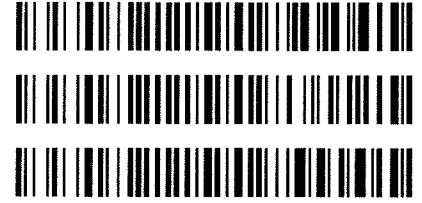


**Worklist: 2355**

<u>LAB CASE</u>	<u>ITEM</u>	<u>TASK ID</u>	<u>DESCRIPTION</u>
C2018-0577	1	114242	AM 27 Blood THC Quant by LC-QQQ
C2018-0716	1	114243	AM 27 Blood THC Quant by LC-QQQ
C2018-0766	2	114244	AM 27 Blood THC Quant by LC-QQQ



A handwritten signature or scribble in black ink, consisting of several overlapping loops and lines.

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

Extraction Date: 4-25-18

Analyst: Ann Nord

Plate lot#: 0515037

Plate Expiration: 9/28/2018

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

**Mobile phase B:** 0.1% Formic acid in Acetonitrile  
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.
- 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.
- 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)**
- 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: 66819*
- 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: 04252018 cann quant Batch Name: 042518 cann quant
- 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:

Toxicology AM method 27 external prep information

working solution 1 ug/ml in meoh C-THC, THC-OH, THC

Stock solution 1mg/ml 10 ul THC, 100 ug/ml 100 ul each THC-OH, C-THC in 9880 ul meOH

Ppd 3/19/18 Exp: 9/19/18 lot 91918 by AMN

Drug	lot (cerilliant)	expiration
C-THC	FE07171501	9/1/2020
THC-OH	FE07221601	7/1/2021
THC	FE01041701	3/1/2022

AM 27 control 100 ul working solution lot (91918) in 9900 ul blood lot (17J20718)

ppd 3/19/18 Exp 6/19/18 lot 61918 Concentration 10 ng/ml each by AMN

# ISP FORENSICS - Cd'A Instrument # 62340

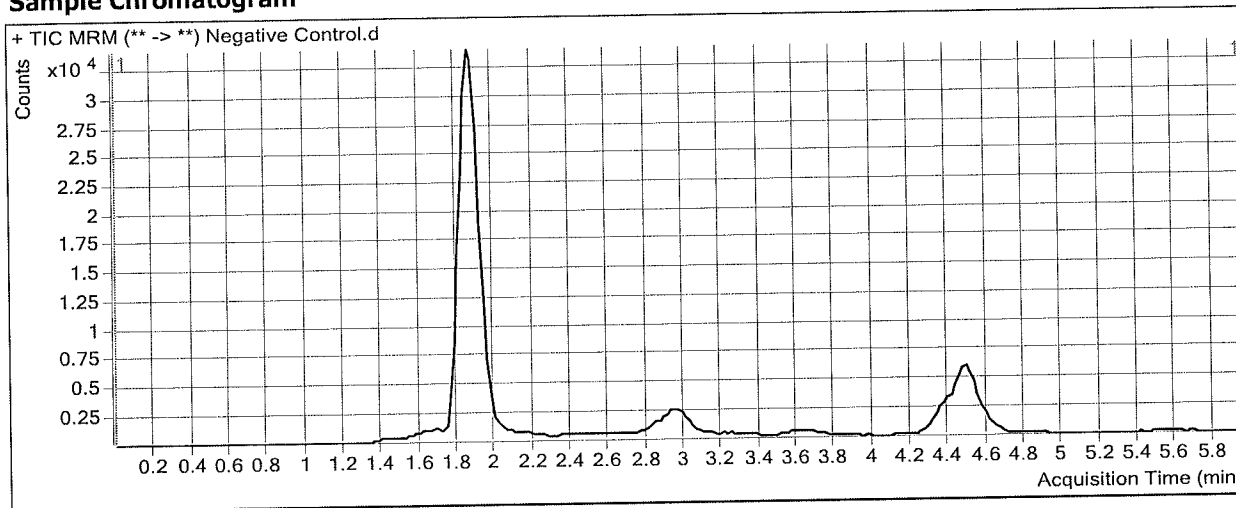
## Cannabinoids Analysis Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin  
**Analysis Time** 4/26/2018 7:24 AM **Analyst Name** ISP Tox  
**Report Time** 4/26/2018 7:32 AM **Reporter Name** ISP Tox  
**Last Calib Update** 4/26/2018 7:24 AM **Batch State** Processed

### Analysis Info

**Acq Time** 2018-04-25 17:18 **Data File** Negative Control.d  
**Sample Type** Sample **Sample Name** Negative Control  
**Dilution** 1 **Acq Method** AM 27 Quant THC 7-2017.m  
**Position** P1-C2 **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	2.155	2437	192567	0.0127	1.4604
THC-COOH	THC-COOH-d9	1.945	3783	64545	0.0586	0.9672

