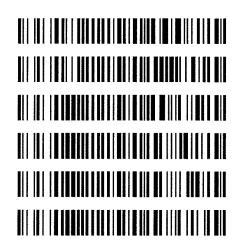


Worklist: 2733

<u>LAB CASE</u> C2018-1903	<u>ITEM</u> 1	<u>TASK ID</u> 129059	DESCRIPTION AM 27 Blood THC Quant by LC-QQQ
C2018-1997	1	129058	AM 27 Blood THC Quant by LC-QQQ
P2018-2727	1	129057	AM 27 Blood THC Quant by LC-QQQ
P2018-2844	1	129056	AM 27 Blood THC Quant by LC-QQQ
P2018-2845	1	129055	AM 27 Blood THC Quant by LC-QQQ
P2018-2847	1	129054	AM 27 Blood THC Quant by LC-QQQ





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

Extraction Date: 10/10/18 Analyst: Anne Nord

Plate lot#: 0539904 Plate Expiration: 09/10/2019

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: 18G207D7 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.

Analytic:

- ☑ 1. Remove standards, plate, controls, and samples from cold storage. Allow to reach room temperature.
- Σ 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
- Δ 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.
- 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.
- □ 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)
- \boxtimes 13. Wait 5 minutes.
- △ 14. Apply positive pressure for approx. 15 seconds. (10-15 PSI- Selector to the left).
- 🗵 16. Reconstitute in 100μL 100% MeOH and heat seal plate with foil. Place in autosampler and run worklist.

Post-Analytic

- \boxtimes 1. Create batch and process data.
 - Batch path: 1010118 cann quant Batch Name: cann quant
- \boxtimes 2. Make any necessary integration changes, Curve weighting of Linear 1/x with r² values \ge 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
- ⊠ 6 Enter QCs into control charting.
- ☑ 7 Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS:



Toxicology AM method 27 external prep information

working solution 1 $\rm ug/ml$ in meoh C-THC, THC-OH, THC Stock solution 1 $\rm ug/ml$ 10 $\rm ul$ each THC, THC-OH 100 $\rm ug/ml$ 100 $\rm ul$ C-THC in 9890 $\rm ul$ meOH

Ppd 6/5/18 Exp: 4/1/19 lot 6518

by AMN

THC	THC-OH	C-THC	Drug
FE04231406	FE01141502	FE03121501	lot (cerilliant)
4/1/2019	1/1/2020	3/1/2020	expiration

AM 27 control 50 ul working solution lot (6518) in 4950 ul blood lot (17J20718) ppd 6/5/18 Exp 4/1/19 lot 6518 Concentration 10 ng/ml $^{\circ}$

lot 6518 Concentration 10 ng/ml each by AMN lot 92018 Concentration 10 ng/ml each by AMN

ppd 9/20/18 Exp 4/1/19

Batch Data Path D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin

 Analysis Time
 10/12/2018 11:12 AM
 Analyst Name
 ISP Tox

 Report Time
 10/12/2018 11:15 AM
 Reporter Name
 ISP Tox

 Last Calib Update
 10/12/2018 11:12 AM
 Batch State
 Processed

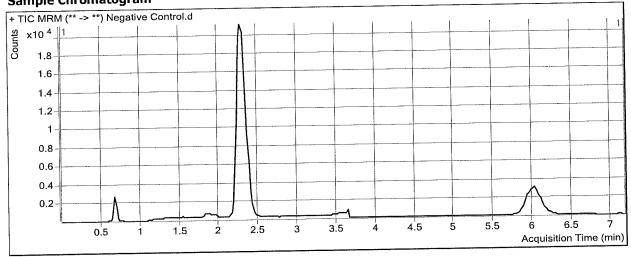
Analysis Info

Acq Time 2018-10-11 18:45 Data File Negative Control.d Sample Type Sample Sample Negative Control

Dilution 2 Acq Method AM 27 Quant THC 7-2017.m

Position P1-A2 Sample Info
Inj Vol -1 Comment AM 27 Cannabinoid Confirmation

Sample Chromatogram



Results Compound THC-OH	ISTD Compound THC-OH-d3	RT 1.855	Response 1614	ISTD Resp 120922	Resp Ratio 0.0134	Final Conc 1.6715 4 4 4
-------------------------------	-------------------------	--------------------	------------------	---------------------	----------------------	----------------------------

D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin **Batch Data Path**

Analyst Name ISP Tox 10/12/2018 11:12 AM **Analysis Time** Reporter Name ISP Tox 10/12/2018 11:15 AM **Report Time** Processed **Batch State** 10/12/2018 11:12 AM **Last Calib Update**

