reviewed 7/23/18 amn

7/10/2018

Worklist: 2555

LAB_CASE	ITEM	TASK_ID	DESCRIPTION
M2018-2977	1	120819	AM 27 Blood THC Quant by LC-QQQ
M2018-3061	1	120820	AM 27 Blood THC Quant by LC-QQQ
P2018-1849	1	120821	AM 27 Blood THC Quant by LC-QQQ
P2018-1873	1	120822	AM 27 Blood THC Quant by LC-QQQ
P2018-1875	1	120823	AM 27 Blood THC Quant by LC-QQQ
P2018-1890	1	120824	AM 27 Blood THC Quant by LC-QQQ
P2018-1892	1	120825	AM 27 Blood THC Quant by LC-QQQ
P2018-1894	1	120826	AM 27 Blood THC Quant by LC-QQQ

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

P

Extraction Date: 07/10/18 Analyst: Sarah Pickle
Plate lot#: 0515037 Plate Expiration: 09/28/18

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: 361331-1 **Column**: UCT Selectra DA 100 x 2.1mm 3um
Blank Urine: POC062718
LCMS-QQQ ID: 59740

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: Data Path: _____

Urine Hydrolysis:

☒ 1. Pipette 1.5 mL urine into empty 48 well plate.
☒ 2. Add 250ul 1N KOH to urine samples.
☒ 3 P 2. Place on shaking incubator at 40 degrees for 15 mins.

Analytic:

☒ 1. Remove standards, plate, controls, and samples from cold storage. Allow to reach room temperature.
☒ 2. Pipette **1000µL blood/urine (calibrated pipette) Pipette ID: 3382167** in wells of analytical (standards) plate.
☒ 3. Place on shaking incubator at ambient temp., 900rpm for 15 minutes. *Shaker ID: 067105*
☒ 4. Pipette **500µL 0.1% formic acid in water** in wells of analytical plate for blood samples. Pipette **500ul saturated phosphate buffer** for urine samples.
☒ 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: 067104
☒ 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: 067103*
☒ 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: 071018 THC Quant Urine Validation SP worklist 2555
Batch Name: 071018 THCQ SP
☒ 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: *Curve Range Limited: THC-COOH 5-250*

Idaho State Police Forensic Services

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

Analyst: Sarah Pickle
Extraction Date: 07/10/18
Worklist Number: 2555

Reagent	*Lot Number*	*Expiration Date*	*Date in Service*	*Date Out of Service*	*Initials*
ToxBox THC/THC Metabolite Plate	0515037	09/28/18			
Negative Blood	361331-1		05/25/18		
Negative Urine	POC062718		06/27/18		
Methanol External Control Solution	WS020718	02/07/19	02/07/18		
Blood External Control Solution	061718	06/17/19	06/17/18		
Urine External Controls	062818	06/28/19	06/28/18		
Methyl Tert-Butyl Ether (MTBE) 99.9%	A0375555		06/26/17		
Hexanes (ACS)	101642		10/26/17		
Methanol (LCMS Grade)	177145		04/11/18		
1 N KOH	091817		09/18/18		
Saturated Phosphate Buffer pH 1.8	020118		02/01/18		
0.1% Formic Acid in Water (Mobile Phase A)	166541		06/26/17		
0.1% Formic Acid in Acetonitrile (Mobile Phase B)	176190		02/08/18		
Needle Rinse--75% LCMS MeOH in LCMS Water	070518		07/05/18		

Methanol External Control Solution (Lot: WS020718)
10 ul of 1mg/mL THC, 100 ul of 100 ug/mL THC-OH, C-THC in 9790 ul MeOH

Component	*Source*	*Source Lot Number*	*Expiration Date*
Methanol (LCMS)	Fisher	172516	
THC	Cerilliant	FE04231406	04/30/2019
C-THC	Cayman	0497429	02/08/2019
THC-OH	Cerilliant	FE01121503	01/31/2020
Prepared:	02/07/18		
Prepared By:	Tamara Salazar		
Expires:	02/07/19		

Blood External Control Solution (Lot: 061718)
100 ul of methanol external control solution was added to 9900 ul of blood.

Component	*Source*	*Source Lot Number*
Negative Blood	Hemostat	361331-1
Methanol External Control Solution		WS020718

PS

Prepared:	06/17/18
Prepared by:	Tamara Salazar
Expires:	06/17/19

Urine External Control Solutions (Lot: 062818)

Three Controls were made by adding 50 ul, 100 ul, and 200 ul of methanol external control to 4.5 mL of negative urine.

Component	*Source*	*Source Lot Number*
Negative Urine		POC062718
Methanol External Control Solution		WS020718
Prepared:	06/28/18	
Prepared by:	Sarah Pickle	
Expires:	06/28/19	

1 N KOH (Lot: 091817)

Component	*Source*	*Source Lot Number*
Potassium Hydroxide	Fisher	034727
DI Water	-	-
Prepared:	09/18/17	
Prepared By:	Celena Shrum	

Saturated Phosphate Buffer pH 1.8 (Lot: 020118)

Component	*Source*	*Source Lot Number*
Potassium Phosphate monobasic	Fisher	L10-021-61
DI Water	-	-
Prepared:	02/01/18	
Prepared By:	Celena Shrum	

Needle Rinse (75% LCMS MeOH in LCMS Water) (Lot: 070518)

Component	*Source*	*Source Lot Number*
MeOH (LCMS Grade)	Fisher	177145
Water (LCMS Grade)	Fisher	177528
Prepared:	07/05/18	
Prepared By:	Tamara Salazar	