# ISP FORENSICS - Cd'A Instrument # 62340

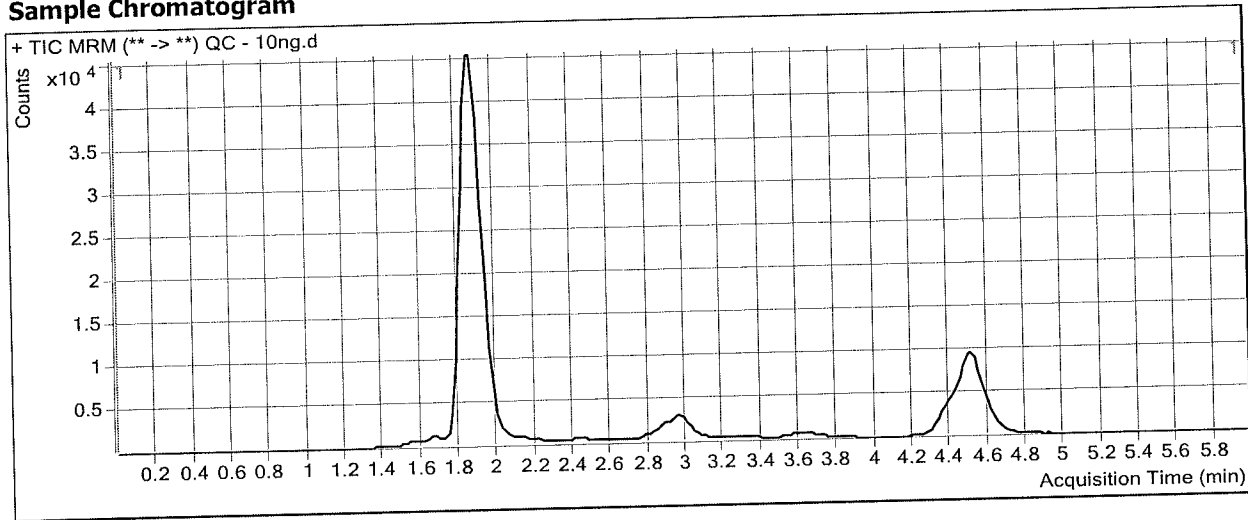
## Cannabinoids Analysis Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin  
**Analysis Time** 4/26/2018 7:24 AM **Analyst Name** ISP Tox  
**Report Time** 4/26/2018 7:32 AM **Reporter Name** ISP Tox  
**Last Calib Update** 4/26/2018 7:24 AM **Batch State** Processed

### Analysis Info

**Acq Time** 2018-04-25 17:30 **Data File** QC - 10ng.d  
**Sample Type** Sample **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

### Sample Chromatogram



### Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	1.895	22189	232233	0.0955	9.6302
THC-COOH	THC-COOH-d9	1.945	17664	76910	0.2297	9.7862
THC	THC-d3	4.511	12129	94159	0.1288	10.1103

# ISP FORENSICS - Cd'A Instrument # 62340

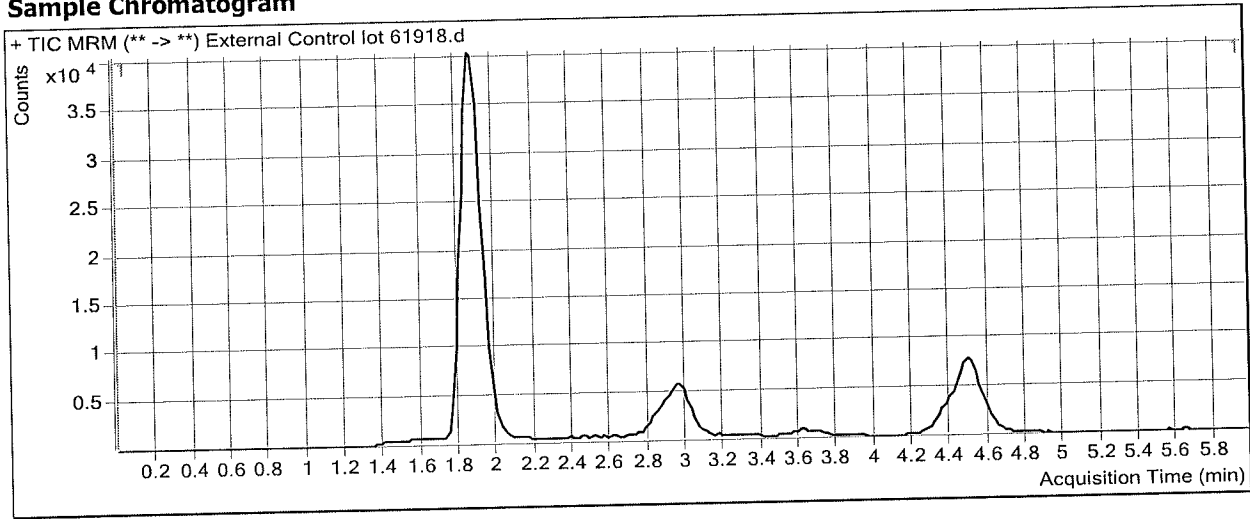
## Cannabinoids Analysis Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin  
**Analysis Time** 4/26/2018 7:24 AM **Analyst Name** ISP Tox  
**Report Time** 4/26/2018 7:32 AM **Reporter Name** ISP Tox  
**Last Calib Update** 4/26/2018 7:24 AM **Batch State** Processed

