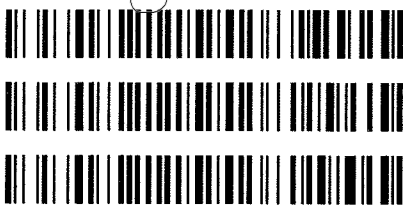
6/7/2018

Worklist: 2453

reviewed 6/8/18

LAB_CASE	ITEM	TASK_ID	DESCRIPTION
C2018-0998	1	117693	AM 27 Blood THC Quant by LC-QQQ
C2018-1019	1	117694	AM 27 Blood THC Quant by LC-QQQ
C2018-1020	1	117695	AM 27 Blood THC Quant by LC-QQQ



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

Extraction Date: 6-6-18 Analyst: Anne Nord
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.
- ☒ 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: 06062018 AM27 cann Quant Batch Name: cann quant
- ☒ 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:

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 each THC, THC-OH 100 ug/ml 100 ul C-THC in 9890 ul meOH
Ppd 6/5/18 Exp: 4/1/19 lot 6518 by AMN

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

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 each by AMN

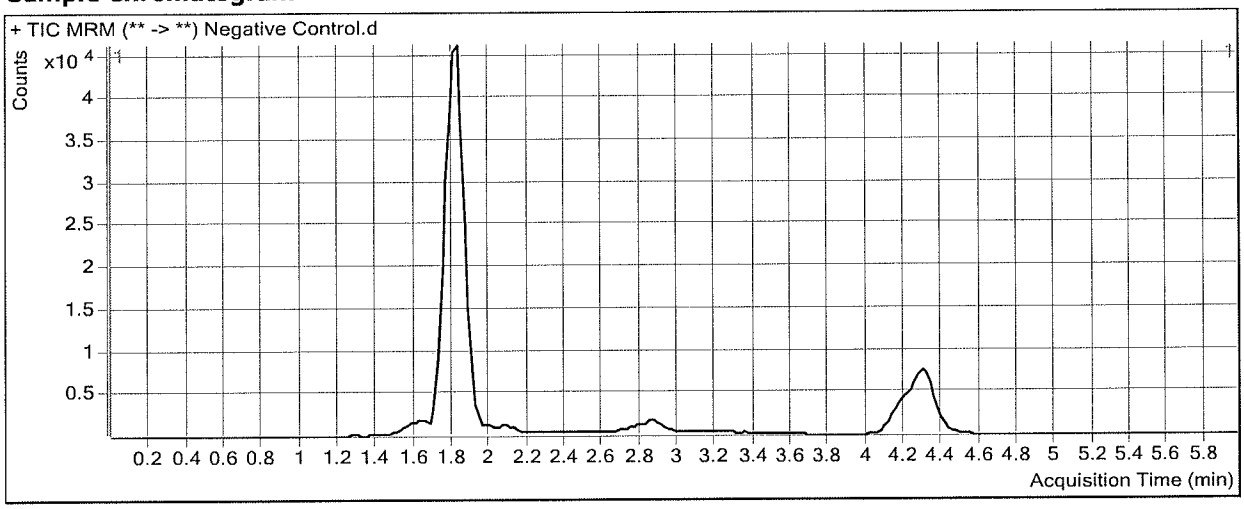
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin		
Analysis Time	6/7/2018 7:26 AM	**Analyst Name**	ISP Tox
Report Time	6/7/2018 8:07 AM	**Reporter Name**	ISP Tox
Last Calib Update	6/7/2018 7:26 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-06-06 15:10	**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-A2	**Sample Info**	
Inj Vol	-1	**Comment**	AM 27 Cannabinoid Confirmation