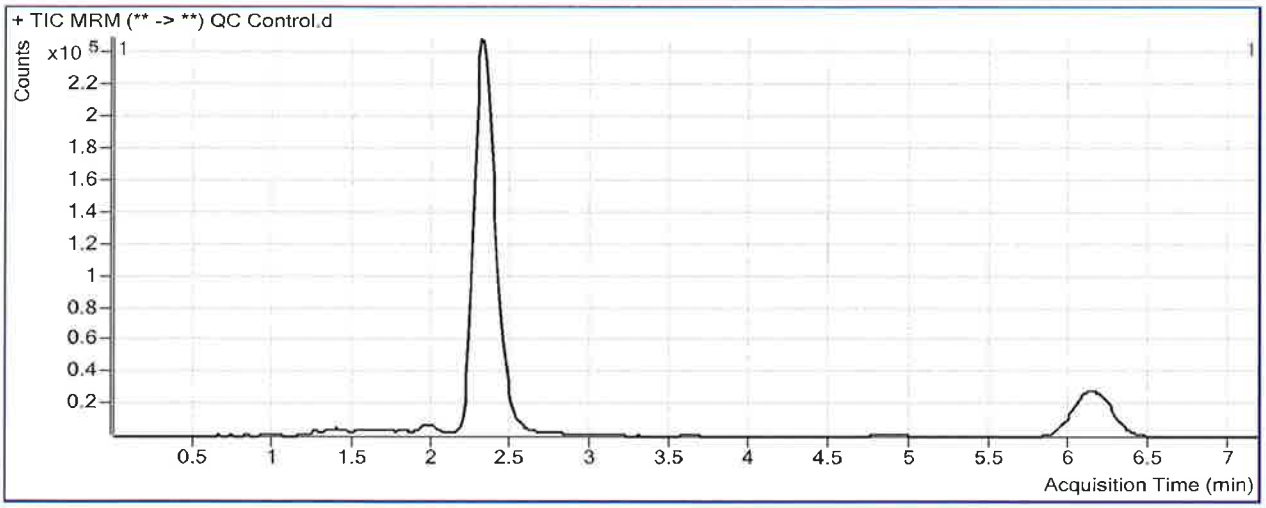
ISP FORENSICS - Pocatello Instrument # 59740
Cannabinoids Analysis Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071		
Analysis Time	7/11/2018 7:39 AM	**Analyst Name**	ISPUser
Report Time	7/11/2018 7:41 AM	**Reporter Name**	ISPUser
Last Calib Update	7/11/2018 7:39 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-07-10 13:16	**Data File**	QC Control.d
Sample Type	Sample	**Sample Name**	QC Control
Dilution	1	**Acq Method**	THC Quant 051517 workingmm.m
Position	P1-A6	**Sample Info**	
Inj Vol	-1	**Comment**	

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.319	140200	1578708	0.0888	8.9710
THC-COOH	THC-COOH-D9	2.432	97725	415636	0.2351	10.1395
THC	THC-D3	6.199	44720	439788	0.1017	9.5897

ISP FORENSICS - Pocatello Instrument # 59740
Cannabinoids Analysis Report

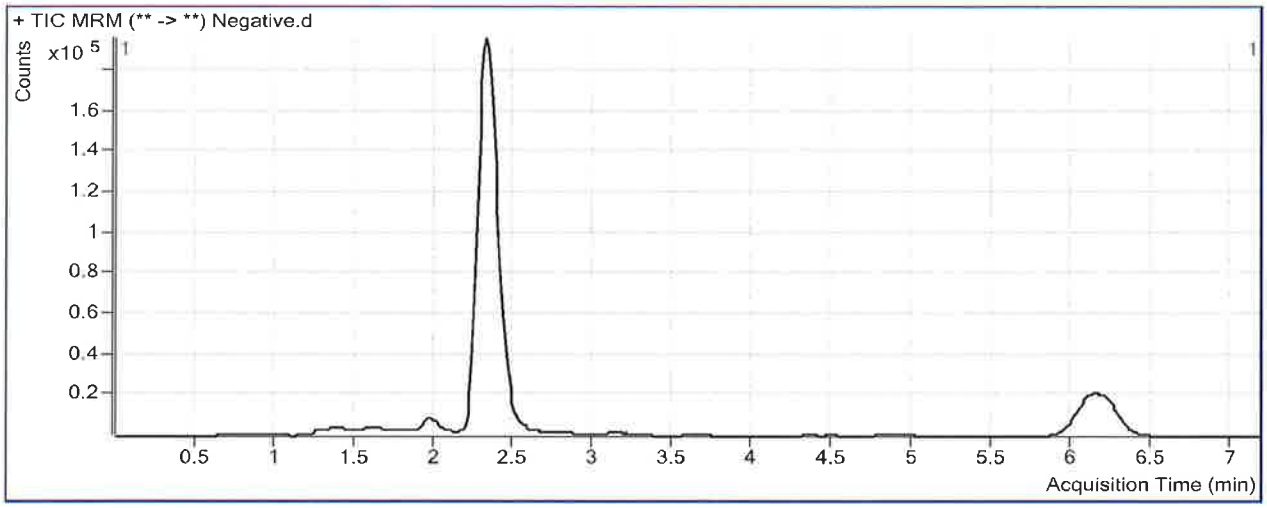
P

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071		
Analysis Time	7/11/2018 7:39 AM	**Analyst Name**	ISPUser
Report Time	7/11/2018 7:41 AM	**Reporter Name**	ISPUser
Last Calib Update	7/11/2018 7:39 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-07-10 13:39	**Data File**	Negative.d
Sample Type	Sample	**Sample Name**	Negative
Dilution	1	**Acq Method**	THC Quant 051517 workingmm.m
Position	P1-H5	**Sample Info**	
Inj Vol	-1	**Comment**	Hemostat 361331-1