**Analysis Info**

**Acq Time** 2018-04-25 17:42 **Data File** External Control lot 61918.d  
**Sample Type** Sample **Sample Name** External Control lot 61918  
**Dilution** 1 **Acq Method** AM 27 Quant THC 7-2017.m  
**Position** P1-E2 **Sample Info**  
**Inj Vol** -1 **Comment** AM 27 Cannabinoid Confirmation

**Sample Chromatogram**



**Results**

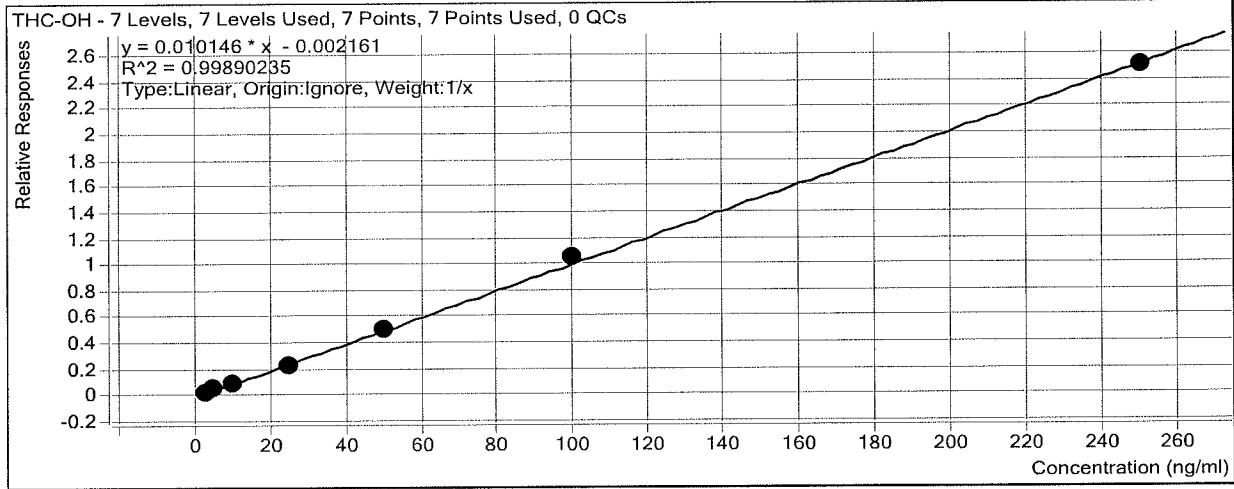
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	1.875	16351	213104	0.0767	7.7754
THC-COOH	THC-COOH-d9	1.945	12632	69153	0.1827	7.3629
THC	THC-d3	4.491	7299	79503	0.0918	7.2269

# ISP Forensics Calibration Curve Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin

**Last Calib Update** 4/26/2018 7:24 AM **Analyst Name** ISP TOX

**Target Compound** *THC-OH*  
**Internal Standard** *THC-OH-d3*



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1 - 3ng	1	<input checked="" type="checkbox"/>	3	3.1	103.8
Cal 2 - 5ng	2	<input checked="" type="checkbox"/>	5	5.1	102.7
Cal 3 - 10ng	3	<input checked="" type="checkbox"/>	10	9.8	98.0
Cal 4 - 25ng	4	<input checked="" type="checkbox"/>	25	23.2	92.8
Cal 5 - 50ng	5	<input checked="" type="checkbox"/>	50	49.3	98.7
Cal 6 - 100ng	6	<input checked="" type="checkbox"/>	100	104.9	104.9
Cal 7 - 250ng	7	<input checked="" type="checkbox"/>	250	247.5	99.0