Sample Chromatogram



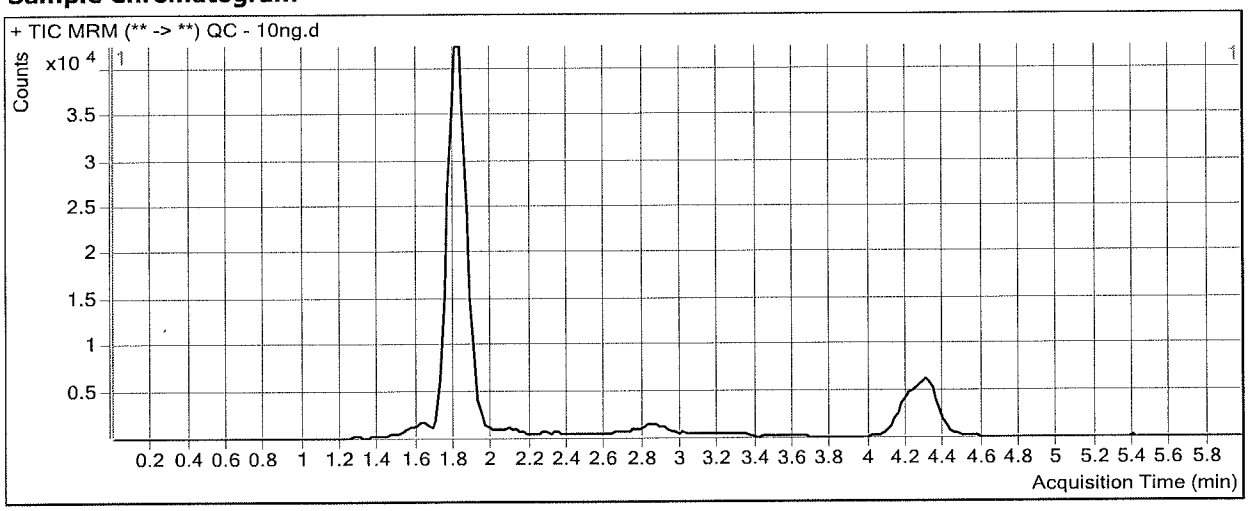
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin		
Analysis Time	6/7/2018 7:26 AM	**Analyst Name**	ISP Tox
Report Time	6/7/2018 8:07 AM	**Reporter Name**	ISP Tox
Last Calib Update	6/7/2018 7:26 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-06-06 15:22	**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

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	1.815	17203	195104	0.0882	9.5581
THC-COOH	THC-COOH-d9	1.885	12592	70496	0.1786	9.5516
THC	THC-d3	4.311	7477	66127	0.1131	10.3752

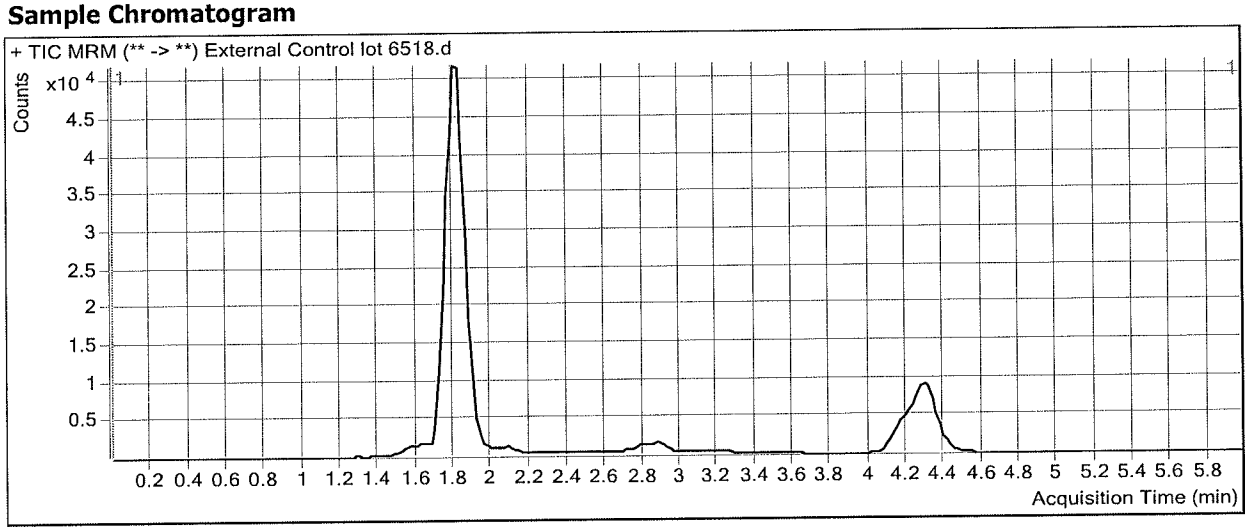
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin		
Analysis Time	6/7/2018 7:26 AM	**Analyst Name**	ISP Tox
Report Time	6/7/2018 8:07 AM	**Reporter Name**	ISP Tox
Last Calib Update	6/7/2018 7:26 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-06-06 15:34	**Data File**	External Control lot 6518.d
Sample Type	QC	**Sample Name**	External Control lot 6518
Dilution	1	**Acq Method**	AM 27 Quant THC 7-2017.m
Position	P1-B2	**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	1.835	20583	250230	0.0823	8.9163
THC-COOH	THC-COOH-d9	1.885	13394	86610	0.1546	8.2183
THC	THC-d3	4.311	10965	89935	0.1219	11.1498