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-COOH	THC-COOH-D9	2.419	21044	362106	0.0581	2.4494 <10 P

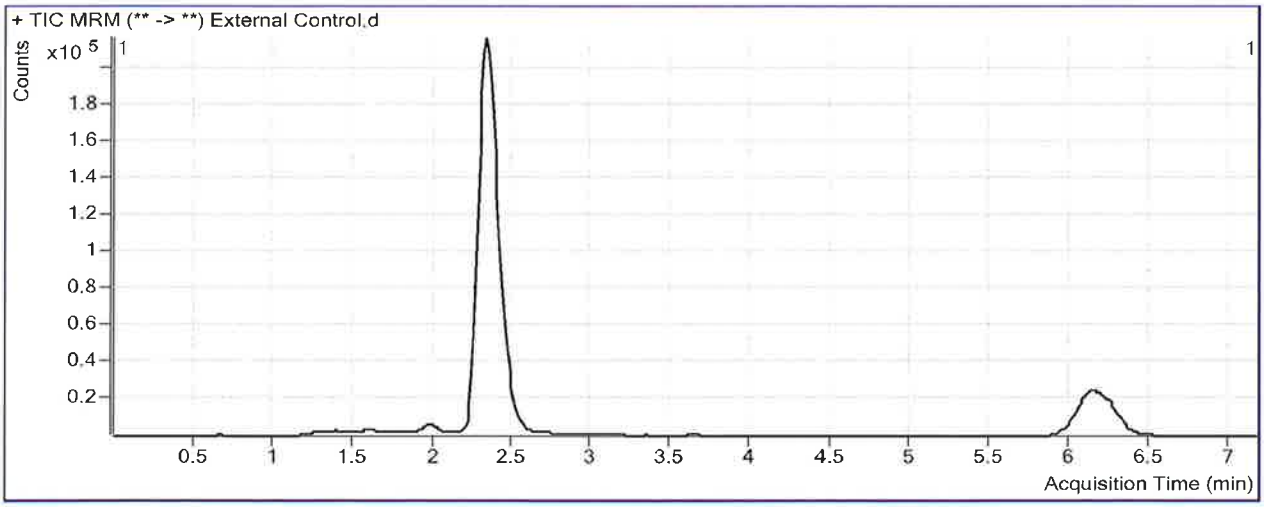
ISP FORENSICS - Pocatello Instrument # 59740
Cannabinoids Analysis Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071		
Analysis Time	7/11/2018 7:48 AM	**Analyst Name**	ISPUser
Report Time	7/11/2018 7:49 AM	**Reporter Name**	ISPUser
Last Calib Update	7/11/2018 7:48 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-07-10 14:03	**Data File**	External Control.d
Sample Type	Sample	**Sample Name**	External Control
Dilution	1	**Acq Method**	THC Quant 051517 workingmm.m
Position	P1-G5	**Sample Info**	
Inj Vol	-1	**Comment**	Hemostat 361331-1 + WS 020718

Sample Chromatogram



Results

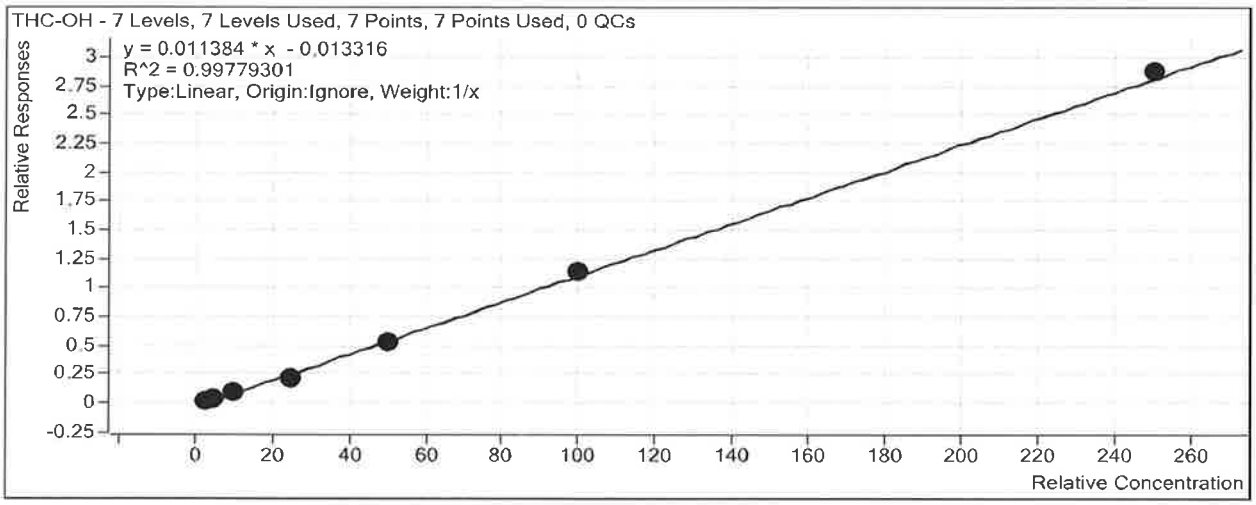
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.332	110760	1330180	0.0833	8.4843
THC-COOH	THC-COOH-D9	2.432	81199	359728	0.2257	9.7312
THC	THC-D3	6.199	34786	359985	0.0966	9.1661