# ISP Forensics Calibration Curve Report

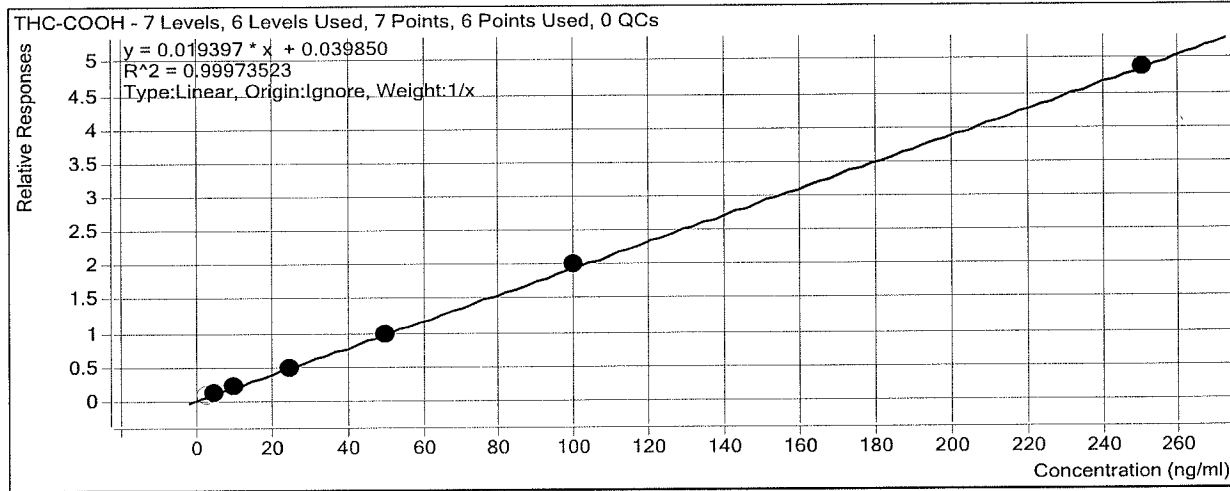
**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin

**Last Calib Update** 4/26/2018 7:24 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	<input type="checkbox"/>	3	3.6	120.7
Cal 2 - 5ng	2	<input checked="" type="checkbox"/>	5	5.1	101.3
Cal 3 - 10ng	3	<input checked="" type="checkbox"/>	10	10.2	101.5
Cal 4 - 25ng	4	<input checked="" type="checkbox"/>	25	24.0	96.2
Cal 5 - 50ng	5	<input checked="" type="checkbox"/>	50	49.6	99.3
Cal 6 - 100ng	6	<input checked="" type="checkbox"/>	100	102.1	102.1
Cal 7 - 250ng	7	<input checked="" type="checkbox"/>	250	249.0	99.6



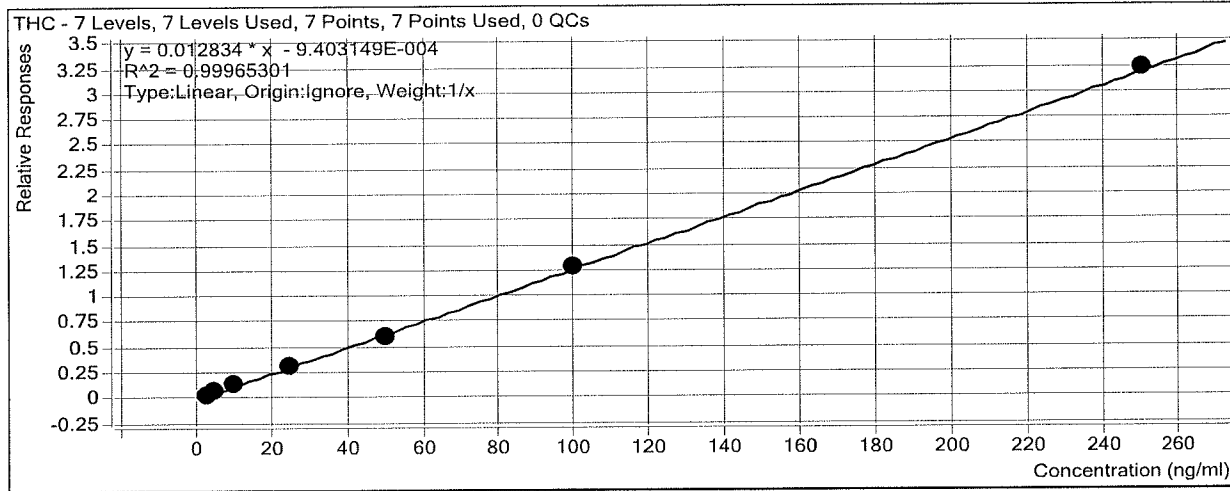
# ISP Forensics Calibration Curve Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin

**Last Calib Update** 4/26/2018 7:24 AM

**Analyst Name** ISP TOX

**Target Compound** *THC*  
**Internal Standard** *THC-d3*



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1 - 3ng	1	<input checked="" type="checkbox"/>	3	3.1	102.8
Cal 2 - 5ng	2	<input checked="" type="checkbox"/>	5	5.0	100.2
Cal 3 - 10ng	3	<input checked="" type="checkbox"/>	10	10.3	102.9
Cal 4 - 25ng	4	<input checked="" type="checkbox"/>	25	24.2	96.6
Cal 5 - 50ng	5	<input checked="" type="checkbox"/>	50	48.1	96.1
Cal 6 - 100ng	6	<input checked="" type="checkbox"/>	100	100.6	100.6
Cal 7 - 250ng	7	<input checked="" type="checkbox"/>	250	251.8	100.7

