Reviewed 5/10/17

5/3/2017

Worklist: 1709

LAB_CASE	ITEM	TASK_ID	DESCRIPTION
C2017-0651	1	83774	AM 27 Blood THC Quant by LC
C2017-0695	1	83778	AM 27 Blood THC Quant by LC
C2017-0696	1	83776	AM 27 Blood THC Quant by LC
C2017-0708	1	83779	AM 27 Blood THC Quant by LC
C2017-0710	1	83773	AM 27 Blood THC Quant by LC
M2017-0629	1	79158	AM 27 Blood THC Quant by LC
M2017-1456	3	83775	AM 27 Blood THC Quant by LC
M2017-1640	2	83777	AM 27 Blood THC Quant by LC
M2017-1645	1	83772	AM 27 Blood THC Quant by LC
M2017-1763	3	83770	AM 27 Blood THC Quant by LC
P2017-0433	3	79159	AM 27 Blood THC Quant by LC
P2017-0910	1	83771	AM 27 Blood THC Quant by LC

Quantitation of THC and Metabolites in Blood by LC-MS/MS

Extraction Date: 5/2/17 **Analyst:** B. Wyline

PRE-ANALYTIC 0499102

Plate Lot# Custom -~~0490364~~ ★ Plate Exp. 1-28-2018 ~~9-21-2017~~ ★ **External QC** Lot 61317, exp 6-13-17

✓ 1. Ensure all solutions are within expiration date.

- Mobile Phase A: ***0.1% Formic Acid in LCMS Water***
- Mobile Phase B: ***0.1% Formic Acid in LCMS Acetonitrile***
- ***LCMS Methanol***
- **Blank/Negative Blood: Lot *321632-1***
- ***0.1% Formic Acid in water***
- ***MTBE***
- ***Hexane***

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

____ 2. Check levels of mobile phases and needle wash and refill as necessary. Ensure waste is not full.

____ 3. Purge Pump and Load appropriate Acq. Method, allow system to equilibrate for approx. 30 min.

____ 4. Create worklist. Data path name: _____

ANALYTIC

✓ 1. Remove standards plate, blood, and samples from cold storage. Allow to reach room temperature.

✓ 2. Add ***1000 µL blood*** to wells of analytical (standards) plate. Mix via aspirate and dispense. Place cover on Plate
Blank blood for locations containing standards/QCs and internal standards
Sample blood for locations containing only internal standards

✓ 3. Place on shaking incubator at ambient temp., **900rpm for 15 minutes**. *Shaker ID 66759*

✓ 4. Pipette ***500µL 0.1% formic acid*** to all wells of standards 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). Wait 5 min. ***(Load blood samples at 85- 100 PSI- Selector to Right)***

✓ 8. Add ***2.25mL MTBE*** and allow to flow under gravity for 5 minutes. ***(add in 3 increments of 750uL)***

✓ 9. Apply positive pressure for approx. 15 seconds ***(10-15 PSI- Selector to left -.***

✓ 10. Add ***2.25mL Hexane*** and allow to flow under gravity for 5 minutes.. ***(add in 3 increments of 750uL)***

✓ 11. Apply positive pressure for approx. 15 seconds. ***(10-15 PSI Selector to the left)***

✓ 12. Remove collection plate containing eluate.

✓ 13. Place collection plate on SPE Dry and evaporate to dryness at approx. 35°C. *SPE Dry ID 66819*

✓ 14. Reconstitute in ***100 µL MeOH*** and heat seal plate with foil. Place in autosampler and run worklist.

POST-ANALYTIC

✓ 1. Open quantitation software and create a new quantitation batch.
✓ Batch name: 5-3-17 THC Quant

✓ 2. Make any necessary integration changes. Limit curves based on validated linear ranges (3-50ng/mL).

✓ 3. Were all appropriate standards used in the curve for each analyte? (Y) / N
Are r^2 values ≥0.98 for each analyte? (Y) / N

✓ 4. Did all QCs pass for each analyte? (Y) / N Were QCs entered into QC charting? (Y) / N

✓ 5. Central File Packet to include: ✓ LIMS Worklist: ✓ Method Checklist ✓ Calibration and Control Reports