Analysis Info

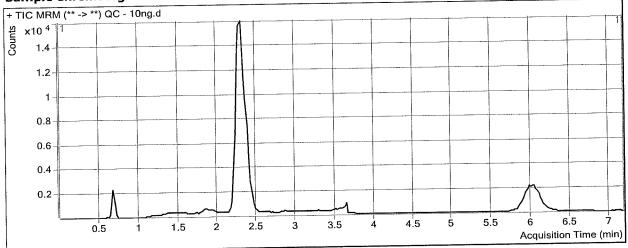
QC-10ng.d 10 Ng CTHC, STHC, STHCOH A **Data File** 2018-10-11 18:57 **Acq Time** QC - 10ng Sample Name Sample Type QC

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

Sample Info P1-H1 **Position**

AM 27 Cannabinoid Confirmation Comment -1 Inj Vol

Sample Chromatogram



D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin **Batch Data Path**

Analyst Name ISP Tox **Analysis Time** 10/12/2018 11:12 AM Reporter Name ISP Tox 10/12/2018 11:15 AM Report Time **Batch State** Processed 10/12/2018 11:12 AM **Last Calib Update**

Analysis Info

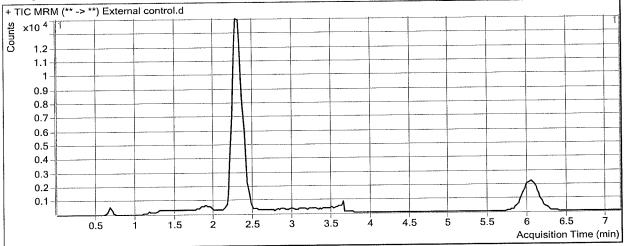
Data File External control.d 2018-10-11 19:09 **Acq Time** Sample Name External control Sample Sample Type **Acq Method** AM 27 Quant THC 7-2017.m

Dilution 1

Sample Info P1-B2 **Position**

Comment AM 27 Cannabinoid Confirmation Inj Vol -1

Sample Chromatogram



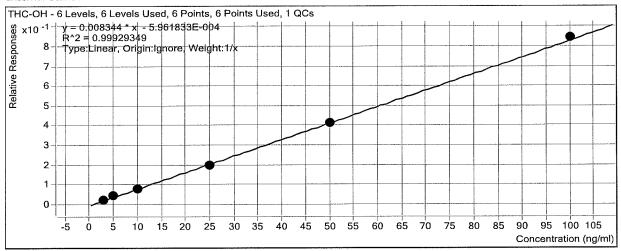
Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	2.296	5849	77649	0.0753	9.0992
THC-COOH	THC-COOH-d9	2.386	4576	34306	0.1334	8.0435
THC	THC-d3	6.073	3072	27737	0.1108	11.5509
TITC	1110 05					

ISP Forensics Calibration Curve Report

Batch Data Path D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin

Last Calib Update 10/12/2018 11:12 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	98.0
Cal 2 - 5ng	2	\square	5	5.4	108.1
Cal 3 - 10ng	3	\square	10	9.7	97.3
Cal 4 - 25ng	4	\square	25	23.9	95.7
Cal 5 - 50ng	5	☑	50	49.9	99.8
Cal 6 - 100ng	6	☑	100	101.1	101.1
QC - 10ng	7	☑	5	4.9	98.9



ISP Forensics Calibration Curve Report

Batch Data Path

D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin

Last Calib Update

10/12/2018 11:12 AM

Analyst Name

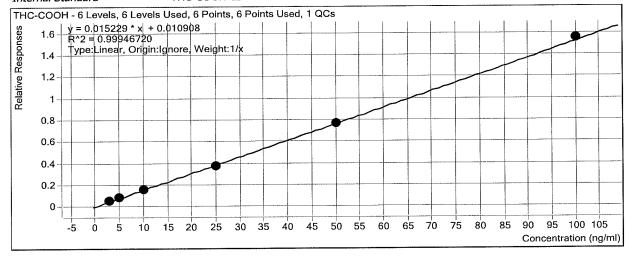
ISP TOX

Target Compound

THC-COOH

Internal Standard

THC-COOH-d9



Sample	Levei	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1 - 3ng	1	\square	3	3.2	105.8
Cal 2 - 5ng	2	\square	5	4.9	97.8
Cal 3 - 10ng	3	\square	10	10.0	99.9
Cal 4 - 25ng	4	☑	25	23.9	95.7
Cal 5 - 50ng	5	\square	50	49.8	99.6
Cal 6 - 100ng	6	☑	100	101.2	101.2
QC - 10ng	7	☑	10	9.7	97.3