# ISP FORENSICS - Cd'A Instrument # 62340

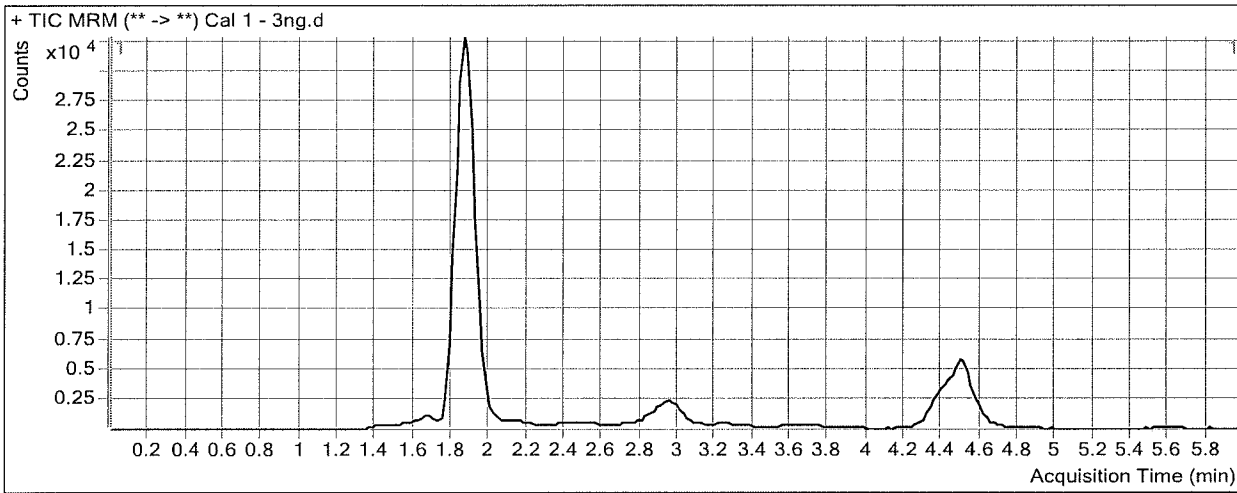
## Cannabinoids Analysis Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin  
**Analysis Time** 4/26/2018 7:24 AM **Analyst Name** ISP Tox  
**Report Time** 4/26/2018 7:32 AM **Reporter Name** ISP Tox  
**Last Calib Update** 4/26/2018 7:24 AM **Batch State** Processed

### Analysis Info

**Acq Time** 2018-04-25 15:43 **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** 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	THC-OH-d3	1.875	5024	170689	0.0294	3.1143
THC-COOH	THC-COOH-d9	1.925	6405	58166	0.1101	3.6223
THC	THC-d3	4.511	2480	64194	0.0386	3.0837

# ISP FORENSICS - Cd'A Instrument # 62340

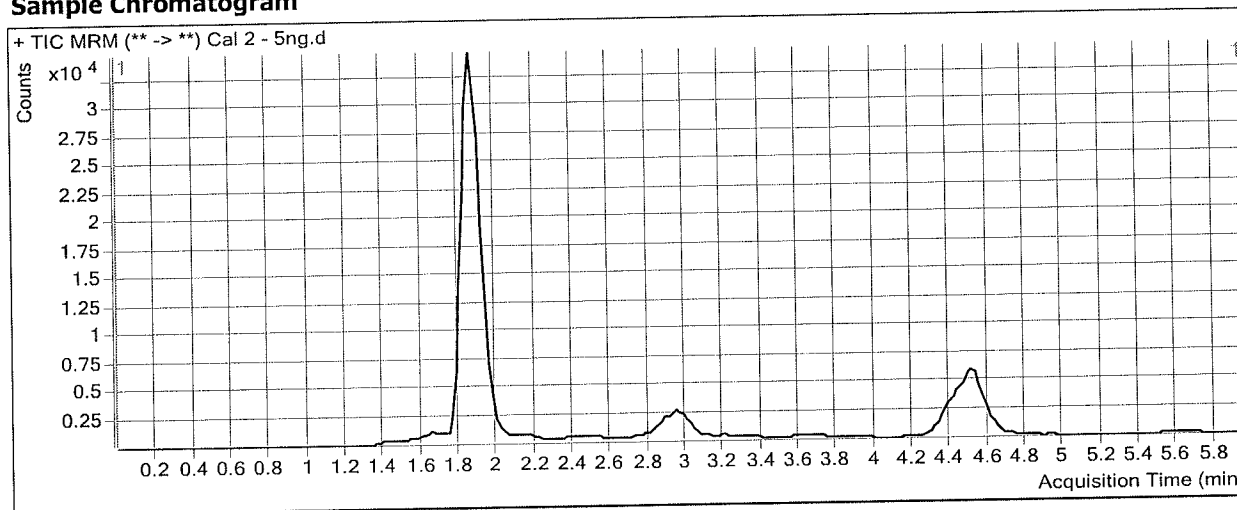
## Cannabinoids Analysis Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin  
**Analysis Time** 4/26/2018 7:24 AM **Analyst Name** ISP Tox  
**Report Time** 4/26/2018 7:32 AM **Reporter Name** ISP Tox  
**Last Calib Update** 4/26/2018 7:24 AM **Batch State** Processed