COMMENTS

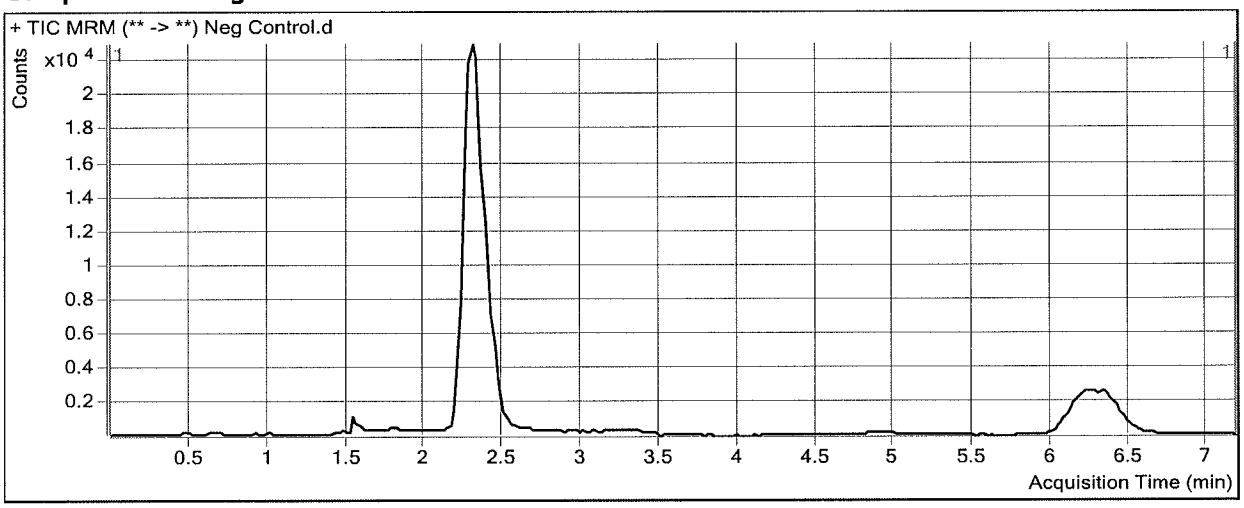
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 09:45	**Data File**	Neg Control.d
Sample Type	Sample	**Sample Name**	Neg Control
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	P2-a2	**Sample Info**	
Inj Vol	-1	**Comment**	AM 27 cannabinoid confirmation

Sample Chromatogram



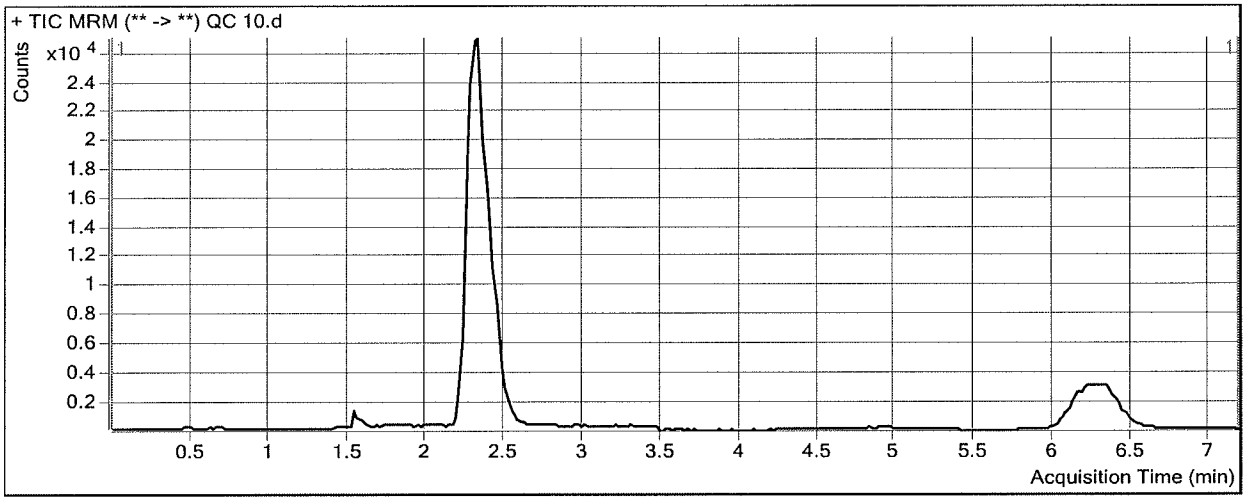
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 09:57	**Data File**	QC 10.d
Sample Type	QC	**Sample Name**	QC 10
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	P2-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	2.336	17193	179160	0.0960	10.1259
THC-COOH	THC-COOH-d9	2.446	12258	53618	0.2286	10.6597
THC	THC-d3	6.313	7064	55465	0.1274	10.6306

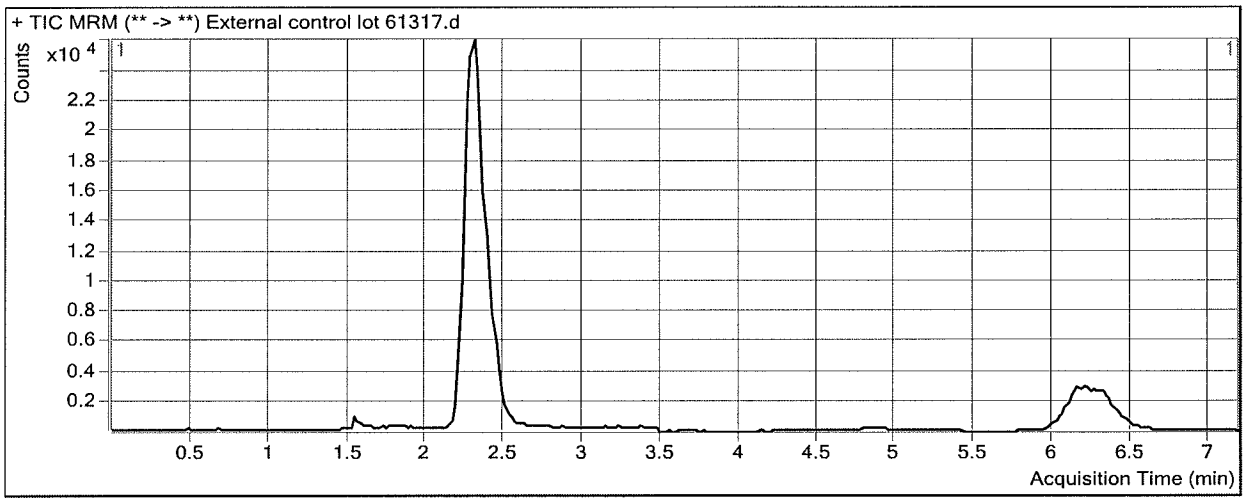
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 10:09	**Data File**	External control lot 61317.d
Sample Type	QC	**Sample Name**	External control lot 61317
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	p2b2	**Sample Info**	
Inj Vol	-1	**Comment**	AM 27 cannabinoid confirmation 10 ng