ISP Forensics Calibration Curve Report

Batch Data Path

D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin

Last Calib Update

10/12/2018 11:12 AM

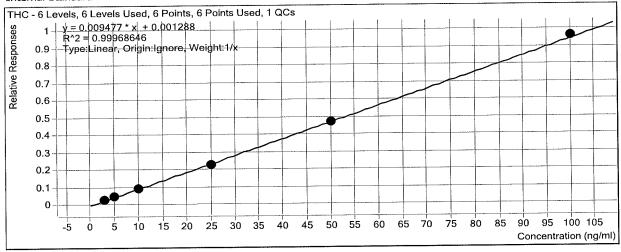
Analyst Name

ISP TOX

Target Compound Internal Standard

THC

THC-d3



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1 - 3ng	1	\square	3	3.1	103.6
Cal 2 - 5ng	2	\square	5	5.0	99.6
Cal 3 - 10ng	3	\square	10	10.0	99.5
Cal 4 - 25ng	4	\square	25	24.2	96.7
Cal 5 - 50ng	5	☑	50	49.7	99.5
Cal 6 - 100ng	6	☑	100	101.1	101.1
QC - 10ng	7	☑	5	5.0	100.9



Batch Data Path

D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin

Analysis Time Report Time

10/12/2018 11:12 AM 10/12/2018 11:15 AM

Analyst Name ISP Tox Reporter Name ISP Tox

Last Calib Update

10/12/2018 11:12 AM

Batch State Processed

Analysis Info

Acq Time

2018-10-11 17:22

Data File

Cal 1 - 3ng.d

Sample Type

Calibration

Sample Name

Cal 1 - 3ng

Dilution

1

Acq Method

AM 27 Quant THC 7-2017.m

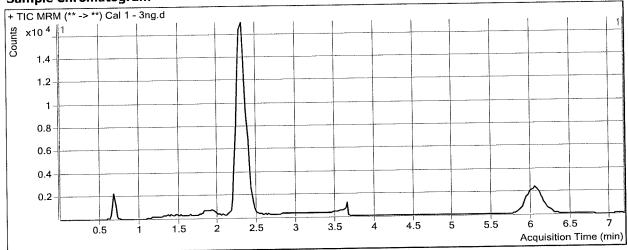
Position Inj Vol

P1-B1 -1

Sample Info Comment

AM 27 Cannabinoid Confirmation

Sample Chromatogram



Results						=:1.0
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	2.316	2354	98295	0.0239	2.9414
THC-COOH	THC-COOH-d9	2.386	2494	42108	0.0592	3.1732
THC	THC-d3	6.073	1023	33277	0.0308	3.1090



Batch Data Path D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin

 Analysis Time
 10/12/2018 11:12 AM
 Analyst Name
 ISP Tox

 Report Time
 10/12/2018 11:15 AM
 Reporter Name
 ISP Tox

 Last Calib Update
 10/12/2018 11:12 AM
 Batch State
 Processed

Analysis Info

 Acq Time
 2018-10-11 17:34
 Data File
 Cal 2 - 5ng.d

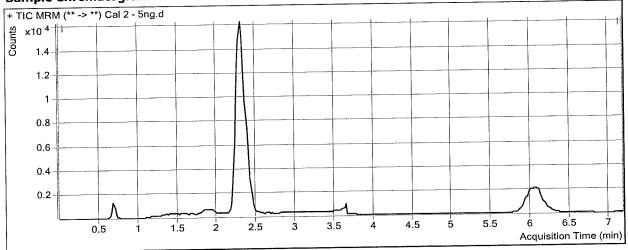
 Sample Type
 Calibration
 Sample Name
 Cal 2 - 5ng

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

Position P1-C1 Sample Info

Inj Vol -1 Comment AM 27 Cannabinoid Confirmation

Sample Chromatogram



Results Compound THC-OH	ISTD Compound THC-OH-d3 THC-COOH-d9	RT 2.296 2.406	Response 4061 3343	ISTD Resp 91285 39141	Resp Ratio 0.0445 0.0854	Final Conc 5.4026 4.8920
THC-COOH THC	THC-COOH-d9 THC-d3	2.406 6.113	3343 1441	39141 29706	0.0854	4.9825

Batch Data Path D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin

 Analysis Time
 10/12/2018 11:12 AM
 Analyst Name
 ISP Tox

 Report Time
 10/12/2018 11:15 AM
 Reporter Name
 ISP Tox

 Last Calib Update
 10/12/2018 11:12 AM
 Batch State
 Processed

Analysis Info

 Acq Time
 2018-10-11 17:46
 Data File
 Cal 3 - 10ng.d

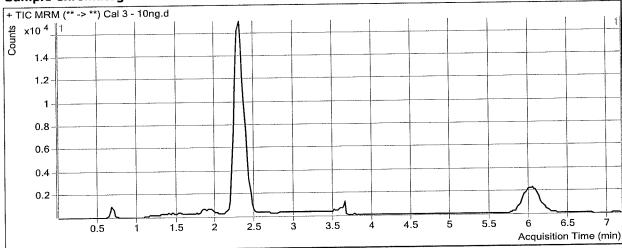
 Sample Type
 Calibration
 Sample Name
 Cal 3 - 10ng.d