### Analysis Info

**Acq Time** 2018-04-25 15:55 **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-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	8659	173317	0.0500	5.1374
THC-COOH	THC-COOH-d9	1.945	8058	58347	0.1381	5.0656
THC	THC-d3	4.531	4180	65959	0.0634	5.0108

# ISP FORENSICS - Cd'A Instrument # 62340

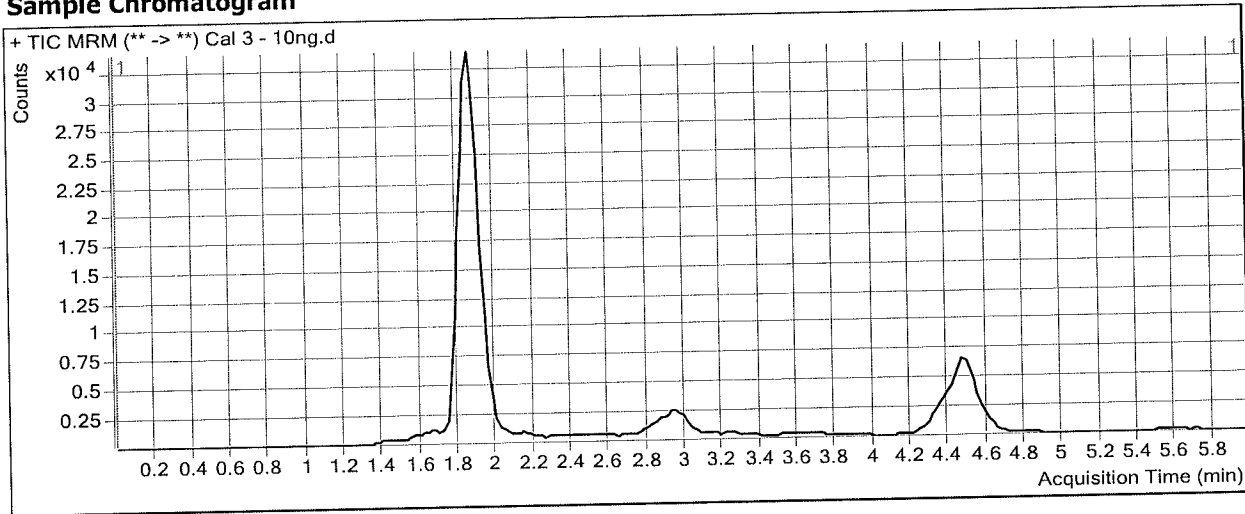
## Cannabinoids Analysis Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin  
**Analysis Time** 4/26/2018 7:24 AM **Analyst Name** ISP Tox  
**Report Time** 4/26/2018 7:32 AM **Reporter Name** ISP Tox  
**Last Calib Update** 4/26/2018 7:24 AM **Batch State** Processed

### Analysis Info

**Acq Time** 2018-04-25 16:07 **Data File** Cal 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

### Sample Chromatogram



### Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	1.875	16337	167885	0.0973	9.8043
THC-COOH	THC-COOH-d9	1.925	13484	56944	0.2368	10.1537
THC	THC-d3	4.491	8500	64816	0.1311	10.2919

# ISP FORENSICS - Cd'A Instrument # 62340

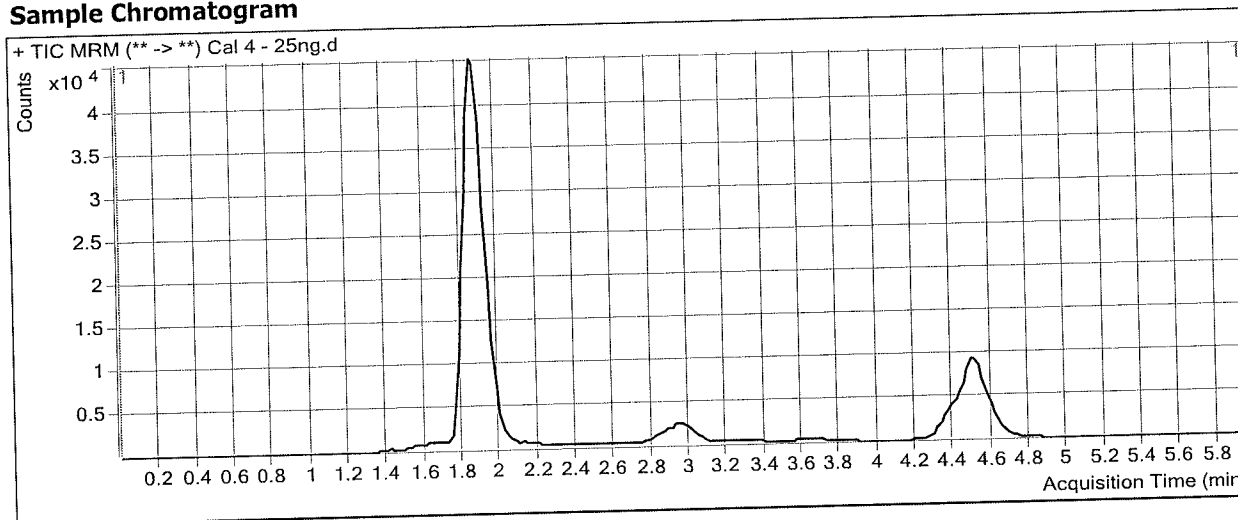
## Cannabinoids Analysis Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin  
**Analysis Time** 4/26/2018 7:24 AM **Analyst Name** ISP Tox  
**Report Time** 4/26/2018 7:32 AM **Reporter Name** ISP Tox  
**Last Calib Update** 4/26/2018 7:24 AM **Batch State** Processed