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-d3	2.316	19172	167558	0.1144	12.0192
THC-COOH	THC-COOH-d9	2.426	9071	47510	0.1909	8.8394
THC	THC-d3	6.293	6331	52086	0.1215	10.1387

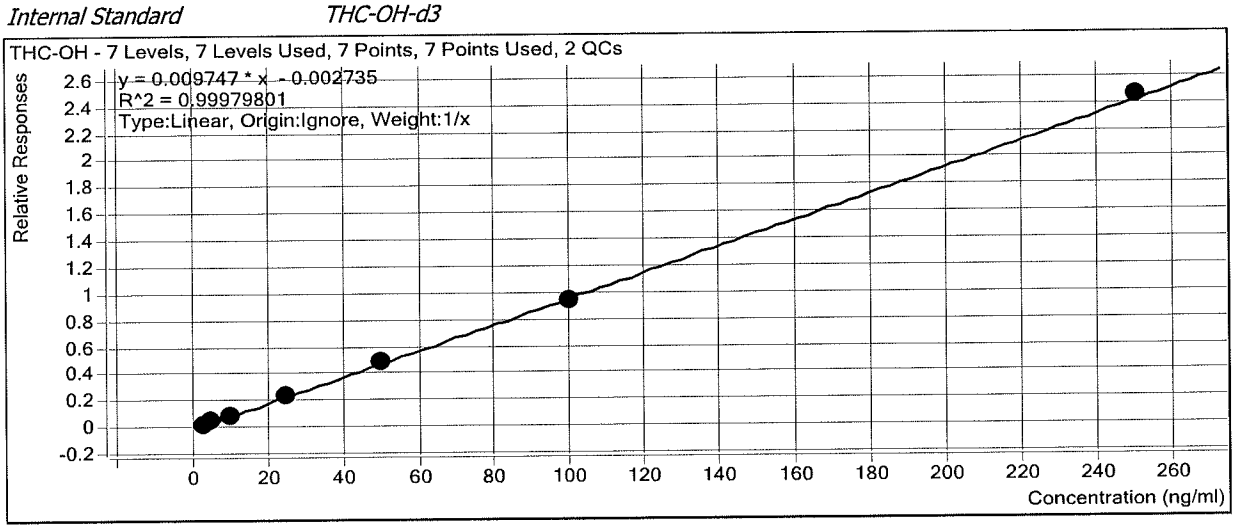
ISP Forensics
Calibration Curve Report

Batch Data Path D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin

Last Calib Update 5/5/2017 4:22 PM **Analyst Name** ISP TOX

Target Compound *THC-OH*

Internal Standard *THC-OH-d3*



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1- 3ng/mL	1	☑	3	3.1	101.8
cal 2- 5ng/mL	2	☑	5	5.0	100.5
cal 3 - 10ng/mL	3	☑	10	9.9	98.9
QC 10	3	☑	10	10.1	101.3
External control lot 61317	3	☑	10	12.0	120.2
cal 4 - 25ng/mL	4	☑	25	25.0	100.1
cal 5 - 50ng/mL	5	☑	50	50.0	100.1
cal 6 - 100ng/mL	6	☑	100	97.7	97.7
cal 7 - 250ng/mL	7	☑	250	252.3	100.9

