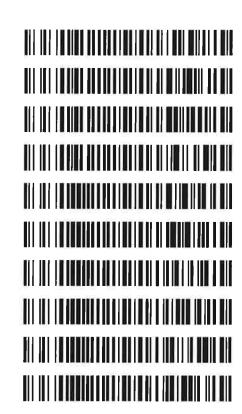
Worklist: 2173

LAB CASE	<u>ITEM</u>	TASK ID	DESCRIPTION
M2017-5822	1	106942	AM 27 Blood THC Quant by LC
M2017-5967	3	106943	AM 27 Blood THC Quant by LC
M2018-0147	1	106944	AM 27 Blood THC Quant by LC
M2018-0204	1	106945	AM 27 Blood THC Quant by LC
P2018-0218	1	106946	AM 27 Blood THC Quant by LC
P2018-0224	1	106947	AM 27 Blood THC Quant by LC
P2018-0251	1	106948	AM 27 Blood THC Quant by LC
P2018-0262	1	106949	AM 27 Blood THC Quant by LC
P2018-0265	1	106950	AM 27 Blood THC Quant by LC
P2018-0272	1	106951	AM 27 Blood THC Quant by LC





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

Extraction Date: 02/07/18

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: 361331-3

Column: UCT Selectra DA 100 x 2.1mm 3um

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:

Analytic:

- ☑ 1. Remove standards, plate, controls, and samples from cold storage. Allow to reach room temperature.
- Q 2. Pipette 1000μL blood (calibrated pipette) Pipette ID: #27 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.
- 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 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
- **1** 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

Create batch and process data.

Worklist path:020 718 THC Quant 59 75 Batch Name: 020718 SP THE Quant

- Make any necessary integration changes, r^2 values ≥ 0.98 for each analyte.
 - Did all QCs pass for each analyte? Y/N Enter QCs into control charting?
- Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS:

Curve Range Limited: Carboxy-THC 10-250 due to low ratio.



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, 100ug/ml 100ul THC-OH, C-THC in 9790 ul MeOH (Fisher Lot # 172516)

Prepared: 02/07/18 Expires: 02/07/19 By:TS

Drug C-THC H ō 497429 Expiration N/A

FE04231406 FE01121503 1/31/2020 4/30/2019

THC-OH

Prepared: 02/07/18

AM 27 control 100 ul working solution lot (WS020718) in 9900ul blood (Hemostat Lot# 361331-2)

Expires: 02/07/19





Batch Data Path C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP THC Quant.batch.bi

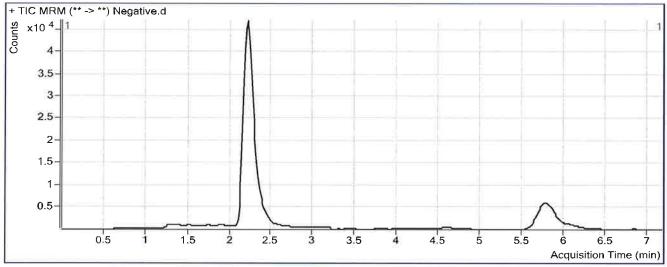
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Analysis Info

Acq Time2018-02-07 14:01Data FileNegative.dSample TypeSampleSample NameNegativeDilution1Acq MethodTHC Quant 051517 workingmm.m

Position P1-A7 Sample Info

Inj Vol -1 Comment Hemostat 361331-3



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.399	2517	340586	0.0074	0.3397 〈 ろ
THC-COOH	THC-COOH-D9	2.299	3417	90577	0.0377	0.3130<10



Batch Data Path C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP THC Quant.batch.bi

Analysis Time2/8/2018 8:27 AMAnalyst NameISPUserReport Time2/8/2018 8:35 AMReporter NameISPUserLast Calib Update2/8/2018 8:27 AMBatch StateProcessed