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						
	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
Compound	THC-OH-d3	2.316	7469	92632	0.0806	9.7341
THC-OH	,,,		6447	39538	0.1631	9.9906
THC-COOH	THC-COOH-d9	2.406				9.9523
THC	THC-d3	6.073	2883	30154	0.0956	9.9323

Batch Data Path D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin

 Analysis Time
 10/12/2018 11:12 AM
 Analyst Name
 ISP Tox

 Report Time
 10/12/2018 11:15 AM
 Reporter Name
 ISP Tox

 Last Calib Update
 10/12/2018 11:12 AM
 Batch State
 Processed

Analysis Info

 Acq Time
 2018-10-11 17:58
 Data File
 Cal 4 - 25ng.d

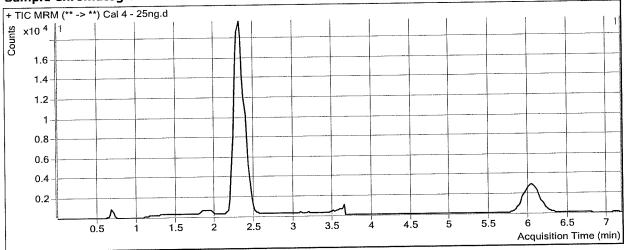
 Sample Type
 Calibration
 Sample Name
 Cal 4 - 25ng

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

Position P1-E1 Sample Info

Inj Vol -1 Comment AM 27 Cannabinoid Confirmation

Sample Chromatogram



Results		DT	Response	ISTD Resp	Resp Ratio	Final Conc
Compound THC-OH	ISTD Compound THC-OH-d3	RT 2.316	19103	96005	0.1990	23.9174
THC-COOH	THC-COOH-d9	2,406	15395	41039	0.3751	23.9167
THC	THC-d3	6.093	7456	32373	0.2303	24.1664

Batch Data Path D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin

 Analysis Time
 10/12/2018 11:12 AM
 Analyst Name
 ISP Tox

 Report Time
 10/12/2018 11:15 AM
 Reporter Name
 ISP Tox

 Last Calib Update
 10/12/2018 11:12 AM
 Batch State
 Processed

Analysis Info

 Acq Time
 2018-10-11 18:10
 Data File
 Cal 5 - 50ng.d

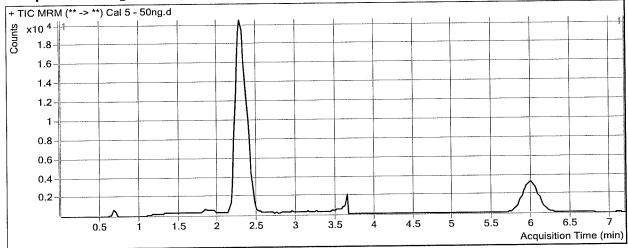
 Sample Type
 Calibration
 Sample Name
 Cal 5 - 50ng

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

Position P1-F1 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	2,296	34644	83345	0.4157	49.8866
THC-COOH	THC-COOH-d9	2,386	27739	36068	0.7691	49.7864
I HC-COOH				28282	0.4726	49,7380
THC	THC-d3	5.993	13367	20202	0.1720	1517500

Batch Data Path D:\2018 Data\1010118 cann quant\QuantResults\cann quant.batch.bin

 Analysis Time
 10/12/2018 11:12 AM
 Analyst Name
 ISP Tox

 Report Time
 10/12/2018 11:15 AM
 Reporter Name
 ISP Tox

 Last Calib Update
 10/12/2018 11:12 AM
 Batch State
 Processed

Analysis Info

 Acq Time
 2018-10-11 18:22
 Data File
 Cal 6 - 100ng.d

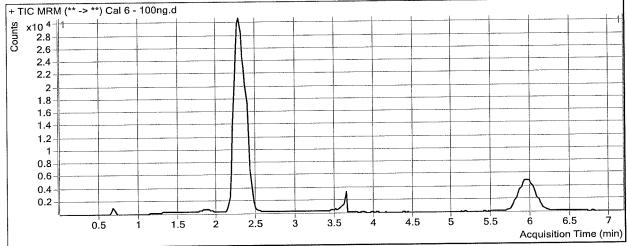
 Sample Type
 Calibration
 Sample Name
 Cal 6 - 100ng

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

Position P1-G1 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	2.276	81308	96431	0.8432	101.1179
THC-COOH	THC-COOH-d9	2,366	62214	40069	1.5527	101.2411
THC	THC-d3	5.953	30610	31921	0.9589	101.0518

7