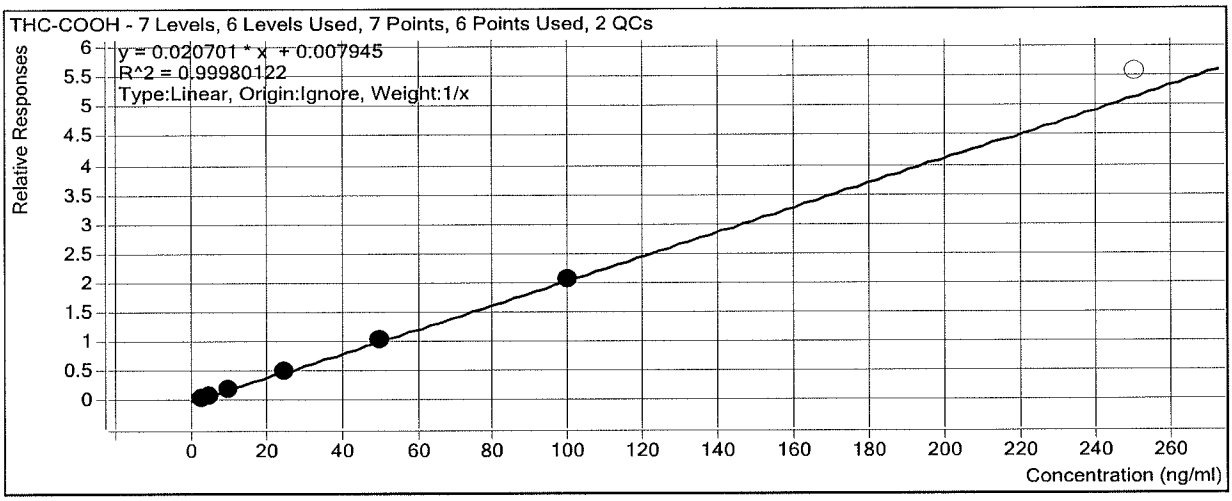
ISP Forensics
Calibration Curve Report

Batch Data Path D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin

Last Calib Update 5/5/2017 4:22 PM **Analyst Name** ISP TOX

Target Compound *THC-COOH*

Internal Standard *THC-COOH-d9*



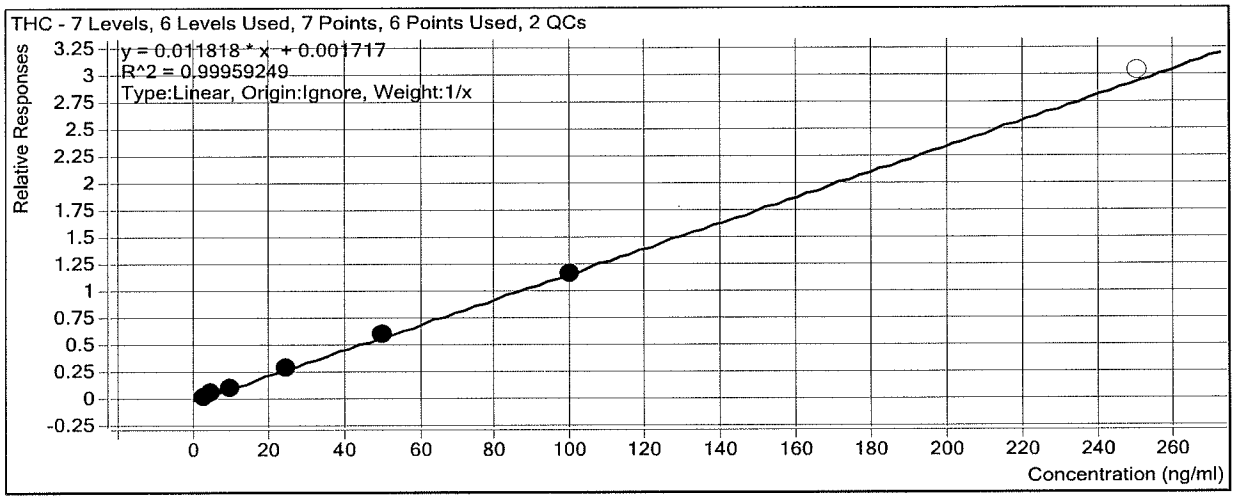
Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1- 3ng/mL	1	☑	3	3.0	101.3
cal 2- 5ng/mL	2	☑	5	4.9	97.8
cal 3 - 10ng/mL	3	☑	10	10.0	99.6
QC 10	3	☑	10	10.7	106.6
External control lot 61317	3	☑	10	8.8	88.4
cal 4 - 25ng/mL	4	☑	25	25.1	100.4
cal 5 - 50ng/mL	5	☑	50	50.9	101.8
cal 6 - 100ng/mL	6	☑	100	99.1	99.1
cal 7 - 250ng/mL	7	☐	250	268.7	107.5

ISP Forensics
Calibration Curve Report

Batch Data Path D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin

Last Calib Update 5/5/2017 4:22 PM **Analyst Name** ISP TOX

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



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1- 3ng/mL	1	☑	3	2.9	96.8
cal 2- 5ng/mL	2	☑	5	5.3	105.2
cal 3 - 10ng/mL	3	☑	10	9.8	97.7
QC 10	3	☑	10	10.6	106.3
External control lot 61317	3	☑	10	10.1	101.4
cal 4 - 25ng/mL	4	☑	25	24.7	98.8
cal 5 - 50ng/mL	5	☑	50	51.1	102.3
cal 6 - 100ng/mL	6	☑	100	99.2	99.2
cal 7 - 250ng/mL	7	☐	250	256.6	102.6