Analysis Info

 Acq Time
 2018-02-07 14:24
 Data File
 External Control.d

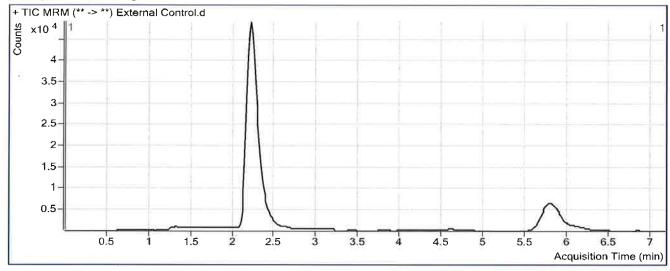
 Sample Type
 Sample
 Sample Name
 External Control

Dilution 1 **Acq Method** THC Quant 051517 workingmm.m

Position P1-B7 **Sample Info**

Inj Vol -1 **Comment** Hemostat 361331-3 + WS 020718

Sample Chromatogram



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.212	31778	327341	0.0971	8.0607
THC-COOH	THC-COOH-D9	2.312	19818	86908	0.2280	8.8384
THC	THC-D3	5.812	10866	100047	0.1086	9.6471

Printed at: 8:35 AM on: 2/8/2018



Batch Data Path C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP THC Quant.batch.bi

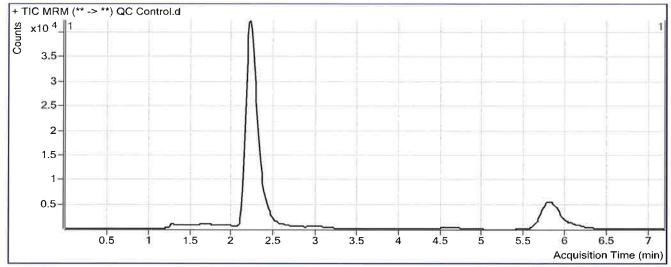
Analysis Time2/8/2018 8:27 AMAnalyst NameISPUserReport Time2/8/2018 8:35 AMReporter NameISPUserLast Calib Update2/8/2018 8:27 AMBatch StateProcessed

Analysis Info

Acq Time2018-02-07 13:37Data FileQC Control.dSample TypeSampleSample NameQC Control

Dilution 1 **Acq Method** THC Quant 051517 workingmm.m

Position P1-H6 Sample Info
Inj Vol -1 Comment



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.225	33719	274840	0.1227	10.2650
THC-COOH	THC-COOH-D9	2.312	19798	67142	0.2949	11.8326
THC	THC-D3	5.839	9586	84283	0.1137	10.1282

15P Forensics Calibration Curve Report

Batch Data Path

C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP

THC Quant.batch.bin

Last Calib Update

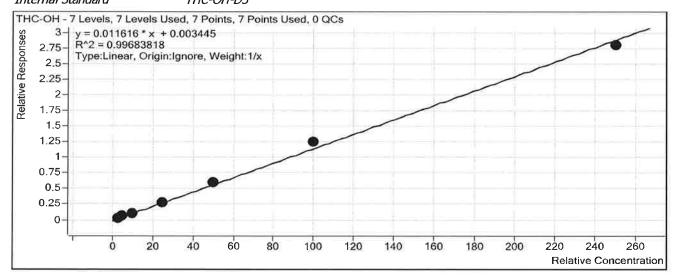
2/8/2018 8:27 AM

Analyst Name

ISP TOX

Target Compound
Internal Standard

THC-OH THC-OH-D3



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1	1		3	2.9	96.9
Cal 2	2	✓	5	5.2	103.3
Cal 3	3	\square	10	9.5	94.7
Cal 4	4		25	24.1	96.3
Cal 5	5	Ø	50	52.0	104.0
Cal 6	6	☑	100	108.4	108.4
Cal 7	7		250	241.0	96.4