### Analysis Info

<b>Acq Time</b>	2018-04-25 16:19	<b>Data File</b>	Cal 4 - 25ng.d
<b>Sample Type</b>	Calibration	<b>Sample Name</b>	Cal 4 - 25ng
<b>Dilution</b>	1	<b>Acq Method</b>	AM 27 Quant THC 7-2017.m
<b>Position</b>	P1-D1	<b>Sample Info</b>	
<b>Inj Vol</b>	-1	<b>Comment</b>	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	46542	199489	0.2333	23.2086
THC-COOH	THC-COOH-d9	1.945	33035	65269	0.5061	24.0390
THC	THC-d3	4.511	23380	75624	0.3092	24.1622

# ISP FORENSICS - Cd'A Instrument # 62340

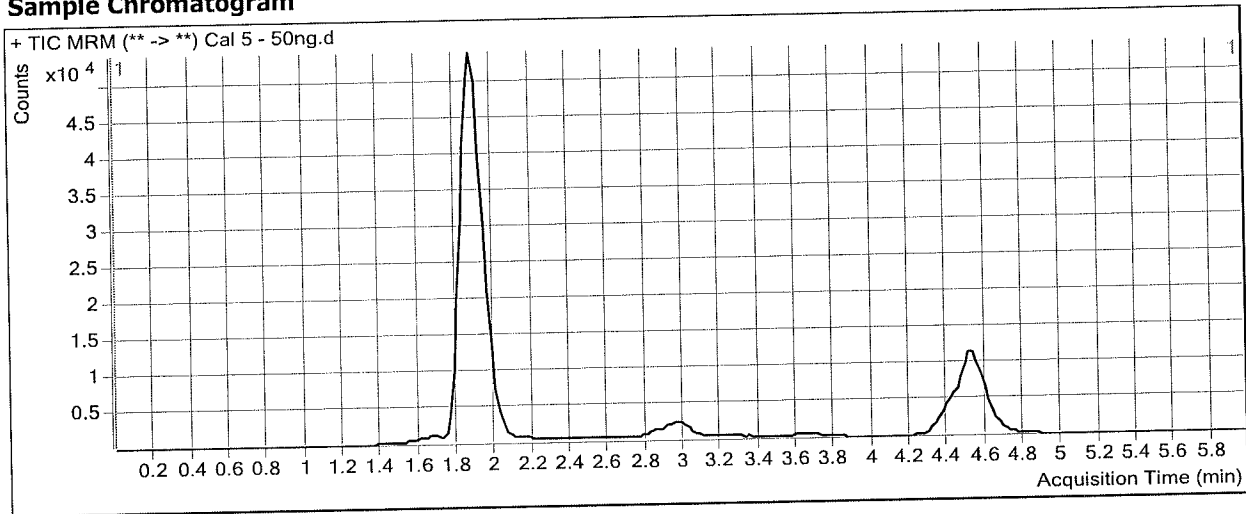
## Cannabinoids Analysis Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin  
**Analysis Time** 4/26/2018 7:24 AM **Analyst Name** ISP Tox  
**Report Time** 4/26/2018 7:32 AM **Reporter Name** ISP Tox  
**Last Calib Update** 4/26/2018 7:24 AM **Batch State** Processed

### Analysis Info

**Acq Time** 2018-04-25 16:31 **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-E1 **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	95234	191057	0.4985	49.3428
THC-COOH	THC-COOH-d9	1.945	62125	61949	1.0028	49.6468
THC	THC-d3	4.511	45001	73053	0.6160	48.0717