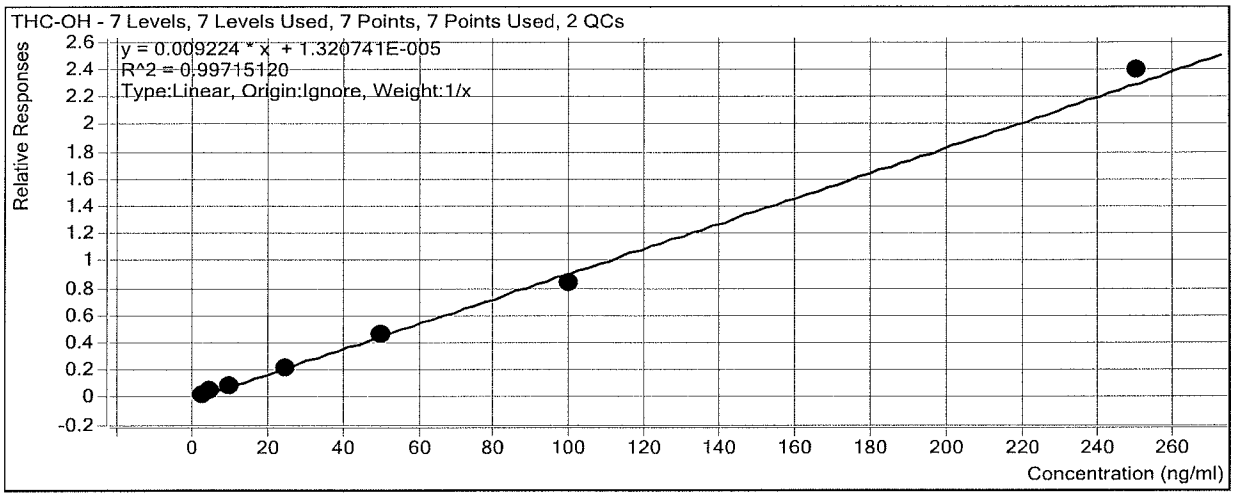
ISP Forensics
Calibration Curve Report

Batch Data Path D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin

Last Calib Update 6/7/2018 7:26 AM **Analyst Name** ISP TOX

Target Compound *THC-OH*

Internal Standard *THC-OH-d3*



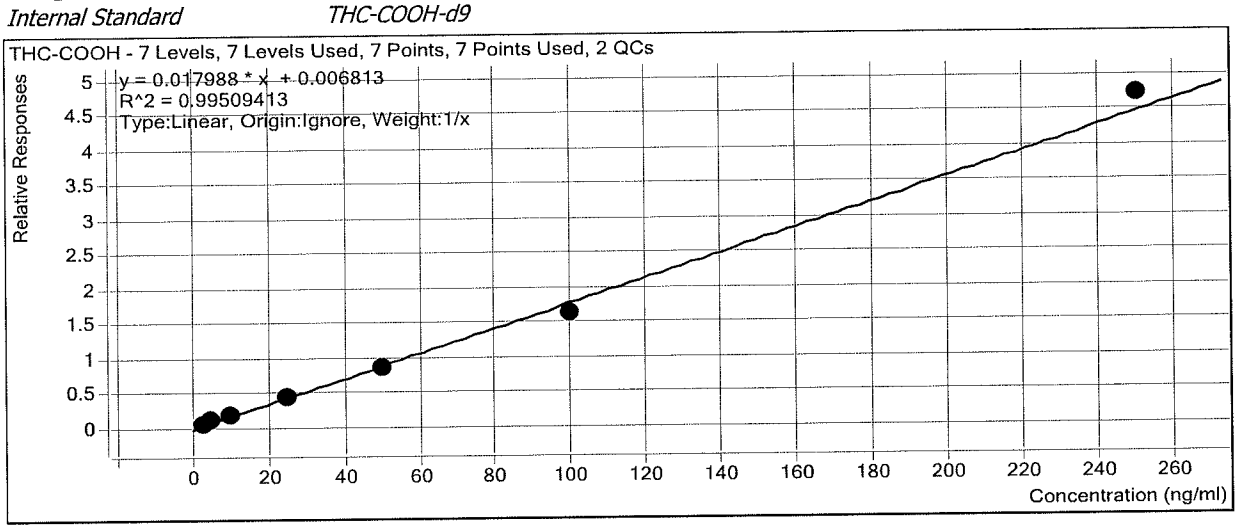
Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1 - 3ng	1	☑	3	3.2	107.4
Cal 2 - 5ng	2	☑	5	5.3	105.4
Cal 3 - 10ng	3	☑	10	9.6	96.2
QC - 10ng	3	☑	10	9.6	95.6
External Control lot 6518	3	☑	10	8.9	89.2
Cal 4 - 25ng	4	☑	25	24.1	96.2
Cal 5 - 50ng	5	☑	50	49.6	99.1
Cal 6 - 100ng	6	☑	100	92.0	92.0
Cal 7 - 250ng	7	☑	250	259.3	103.7