Calibration Curve Report

Batch Data Path

C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP

THC Quant.batch.bin

Last Calib Update

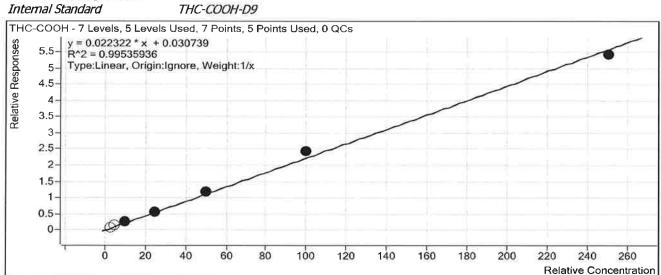
2/8/2018 8:27 AM

Analyst Name

ISP TOX

Target Compound
Internal Standard

THC-COOH



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1	1		3	3.0	101.7
Cal 2	2		5	4.9	97.2
Cal 3	3	\square	10	9.8	97.9
Cal 4	4	\square	25	23.1	92.3
Cal 5	5	\square	50	52.3	104.6
Cal 6	6		100	108.8	108.8
Cal 7	7		250	241.0	96.4

international view near a sea



15P Forensics Calibration Curve Report

Batch Data Path

C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP

THC Quant.batch.bin

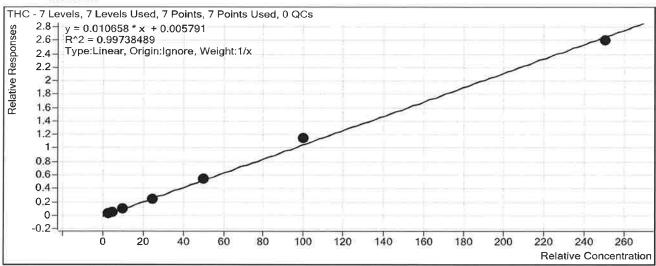
Last Calib Update

2/8/2018 8:27 AM

Analyst Name

ISP TOX

Target CompoundTHCInternal StandardTHC-D3



Sample	Level	Enabled	Exp Conc	Final Conc	Accuracy
Cal 1	1	☑	3	3.1	104.6
Cal 2	2	☑	5	4.9	98.4
Cal 3	3		10	9.8	97.9
Cal 4	4	☑	25	22.9	91.5
Cal 5	5		50	51.4	102.7
Cal 6	6	\square	100	107.6	107.6
Cal 7	7	✓	250	243.4	97.3

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Printed at: 8:34 AM on: 2/8/2018

Batch Data Path C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP THC Quant.batch.bi

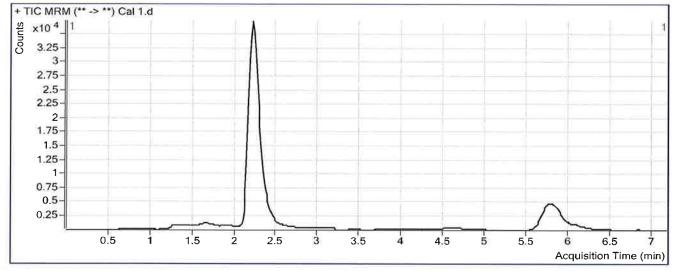
Analysis Time2/8/2018 8:27 AMAnalyst NameISPUserReport Time2/8/2018 8:34 AMReporter NameISPUserLast Calib Update2/8/2018 8:27 AMBatch StateProcessed

Analysis Info

Acq Time2018-02-07 12:02Data FileCal 1.dSample TypeCalibrationSample NameCal 1

Dilution 1 **Acq Method** THC Quant 051517 workingmm.m

Position P1-A6 Sample Info
Inj Vol -1 Comment



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.225	9826	264079	0.0372	2.9067
THC-COOH	THC-COOH-D9	2.312	7149	72352	0.0988	3.0497
THC	THC-D3	5.825	3259	83085	0.0392	3.1370



Batch Data Path C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP THC Quant.batch.bi

Analysis Time2/8/2018 8:27 AMAnalyst NameISPUserReport Time2/8/2018 8:34 AMReporter NameISPUserLast Calib Update2/8/2018 8:27 AMBatch StateProcessed