ISP Forensics
Calibration Curve Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071018 THCQ SP.batch.bin
Last Calib Update	7/11/2018 7:39 AM
Analyst Name	ISP TOX

Target Compound *THC-OH*

Internal Standard *THC-OH-D3*



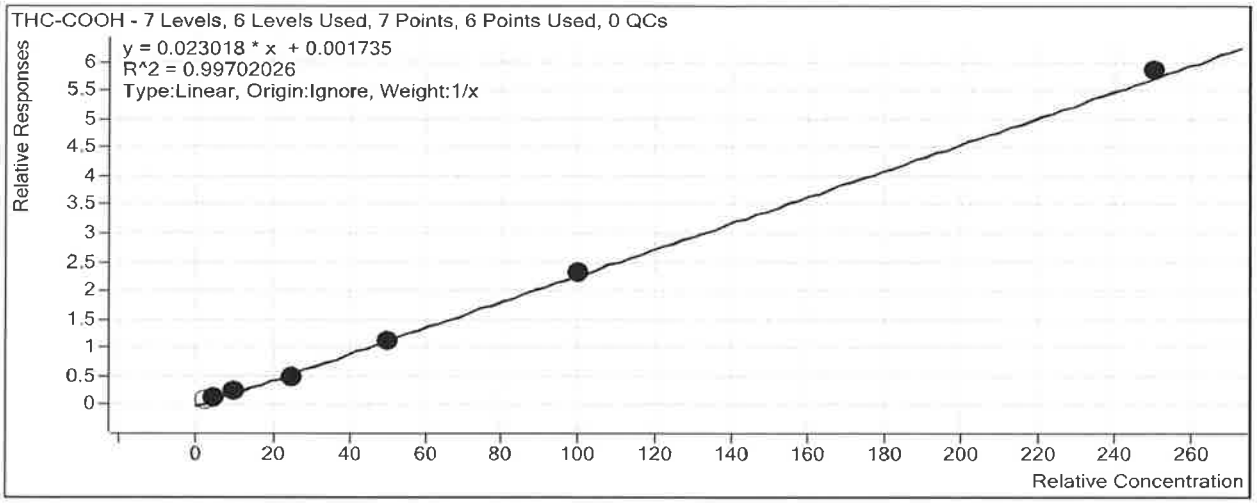
Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1	1	☑	3	3.5	115.7
Cal 2	2	☑	5	5.0	100.4
Cal 3	3	☑	10	10.0	100.1
Cal 4	4	☑	25	21.0	83.9
Cal 5	5	☑	50	48.7	97.3
Cal 6	6	☑	100	101.1	101.1
Cal 7	7	☑	250	253.8	101.5

ISP Forensics
Calibration Curve Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071018 THCQ SP.batch.bin		
Last Calib Update	7/11/2018 7:39 AM	**Analyst Name**	ISP TOX

Target Compound *THC-COOH*

Internal Standard *THC-COOH-D9*



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1	1	☐	3	4.4	147.7
Cal 2	2	☑	5	5.4	107.5
Cal 3	3	☑	10	11.1	111.2
Cal 4	4	☑	25	20.7	82.8
Cal 5	5	☑	50	48.3	96.6
Cal 6	6	☑	100	100.2	100.2
Cal 7	7	☑	250	254.3	101.7

ISP Forensics
Calibration Curve Report

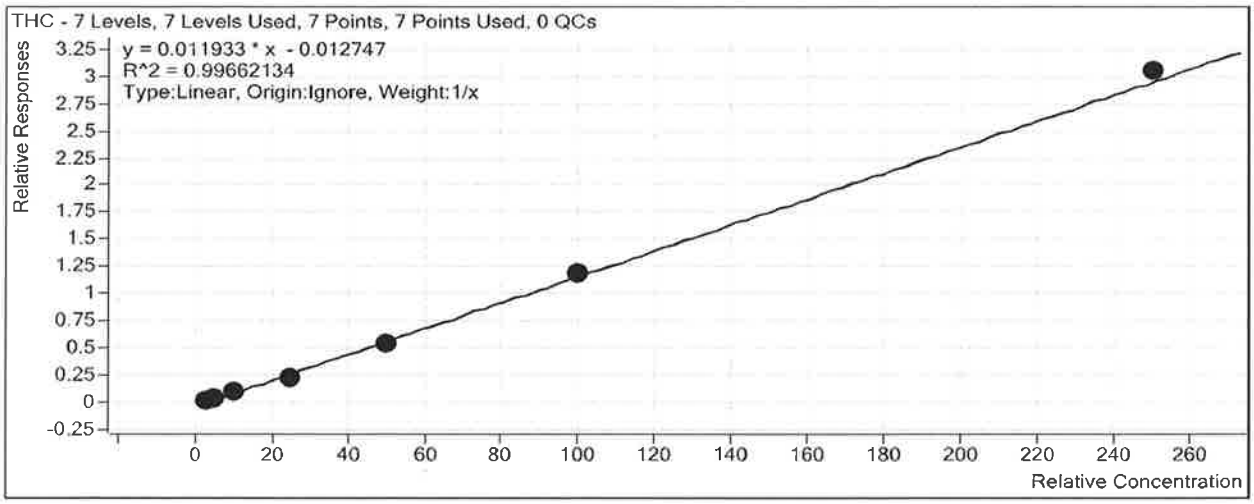
P

Batch Data Path C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071018 THCQ SP.batch.bin