ISP Forensics
Calibration Curve Report

Batch Data Path D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin

Last Calib Update 6/7/2018 7:26 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	3.1	103.2
Cal 2 - 5ng	2	☑	5	5.9	118.0
Cal 3 - 10ng	3	☑	10	9.5	94.6
QC - 10ng	3	☑	10	9.6	95.5
External Control lot 6518	3	☑	10	8.2	82.2
Cal 4 - 25ng	4	☑	25	23.0	92.1
Cal 5 - 50ng	5	☑	50	48.1	96.1
Cal 6 - 100ng	6	☑	100	91.0	91.0
Cal 7 - 250ng	7	☑	250	262.4	105.0

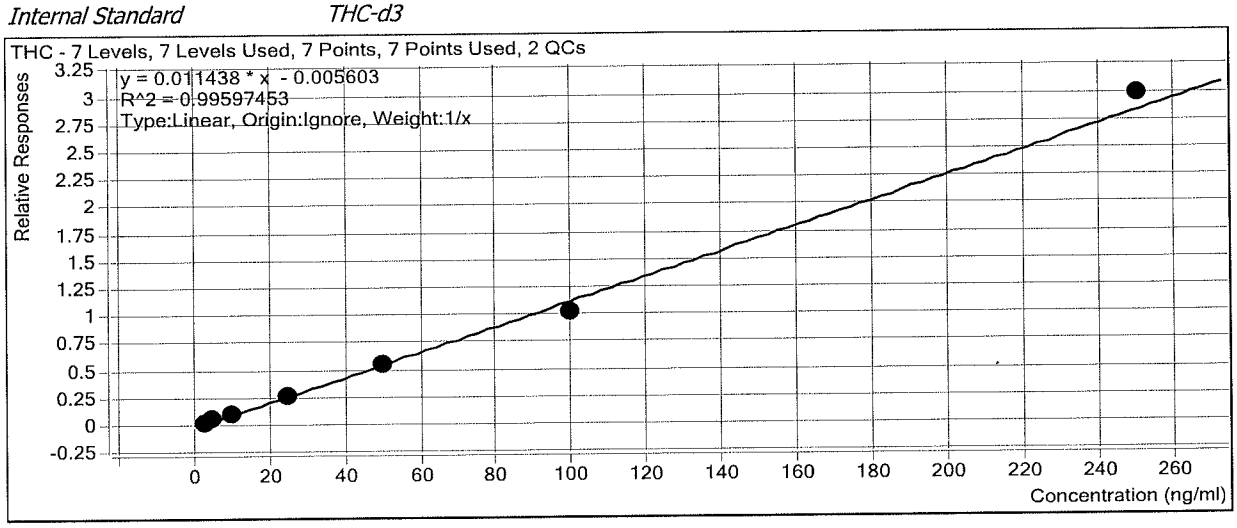
ISP Forensics
Calibration Curve Report

Batch Data Path D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin

Last Calib Update 6/7/2018 7:26 AM **Analyst Name** ISP TOX

Target Compound *THC*

Internal Standard *THC-d3*



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1 - 3ng	1	☑	3	3.2	105.8
Cal 2 - 5ng	2	☑	5	5.5	109.2
Cal 3 - 10ng	3	☑	10	9.8	97.5
QC - 10ng	3	☑	10	10.4	103.8
External Control lot 6518	3	☑	10	11.1	111.5
Cal 4 - 25ng	4	☑	25	23.7	94.7
Cal 5 - 50ng	5	☑	50	48.6	97.3
Cal 6 - 100ng	6	☑	100	90.9	90.9
Cal 7 - 250ng	7	☑	250	261.4	104.6