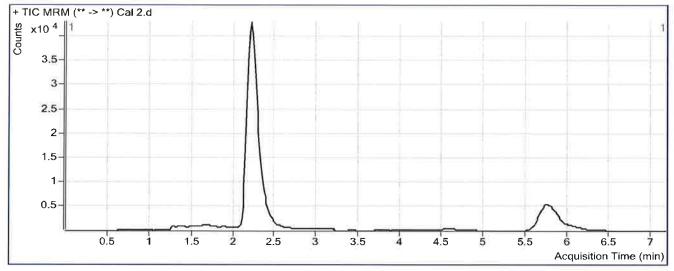
Analysis Info

Acq Time2018-02-07 12:14Data FileCal 2.dSample TypeCalibrationSample NameCal 2

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	2.212	18485	291439	0.0634	5.1636
THC-COOH	THC-COOH-D9	2.312	11415	81990	0.1392	4.8600
THC	THC-D3	5.812	5190	89129	0.0582	4.9199

Printed at: 8:34 AM on: 2/8/2018



Batch Data Path C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP THC Quant.batch.bi

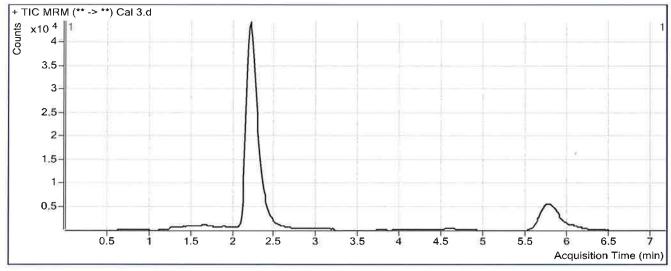
Analysis Time2/8/2018 8:27 AMAnalyst NameISPUserReport Time2/8/2018 8:34 AMReporter NameISPUserLast Calib Update2/8/2018 8:27 AMBatch StateProcessed

Analysis Info

Acq Time2018-02-07 12:26Data FileCal 3.dSample TypeCalibrationSample NameCal 3

Dilution 1 **Acq Method** THC Quant 051517 workingmm.m

Position P1-C6 Sample Info
Inj Vol -1 Comment



Results						
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc
THC-OH	THC-OH-D3	2.212	32675	287927	0.1135	9.4729
THC-COOH	THC-COOH-D9	2.299	19302	77431	0.2493	9.7902
THC	THC-D3	5.799	9610	87226	0.1102	9.7933



Batch Data Path C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP THC Quant.batch.bi

Analysis Time2/8/2018 8:27 AMAnalyst NameISPUserReport Time2/8/2018 8:34 AMReporter NameISPUserLast Calib Update2/8/2018 8:27 AMBatch StateProcessed

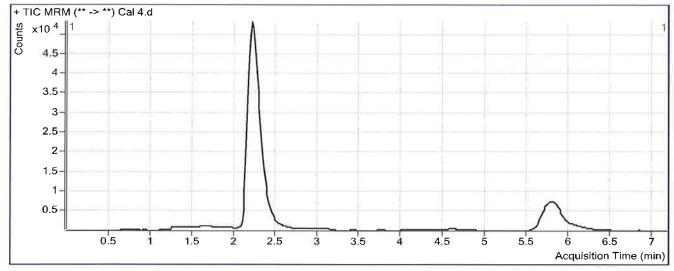
Analysis Info

Acq Time2018-02-07 12:38Data FileCal 4.dSample TypeCalibrationSample NameCal 4

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	2.212	84258	297635	0.2831	24.0738
THC-COOH	THC-COOH-D9	2.312	45220	82863	0.5457	23.0706
THC	THC-D3	5.839	232 44	93152	0.2495	22.8689

Printed at: 8:34 AM on: 2/8/2018



Batch Data Path C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP THC Quant.batch.bi

Analysis Time2/8/2018 8:27 AMAnalyst NameISPUserReport Time2/8/2018 8:34 AMReporter NameISPUserLast Calib Update2/8/2018 8:27 AMBatch StateProcessed