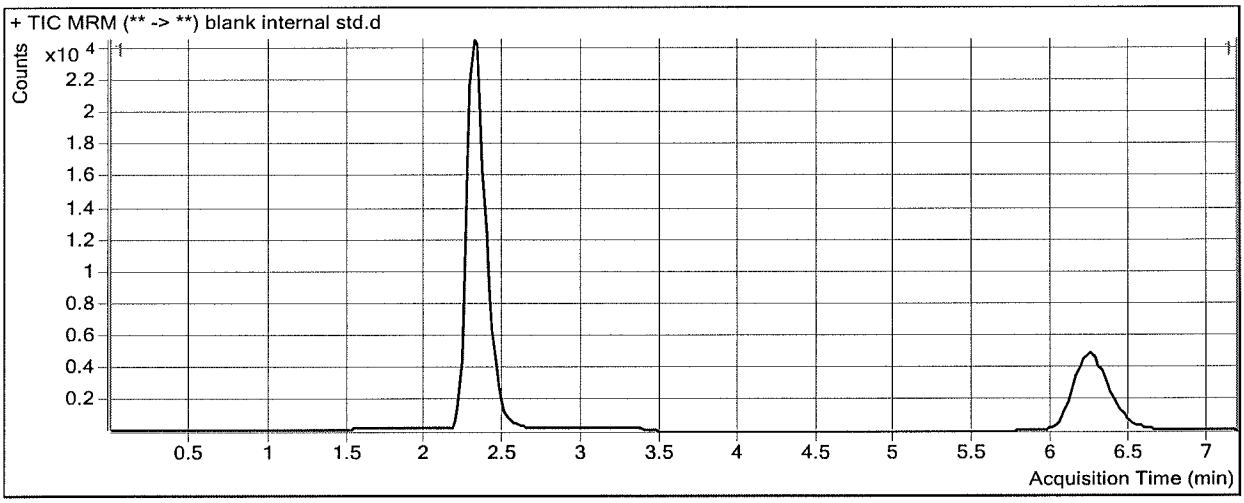
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 09:33	**Data File**	blank internal std.d
Sample Type	Sample	**Sample Name**	blank internal std
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	Vial 2	**Sample Info**	
Inj Vol	-1	**Comment**	AM 27 cannabinoid confirmation

Sample Chromatogram



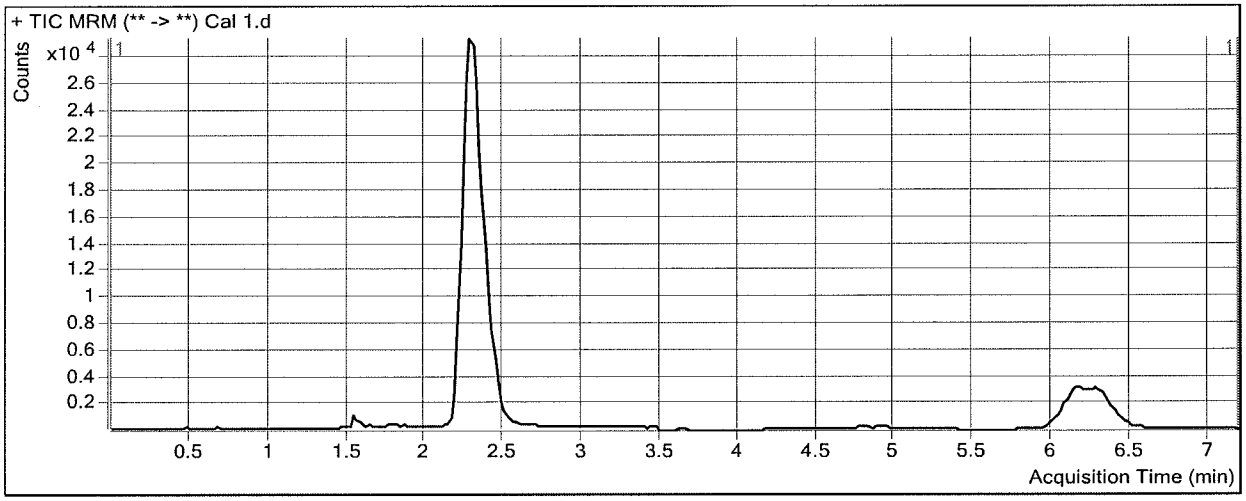
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 08:10	**Data File**	Cal 1.d
Sample Type	Calibration	**Sample Name**	Cal 1- 3ng/mL
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	P2-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	2.296	5600	207060	0.0270	3.0551
THC-COOH	THC-COOH-d9	2.426	4470	63066	0.0709	3.0398
THC	THC-d3	6.273	2249	62426	0.0360	2.9028