# ISP FORENSICS - Cd'A Instrument # 62340

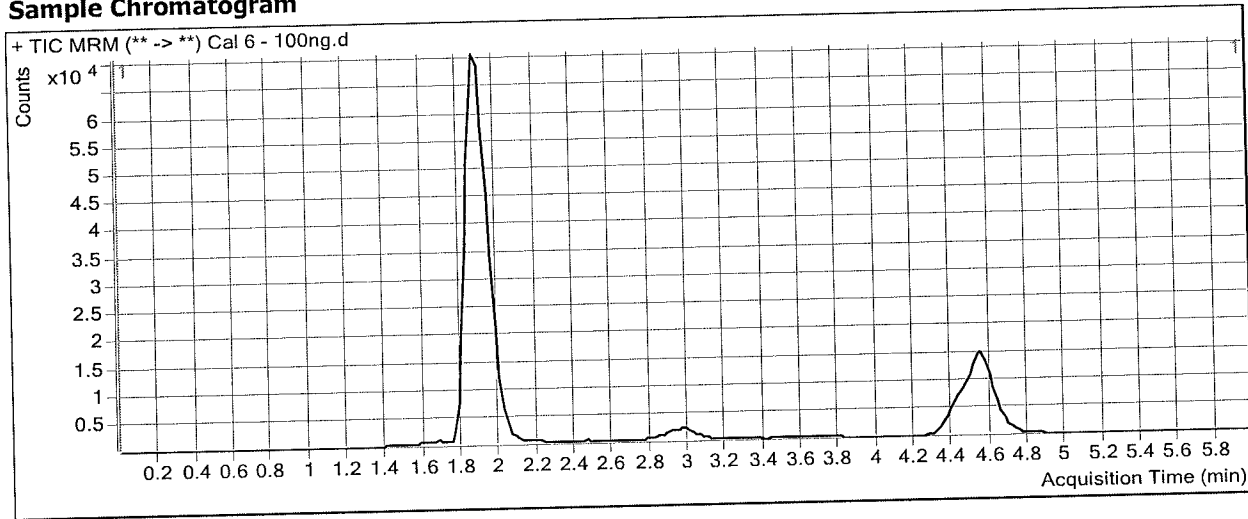
## Cannabinoids Analysis Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin  
**Analysis Time** 4/26/2018 7:24 AM **Analyst Name** ISP Tox  
**Report Time** 4/26/2018 7:32 AM **Reporter Name** ISP Tox  
**Last Calib Update** 4/26/2018 7:24 AM **Batch State** Processed

### Analysis Info

**Acq Time** 2018-04-25 16:42 **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-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	1.895	182559	171929	1.0618	104.8699
THC-COOH	THC-COOH-d9	1.945	112033	55452	2.0204	102.1048
THC	THC-d3	4.551	84131	65240	1.2896	100.5537

# ISP FORENSICS - Cd'A Instrument # 62340

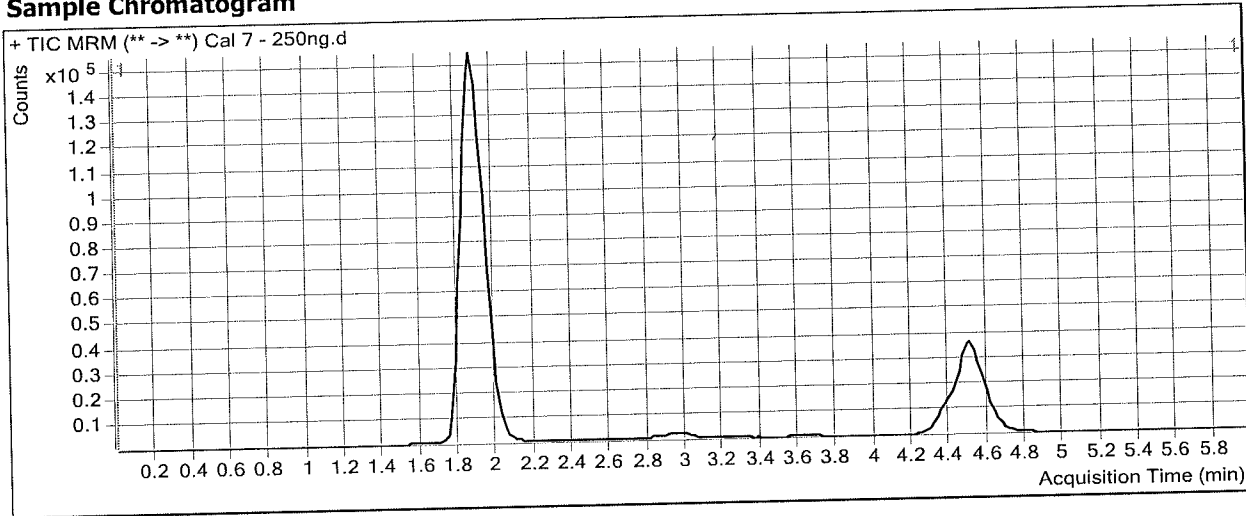
## Cannabinoids Analysis Report

**Batch Data Path** D:\2018 Data\04252018 cann quant\QuantResults\042518 cann quant.batch.bin  
**Analysis Time** 4/26/2018 7:24 AM **Analyst Name** ISP Tox  
**Report Time** 4/26/2018 7:32 AM **Reporter Name** ISP Tox  
**Last Calib Update** 4/26/2018 7:24 AM **Batch State** Processed

### Analysis Info

**Acq Time** 2018-04-25 16:54 **Data File** Cal 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

### Sample Chromatogram



### Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	1.875	542151	216070	2.5091	247.5228
THC-COOH	THC-COOH-d9	1.945	318049	65315	4.8695	248.9901
THC	THC-d3	4.511	256171	79286	3.2310	251.8261