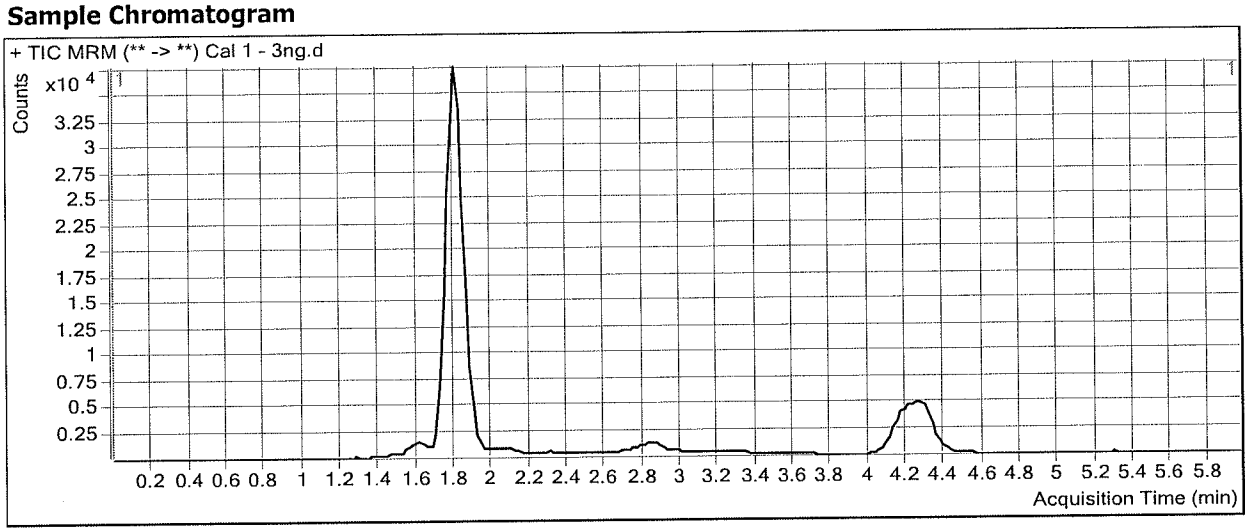
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin		
Analysis Time	6/7/2018 7:26 AM	**Analyst Name**	ISP Tox
Report Time	6/7/2018 8:07 AM	**Reporter Name**	ISP Tox
Last Calib Update	6/7/2018 7:26 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-06-06 13:17	**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	1.815	5129	172463	0.0297	3.2227
THC-COOH	THC-COOH-d9	1.845	3792	60684	0.0625	3.0947
THC	THC-d3	4.271	1891	61570	0.0307	3.1746

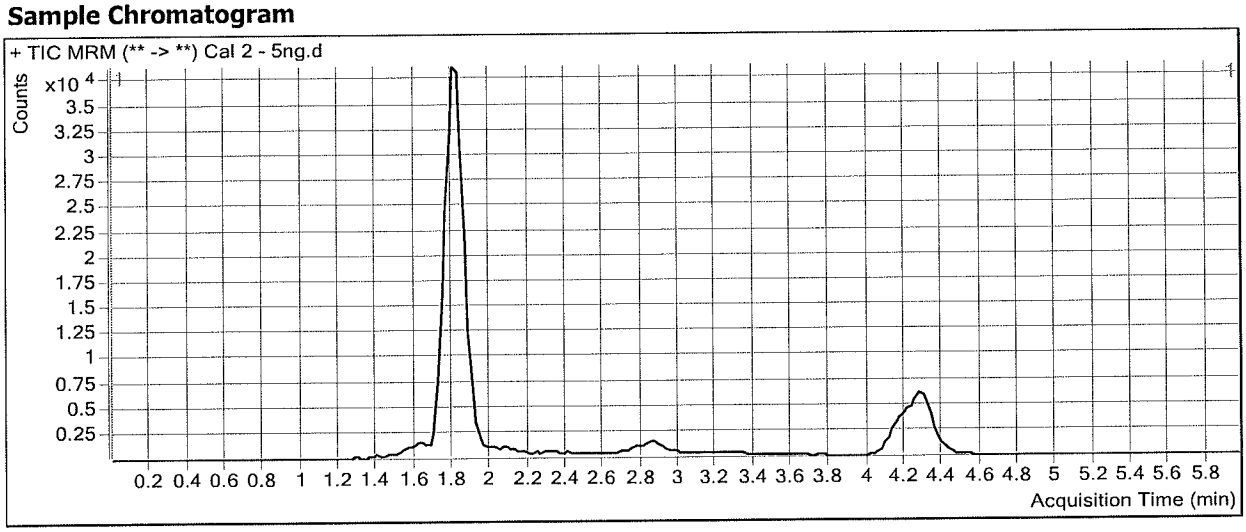
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin		
Analysis Time	6/7/2018 7:26 AM	**Analyst Name**	ISP Tox
Report Time	6/7/2018 8:07 AM	**Reporter Name**	ISP Tox
Last Calib Update	6/7/2018 7:26 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-06-06 13:47	**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	1.815	9145	188155	0.0486	5.2681
THC-COOH	THC-COOH-d9	1.885	7702	68181	0.1130	5.9013
THC	THC-d3	4.291	3849	67701	0.0569	5.4605