Last Calib Update 7/11/2018 7:39 AM **Analyst Name** ISP TOX

Target Compound *THC*

Internal Standard *THC-D3*



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1	1	☑	3	3.6	120.4
Cal 2	2	☑	5	5.3	105.4
Cal 3	3	☑	10	9.4	94.3
Cal 4	4	☑	25	20.7	82.6
Cal 5	5	☑	50	47.1	94.2
Cal 6	6	☑	100	100.5	100.5
Cal 7	7	☑	250	256.5	102.6

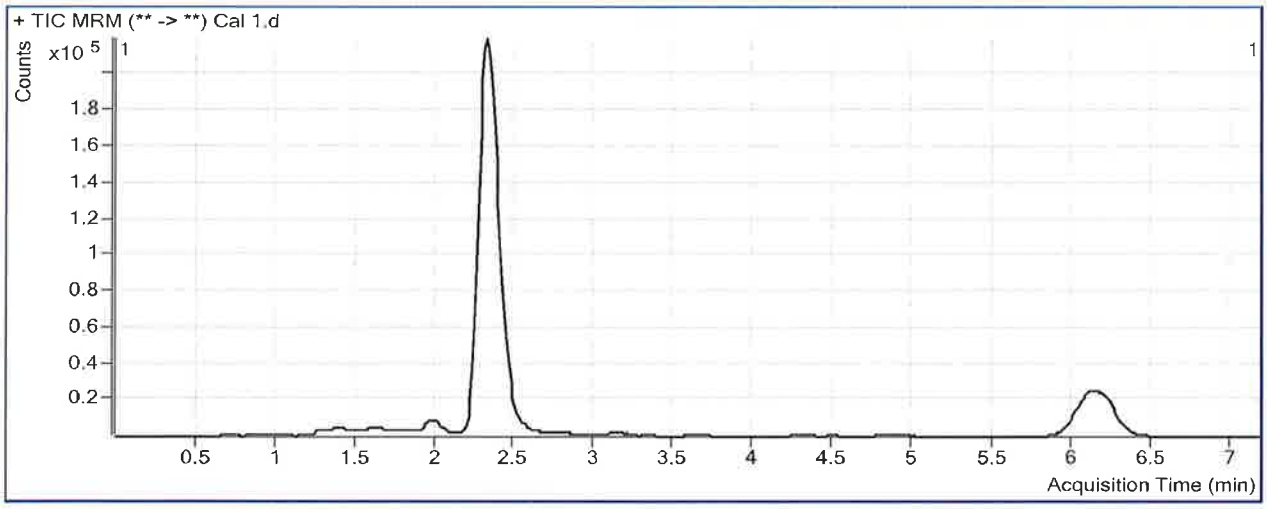
ISP FORENSICS - Pocatello Instrument # 59740
Cannabinoids Analysis Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071		
Analysis Time	7/11/2018 7:39 AM	**Analyst Name**	ISPUser
Report Time	7/11/2018 7:40 AM	**Reporter Name**	ISPUser
Last Calib Update	7/11/2018 7:39 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-07-10 11:41	**Data File**	Cal 1.d
Sample Type	Calibration	**Sample Name**	Cal 1
Dilution	1	**Acq Method**	THC Quant 051517 workingmm.m
Position	P1-H6	**Sample Info**	
Inj Vol	-1	**Comment**	

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.332	38047	1451456	0.0262	3.4724
THC-COOH	THC-COOH-D9	2.432	41312	398396	0.1037	4.4297
THC	THC-D3	6.226	12492	411452	0.0304	3.6125

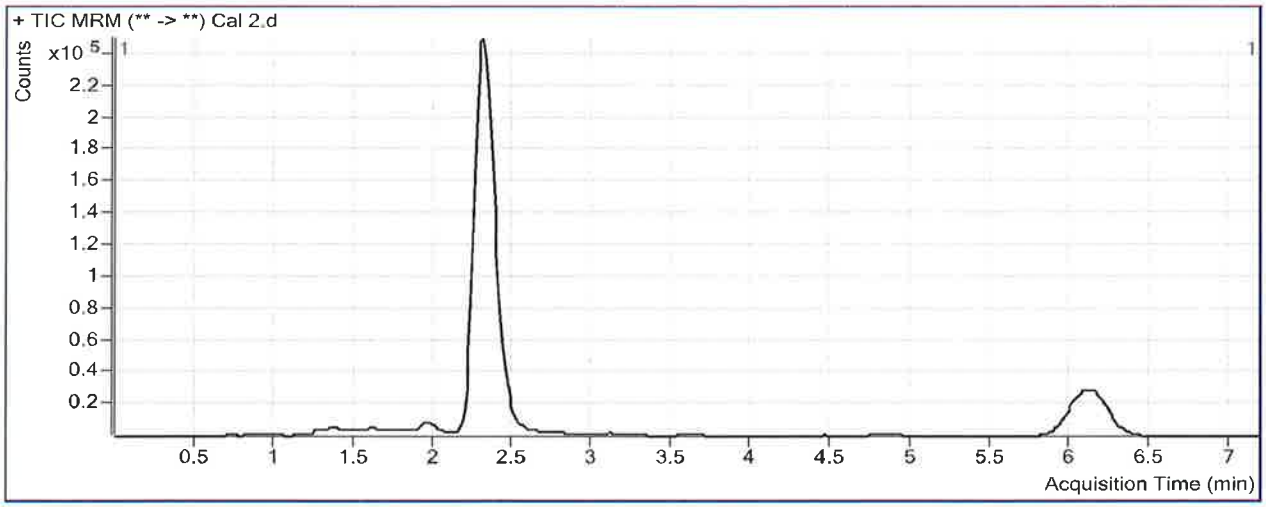
ISP FORENSICS - Pocatello Instrument # 59740
Cannabinoids Analysis Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071		
Analysis Time	7/11/2018 7:39 AM	**Analyst Name**	ISPUser
Report Time	7/11/2018 7:40 AM	**Reporter Name**	ISPUser
Last Calib Update	7/11/2018 7:39 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-07-10 11:53	**Data File**	Cal 2.d
Sample Type	Calibration	**Sample Name**	Cal 2
Dilution	1	**Acq Method**	THC Quant 051517 workingmm.m
Position	P1-G6	**Sample Info**	
Inj Vol	-1	**Comment**	

