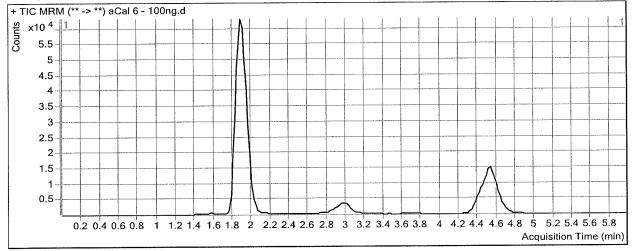
Position Inj Vol

P1-F1 -1

Sample Info

Comment

AM 27 Cannabinoid Confirmation



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	1.875	162308	153438	1.0578	102.7441
THC-COOH	THC-COOH-d9	1.945	98473	47655	2.0664	99.3252
THC	THC-d3	4.531	87032	65040	1.3381	102.8450



Batch Data Path D:\2018 Data\03222018 cann quant\QuantResults\32218 cann quant.batch.bin

 Analysis Time
 3/23/2018 9:09 AM
 Analyst Name
 ISP Tox

 Report Time
 3/23/2018 9:11 AM
 Reporter Name
 ISP Tox

 Last Calib Update
 3/23/2018 9:09 AM
 Batch State
 Processed

Analysis Info

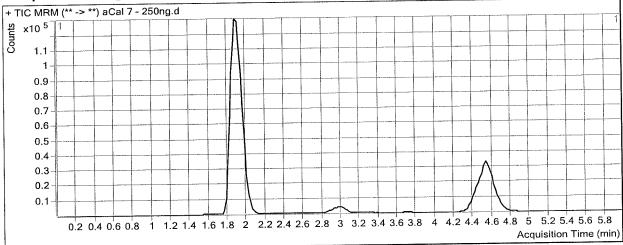
 Acq Time
 2018-03-22 14:49
 Data File
 aCal 7 - 250ng.d

 Sample Type
 Calibration
 Sample Name
 Cal 7 - 250ng

Dilution 1 Acq Method AM 27 Quant THC 7-2017.m

Position P1-G1 Sample Info

Inj Vol -1 Comment AM 27 Cannabinoid Confirmation



Results						
	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
Compound	THC-OH-d3	1.895	461014	180539	2.5535	248.1862
THC-OH			265574	50157	5.2948	254.3362
THC-COOH	THC-COOH-d9	1.945				247.8370
THC	THC-d3	4.531	244678	75994	3.2197	247.0370