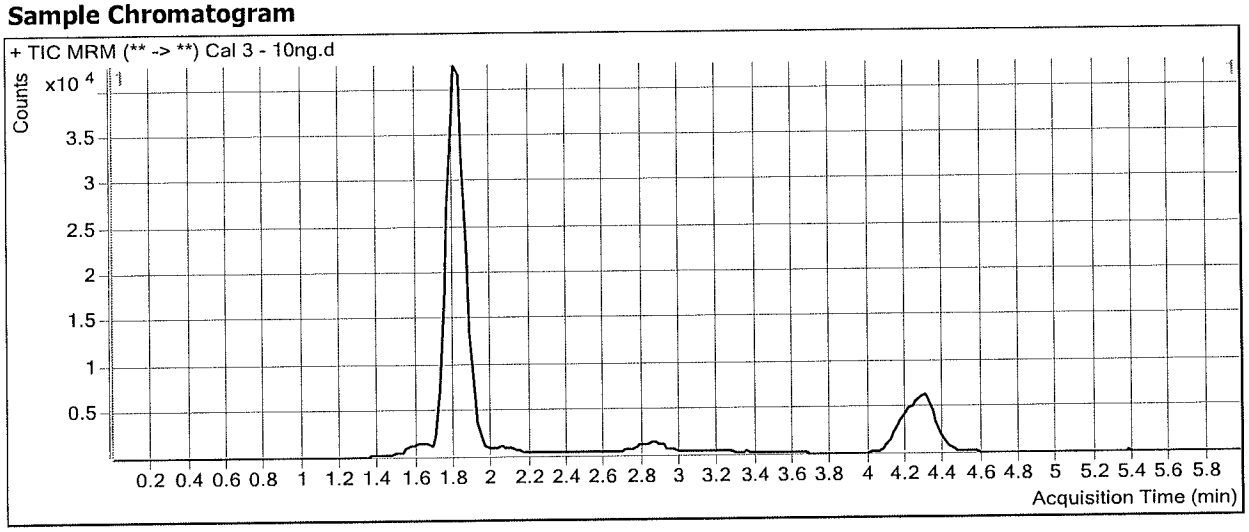
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin		
Analysis Time	6/7/2018 7:26 AM	**Analyst Name**	ISP Tox
Report Time	6/7/2018 8:07 AM	**Reporter Name**	ISP Tox
Last Calib Update	6/7/2018 7:26 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-06-06 13:59	**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	1.815	17100	192770	0.0887	9.6158
THC-COOH	THC-COOH-d9	1.865	12053	68131	0.1769	9.4561
THC	THC-d3	4.311	6986	65958	0.1059	9.7507

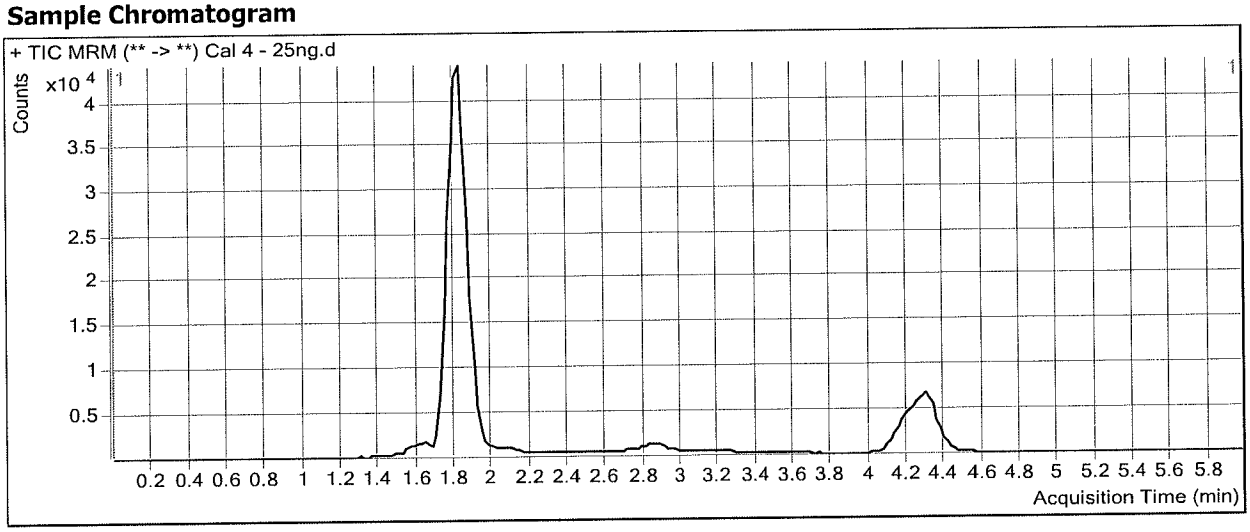
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin		
Analysis Time	6/7/2018 7:26 AM	**Analyst Name**	ISP Tox
Report Time	6/7/2018 8:07 AM	**Reporter Name**	ISP Tox
Last Calib Update	6/7/2018 7:26 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-06-06 14:11	**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-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	THC-OH-d3	1.815	38418	173172	0.2218	24.0504
THC-COOH	THC-COOH-d9	1.865	27638	65631	0.4211	23.0319
THC	THC-d3	4.291	15633	58930	0.2653	23.6837