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.319	71816	1639341	0.0438	5.0180
THC-COOH	THC-COOH-D9	2.419	57248	456278	0.1255	5.3756
THC	THC-D3	6.199	23278	464124	0.0502	5.2714

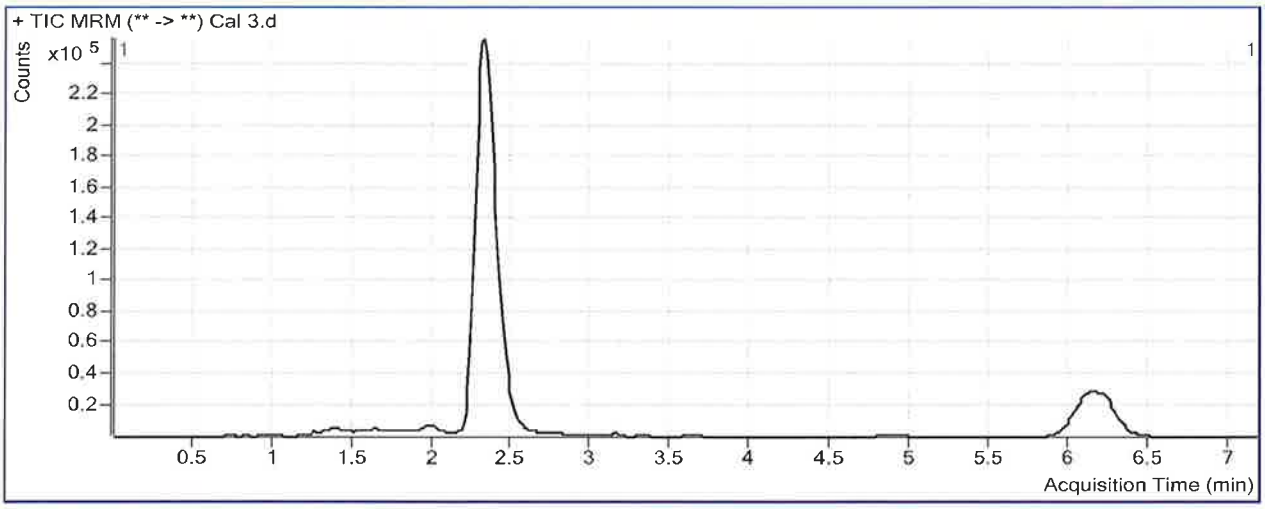
ISP FORENSICS - Pocatello Instrument # 59740
Cannabinoids Analysis Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071		
Analysis Time	7/11/2018 7:39 AM	**Analyst Name**	ISPUser
Report Time	7/11/2018 7:40 AM	**Reporter Name**	ISPUser
Last Calib Update	7/11/2018 7:39 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-07-10 12:05	**Data File**	Cal 3.d
Sample Type	Calibration	**Sample Name**	Cal 3
Dilution	1	**Acq Method**	THC Quant 051517 workingmm.m
Position	P1-F6	**Sample Info**	
Inj Vol	-1	**Comment**	

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.332	160903	1599698	0.1006	10.0055
THC-COOH	THC-COOH-D9	2.432	107707	417988	0.2577	11.1196
THC	THC-D3	6.212	43957	440528	0.0998	9.4303