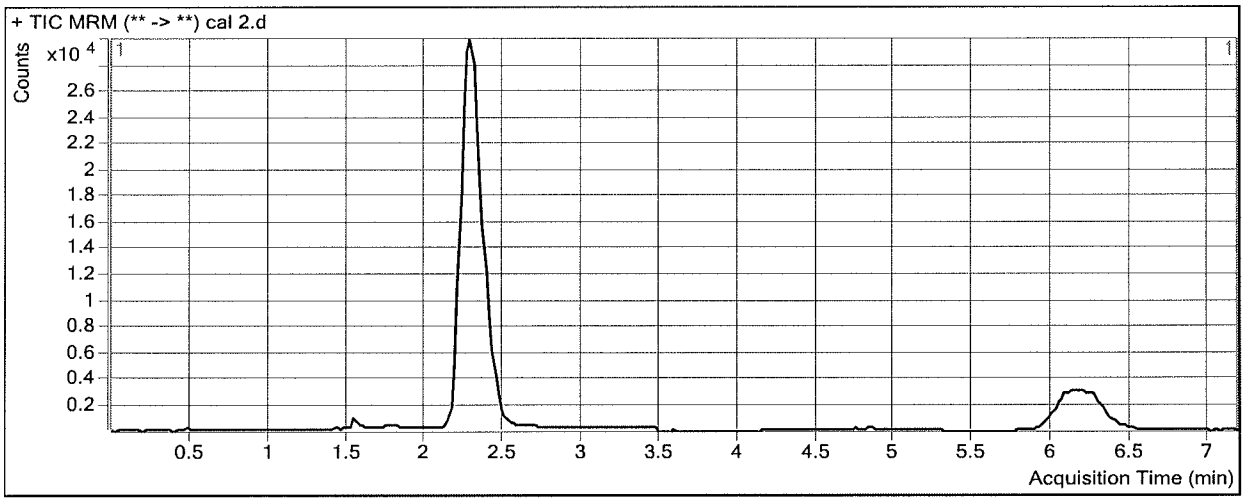
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 08:22	**Data File**	cal 2.d
Sample Type	Calibration	**Sample Name**	cal 2- 5ng/mL
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	P2-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	2.296	9437	204126	0.0462	5.0238
THC-COOH	THC-COOH-d9	2.386	6893	63149	0.1092	4.8893
THC	THC-d3	6.253	3700	57908	0.0639	5.2605

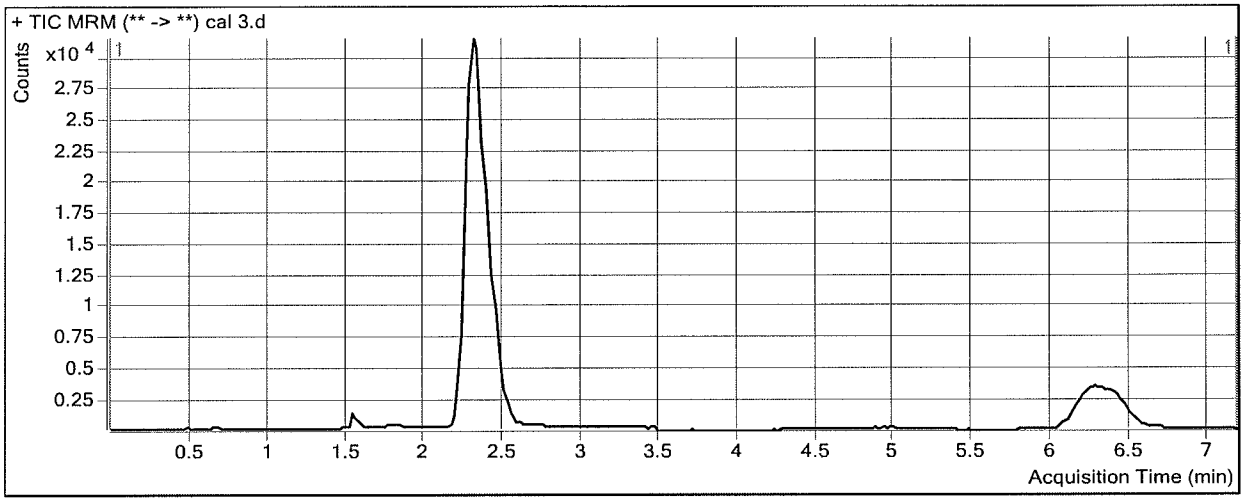
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 08:34	**Data File**	cal 3.d
Sample Type	Calibration	**Sample Name**	cal 3 - 10ng/mL
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	P2-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	2.336	19674	210069	0.0937	9.8888
THC-COOH	THC-COOH-d9	2.446	13557	63343	0.2140	9.9556
THC	THC-d3	6.373	7365	62823	0.1172	9.7748