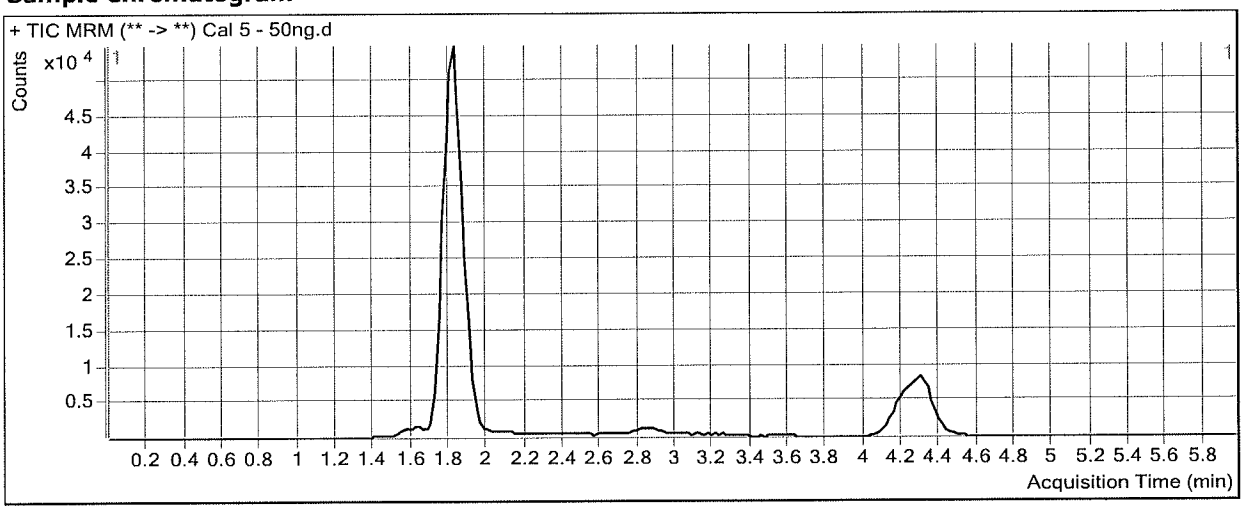
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin		
Analysis Time	6/7/2018 7:26 AM	**Analyst Name**	ISP Tox
Report Time	6/7/2018 8:07 AM	**Reporter Name**	ISP Tox
Last Calib Update	6/7/2018 7:26 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-06-06 14:22	**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	1.815	77859	170276	0.4573	49.5716
THC-COOH	THC-COOH-d9	1.885	52550	60314	0.8713	48.0578
THC	THC-d3	4.291	31679	57534	0.5506	48.6302