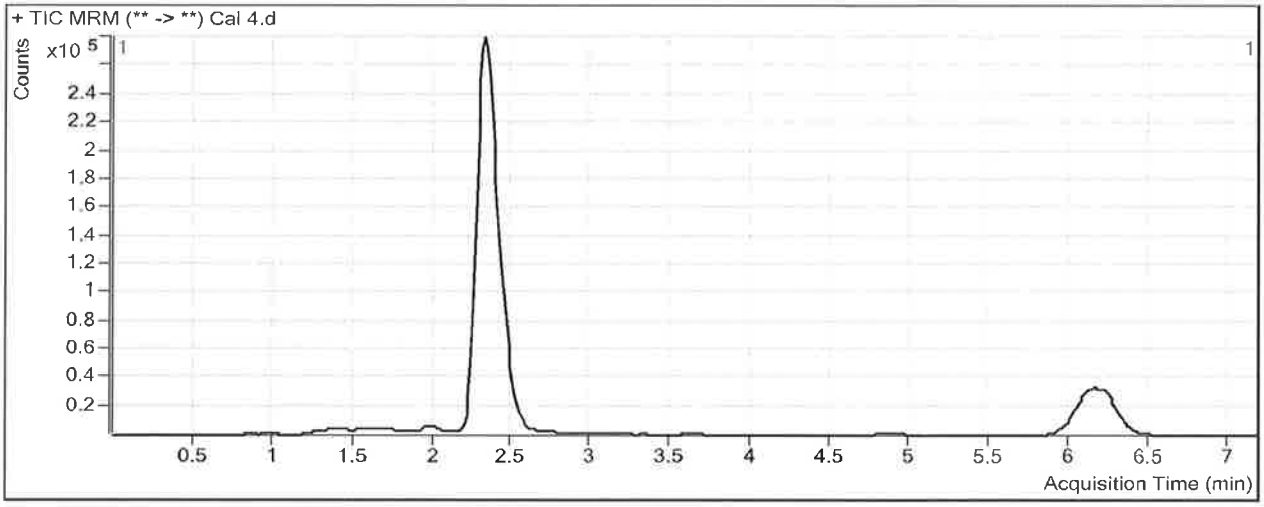
ISP FORENSICS - Pocatello Instrument # 59740
Cannabinoids Analysis Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071		
Analysis Time	7/11/2018 7:39 AM	**Analyst Name**	ISPUser
Report Time	7/11/2018 7:40 AM	**Reporter Name**	ISPUser
Last Calib Update	7/11/2018 7:39 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-07-10 12:16	**Data File**	Cal 4.d
Sample Type	Calibration	**Sample Name**	Cal 4
Dilution	1	**Acq Method**	THC Quant 051517 workingmm.m
Position	P1-E6	**Sample Info**	
Inj Vol	-1	**Comment**	

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.332	351070	1557201	0.2254	20.9744
THC-COOH	THC-COOH-D9	2.432	204411	427492	0.4782	20.6985
THC	THC-D3	6.212	99419	425390	0.2337	20.6540

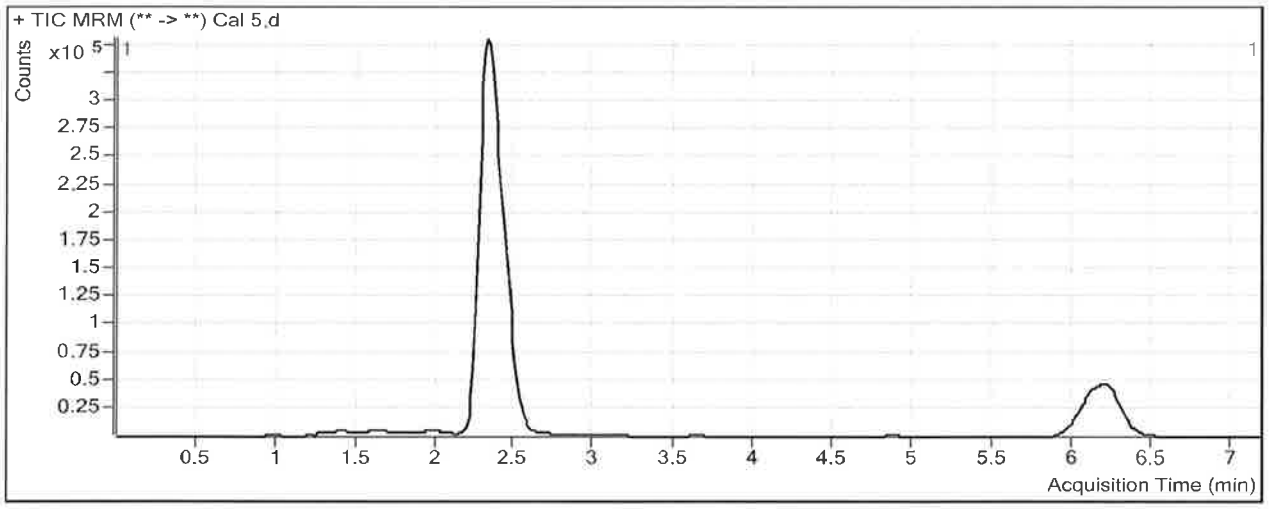
ISP FORENSICS - Pocatello Instrument # 59740
Cannabinoids Analysis Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071		
Analysis Time	7/11/2018 7:39 AM	**Analyst Name**	ISPUser
Report Time	7/11/2018 7:40 AM	**Reporter Name**	ISPUser
Last Calib Update	7/11/2018 7:39 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-07-10 12:28	**Data File**	Cal 5.d
Sample Type	Calibration	**Sample Name**	Cal 5
Dilution	1	**Acq Method**	THC Quant 051517 workingmm.m
Position	P1-D6	**Sample Info**	
Inj Vol	-1	**Comment**	

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.332	857242	1585268	0.5408	48.6725
THC-COOH	THC-COOH-D9	2.432	470175	422265	1.1135	48.2990
THC	THC-D3	6.212	239484	436082	0.5492	47.0906