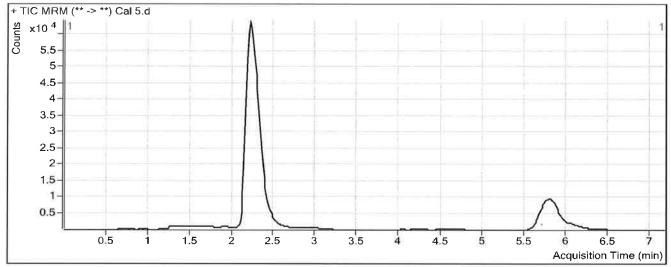
Analysis Info

Acq Time2018-02-07 12:50Data FileCal 5.dSample TypeCalibrationSample NameCal 5

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	2.212	170802	281125	0.6076	52.0069
THC-COOH	THC-COOH-D9	2.312	89703	74876	1.1980	52.2930
THC	THC-D3	5.825	47107	85156	0.5532	51.3582

Samples Report Cannabinoids1_004_Cal 5.xlsx Printed at: 8:34 AM on: 2/8/2018



Batch Data Path C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP THC Quant.batch.bi

Analysis Time2/8/2018 8:27 AMAnalyst NameISPUserReport Time2/8/2018 8:34 AMReporter NameISPUserLast Calib Update2/8/2018 8:27 AMBatch StateProcessed

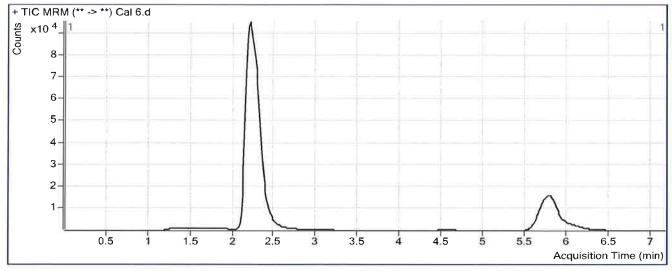
Analysis Info

Acq Time2018-02-07 13:01Data FileCal 6.dSample TypeCalibrationSample NameCal 6

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	2.199	369099	292289	1.2628	108.4132		
THC-COOH	THC-COOH-D9	2.299	188046	76447	2.4598	108.8189		
THC	THC-D3	5.799	104175	90407	1.1523	107.5687		

Printed at: 8:35 AM on: 2/8/2018



Printed at: 8:35 AM on: 2/8/2018

Batch Data Path C:\MassHunter\Data\2018\THC Quant\020718 THC Quant SP TS\QuantResults\020718 SP THC Quant.batch.bi

Analysis Time2/8/2018 8:27 AMAnalyst NameISPUserReport Time2/8/2018 8:35 AMReporter NameISPUserLast Calib Update2/8/2018 8:27 AMBatch StateProcessed

Analysis Info

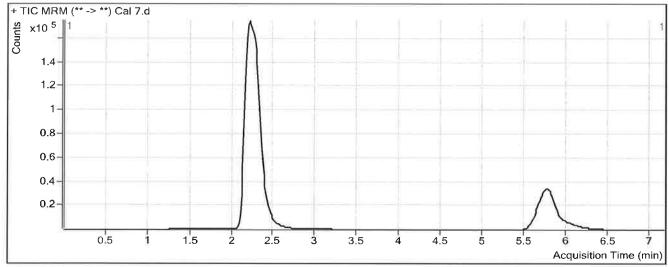
 Acq Time
 2018-02-07 13:13
 Data File
 Cal 7.d

 Sample Type
 Calibration
 Sample Name
 Cal 7

Dilution 1 **Acq Method** THC Quant 051517 workingmm.m

 Position
 P1-G6
 Sample Info

 Inj Vol
 -1
 Comment



Results								
Compound	ISTD Compound	RT	Response	ISTD Resp	Resp Ratio	Final Conc		
THC-OH	THC-OH-D3	2.199	910053	324728	2.8025	240.9629		
THC-COOH	THC-COOH-D9	2.299	442935	81859	5.4110	241.0274		
THC	THC-D3	5.772	263720	101449	2.5995	243.3540		