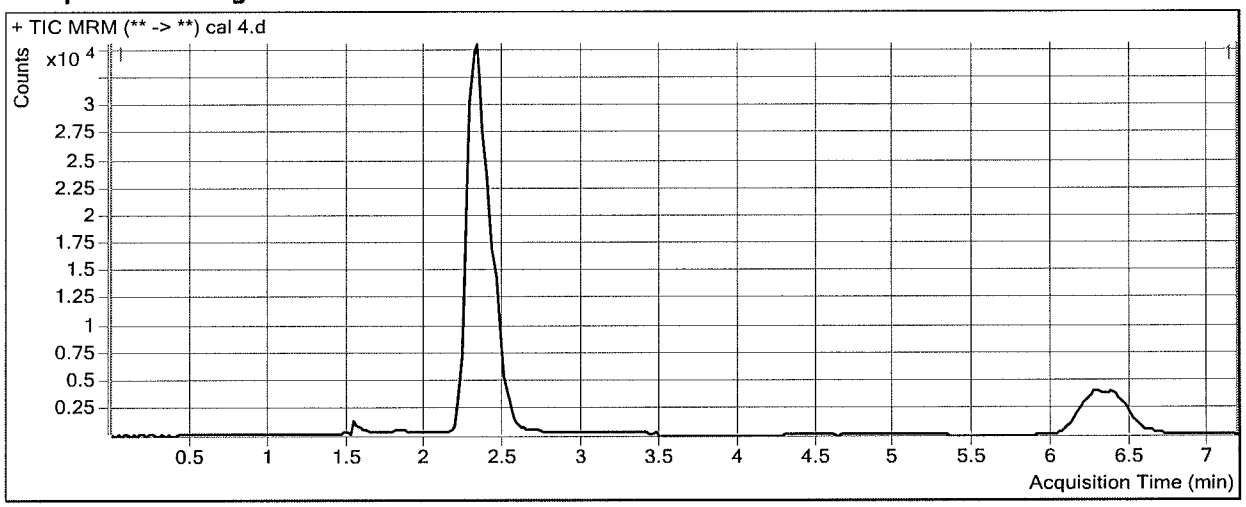
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 08:46	**Data File**	cal 4.d
Sample Type	Calibration	**Sample Name**	cal 4 - 25ng/mL
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	P2-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	2.336	49897	206883	0.2412	25.0248
THC-COOH	THC-COOH-d9	2.446	32925	62408	0.5276	25.1020
THC	THC-d3	6.373	17950	61153	0.2935	24.6914

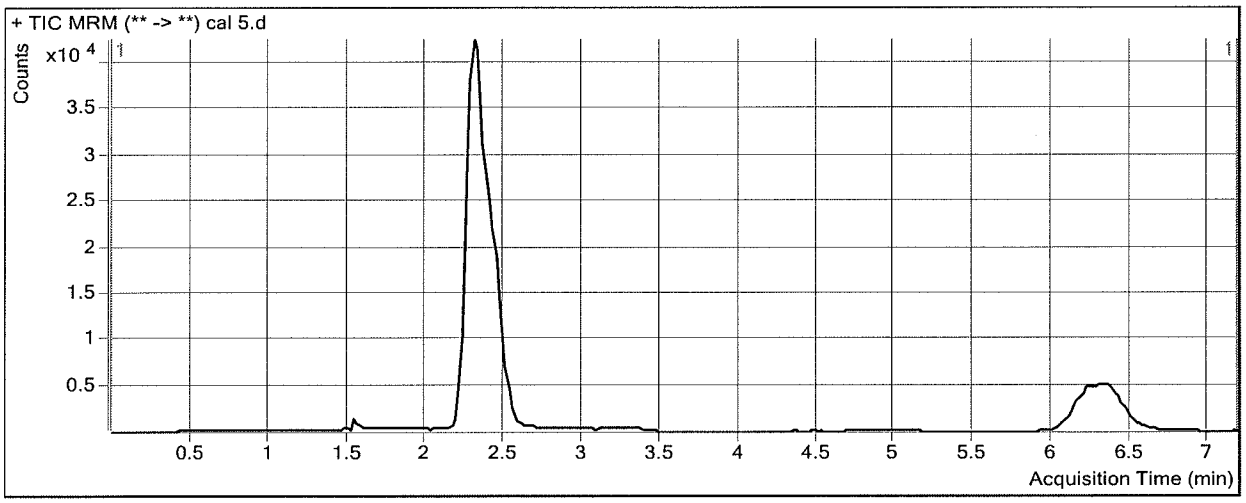
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 08:57	**Data File**	cal 5.d
Sample Type	Calibration	**Sample Name**	cal 5 - 50ng/mL
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	P2-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	2.316	97574	201154	0.4851	50.0463
THC-COOH	THC-COOH-d9	2.446	63597	59894	1.0618	50.9101
THC	THC-d3	6.353	36041	59457	0.6062	51.1449

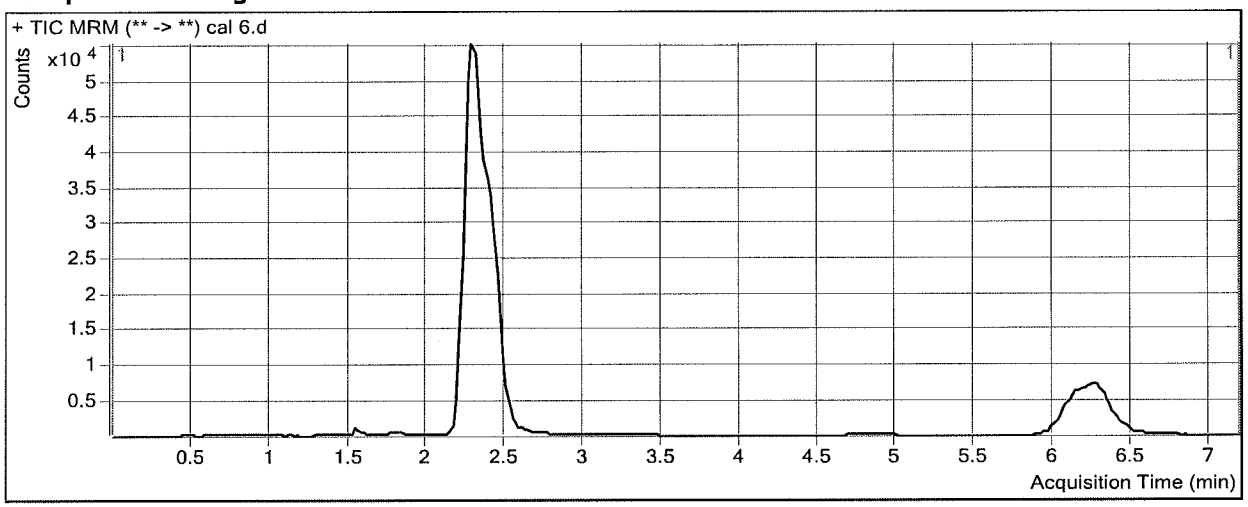
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 09:09	**Data File**	cal 6.d
Sample Type	Calibration	**Sample Name**	cal 6 - 100ng/mL
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	P2-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	187826	197795	0.9496	97.7043
THC-COOH	THC-COOH-d9	2.406	120388	58457	2.0594	99.1032
THC	THC-d3	6.273	65847	56069	1.1744	99.2255