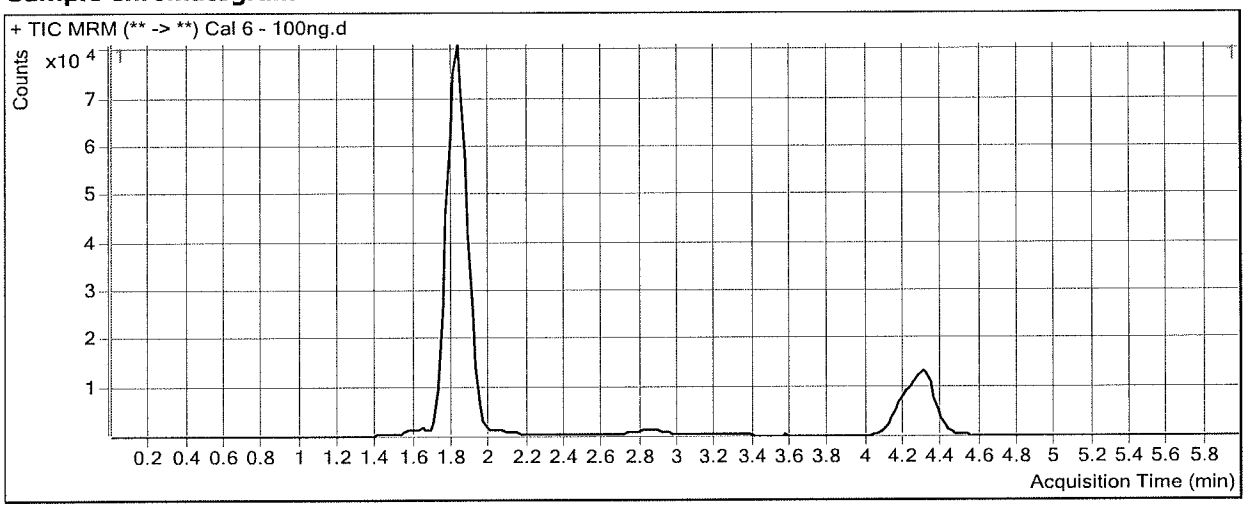
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin		
Analysis Time	6/7/2018 7:26 AM	**Analyst Name**	ISP Tox
Report Time	6/7/2018 8:07 AM	**Reporter Name**	ISP Tox
Last Calib Update	6/7/2018 7:26 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-06-06 14:34	**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	1.815	167404	197256	0.8487	92.0066
THC-COOH	THC-COOH-d9	1.865	112631	68485	1.6446	91.0492
THC	THC-d3	4.291	67704	65465	1.0342	90.9114

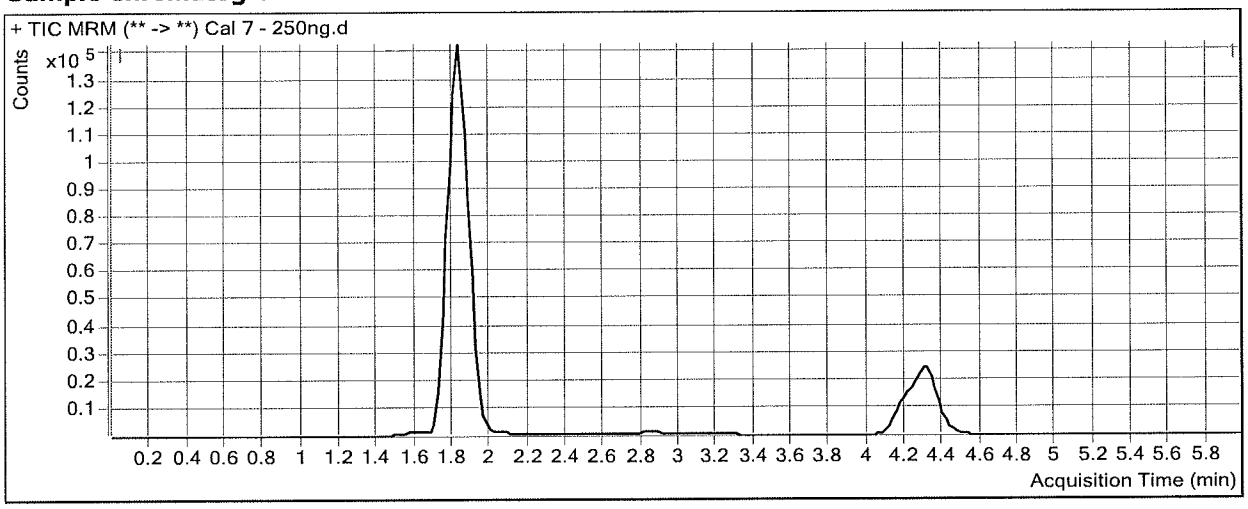
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2018 Data\06062018 AM27 cann quant\QuantResults\cann quant.batch.bin		
Analysis Time	6/7/2018 7:26 AM	**Analyst Name**	ISP Tox
Report Time	6/7/2018 8:07 AM	**Reporter Name**	ISP Tox
Last Calib Update	6/7/2018 7:26 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-06-06 14:46	**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	1.815	421315	176178	2.3914	259.2649
THC-COOH	THC-COOH-d9	1.885	278288	58871	4.7271	262.4089
THC	THC-d3	4.311	171194	57370	2.9841	261.3890