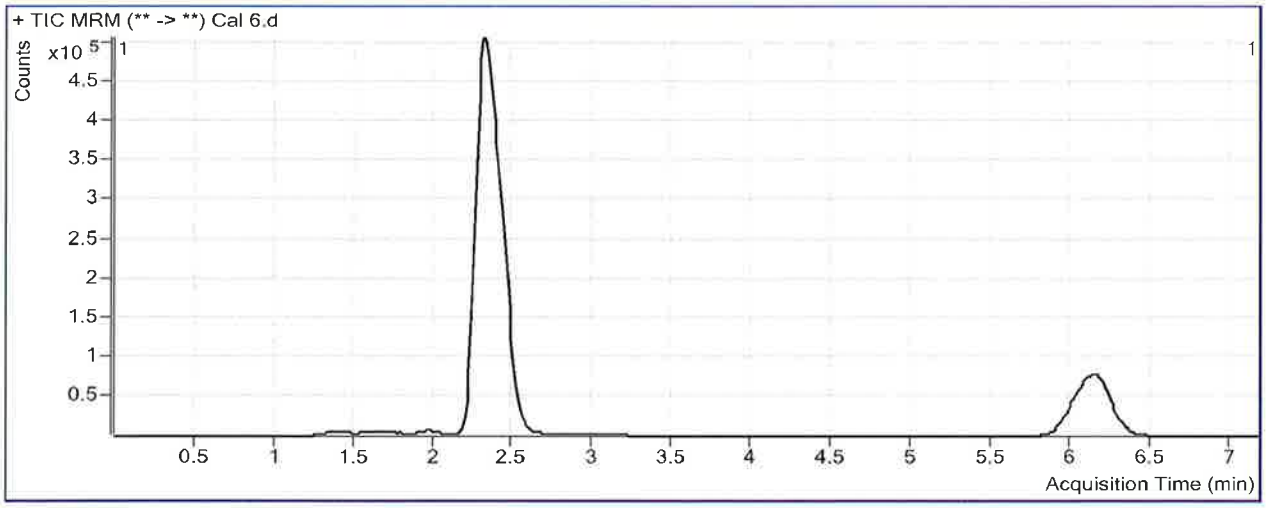
ISP FORENSICS - Pocatello Instrument # 59740
Cannabinoids Analysis Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071		
Analysis Time	7/11/2018 7:39 AM	**Analyst Name**	ISPUser
Report Time	7/11/2018 7:40 AM	**Reporter Name**	ISPUser
Last Calib Update	7/11/2018 7:39 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-07-10 12:40	**Data File**	Cal 6.d
Sample Type	Calibration	**Sample Name**	Cal 6
Dilution	1	**Acq Method**	THC Quant 051517 workingmm.m
Position	P1-C6	**Sample Info**	
Inj Vol	-1	**Comment**	

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.319	1836042	1614264	1.1374	101.0838
THC-COOH	THC-COOH-D9	2.419	973235	421823	2.3072	100.1617
THC	THC-D3	6.159	544539	459111	1.1861	100.4646

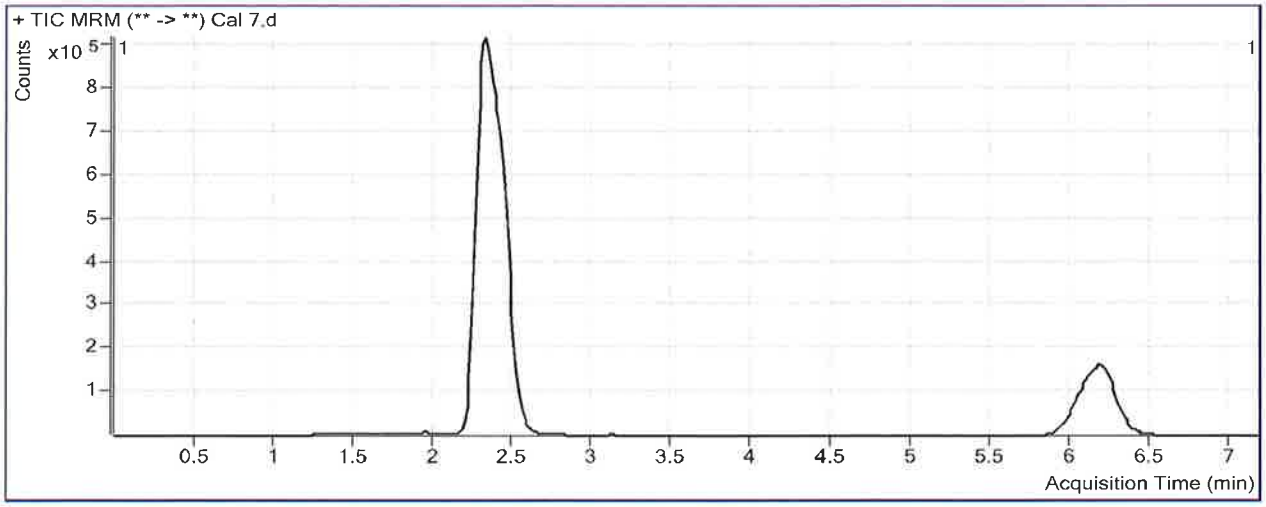
ISP FORENSICS - Pocatello Instrument # 59740
Cannabinoids Analysis Report

Batch Data Path	C:\MassHunter\Data\2018\THC Quant\071018 THC Quant Urine Validation SP worklist 2555\QuantResults\071		
Analysis Time	7/11/2018 7:39 AM	**Analyst Name**	ISPUser
Report Time	7/11/2018 7:41 AM	**Reporter Name**	ISPUser
Last Calib Update	7/11/2018 7:39 AM	**Batch State**	Processed

Analysis Info

Acq Time	2018-07-10 12:52	**Data File**	Cal 7.d
Sample Type	Calibration	**Sample Name**	Cal 7
Dilution	1	**Acq Method**	THC Quant 051517 workingmm.m
Position	P1-B6	**Sample Info**	
Inj Vol	-1	**Comment**	

Sample Chromatogram



Results

Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.319	4596310	1598412	2.8755	253.7733
THC-COOH	THC-COOH-D9	2.419	2343351	400153	5.8561	254.3455
THC	THC-D3	6.186	1399097	459064	3.0477	256.4766