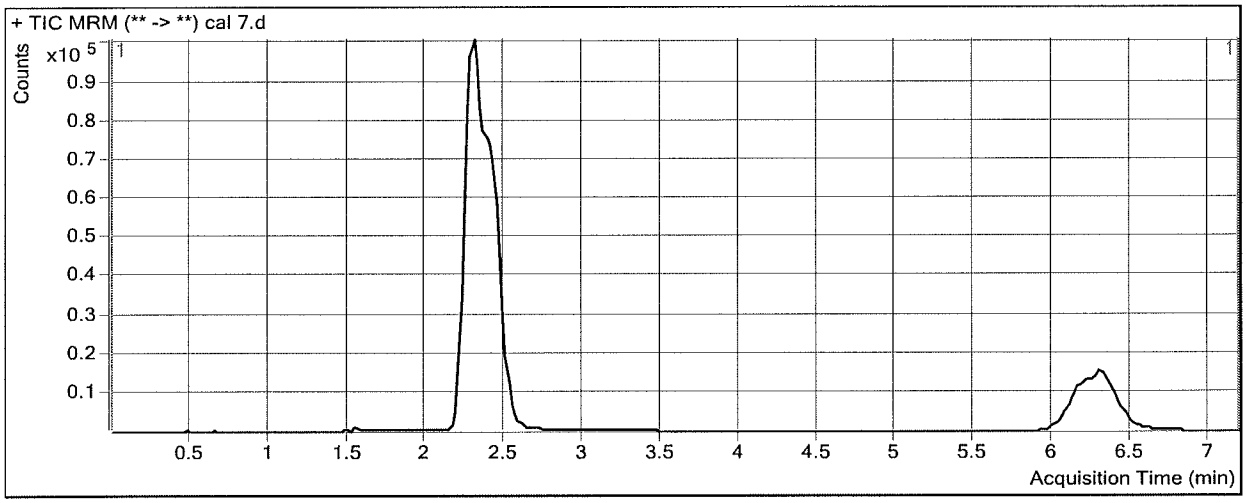
ISP FORENSICS - Cd'A Instrument # 62340
Cannabinoids Analysis Report

Batch Data Path	D:\2017 Data\5-3-17 THC Quant\QuantResults\5-3-17 THC Quant.batch.bin		
Analysis Time	5/5/2017 4:22 PM	**Analyst Name**	ISP Tox
Report Time	5/5/2017 4:23 PM	**Reporter Name**	ISP Tox
Last Calib Update	5/5/2017 4:22 PM	**Batch State**	Processed

Analysis Info

Acq Time	2017-05-05 09:21	**Data File**	cal 7.d
Sample Type	Calibration	**Sample Name**	cal 7 - 250ng/mL
Dilution	1	**Acq Method**	Quant THC 2017.m
Position	P2-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	2.316	502950	204780	2.4561	252.2569
THC-COOH	THC-COOH-d9	2.426	304736	54710	5.5701	268.6943
THC	THC-d3	6.293	178411	58795	3.0345	256.6134

Run Date: 5-3-2017 extracted

Worklist ID: _____

	1	2	3	4	5	6
A	Std 1 + ISTD (3ng/mL)	neg *	C651			QC 1 + ISTD (10ng/ml)
B	Std 2 + ISTD (5ng/ml)	ext ctrl *	m1456-3			Std 7 + ISTD (250ng/ml)
C	Std 3 + ISTD (10ng/ml)	P433	purple for C696 (B)			Std 6 + ISTD (100ng/ml)
D	Std 4 + ISTD (25ng/ml)	m629	m1640-2.			Std 5 + ISTD (50ng/ml)
E	Std 5 + ISTD (50ng/ml)	m1763-3	C695			Std 4 + ISTD (25ng/ml)
F	Std 6 + ISTD (100ng/ml)	P910	C708			Std 3 + ISTD (10ng/ml)
G	Std 7 + ISTD (250ng/ml)	m1645-1 (B)	P910R			Std 2 + ISTD (5ng/ml)
H	QC 1 + ISTD (10ng/ml)	C710				Std 1 + ISTD (3ng/mL)

Custom thc plates