5/6/2021

REVIEWED
By Britany Wylie at 12:47 pm, May 10, 2021

Worklist: 4955

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION	
C2021-0896	1	UCK	AM 3 Urine Carboxy-THC	
C2021-0939	1	UCK	AM 3 Urine Carboxy-THC	
C2021-0987	1	UCK	AM 3 Urine Carboxy-THC	
C2021-1019	1	UCK	AM 3 Urine Carboxy-THC	

AM 3: Carboxy-THC Urine Extraction

Extraction Date: 5/6/21 Analyst: Anne Nord

Negative Urine Lot: 2121 **GC/MS ID**: 65198
Hexane Lot: fisher 42712 **Ethyl Acetate Lot:** fisher 020419
Silylating Agent Lot: Cerilliant FN08181601

Pre-Analytic:

☒ 1. *Positive Control Working Solution Preparation Instructions:*
Add 180uL of 100ug/mL 11-nor-9-carboxy-Δ9-THC Stock Solution to 9.82mL Methanol. Other volumes may be prepared. Solution is stable for 1-year or the expiration of the stock reference material (whichever is sooner). Store under refrigeration

☒ 2. Verify Tune and Tune evaluation completed within the previous 7 days. Tune and Tune evaluation reports initialed and filed.

☒ 3. Create GCMS sequence to include controls, case blanks and case samples.

Analytic:

☒ 1. Remove working solutions, external control, negative urine and case samples from cold storage.

☒ 2. To each labeled round bottomed tube add 3mL sample, using negative urine sample for both negative and positive control. Positive control:. added working solution to equal approximately 60 ng/ml Carboxy-THC

☒ 3. Add 500uL 1N KOH to all tubes. Check pH. *(If pH <12, add additional 500ul 1N KOH). (Note: put a mark on the tube or separate the tubes that have a pH<12 as you will need to know this in step 5).*

☒ 4. Place tubes in 40C water bath for ~15 min. Remove and allow to cool.

☒ 5. If original pH was >12, add 1.5mL pH 1.8 Saturated Phosphate Buffer and 3mL Hexane/Ethyl Acetate *(87:13)*
If original pH was <12, add 3mL pH 1.8 Saturated Phosphate Buffer and 4mL Hexane/Ethyl Acetate *(87:13)*

☒ 6. Rock at ambient temp for ~ 10 minutes.

☒ 7. Centrifuge for ~ 10 min at ~3500rpm.

☒ 8. Transfer solvent to tapered bottom tube and evaporate to dryness under nitrogen @ 37C.

☒ 9. Add 50uL Ethyl Acetate and 50uL silylating reagent, cap and vortex. Heat @ 95C for 15min, then allow to cool.

☒ 10. Transfer sample to labeled ALS vial with insert.

☒ 11. Place ALS Vials in appropriate location on GCMS rack and run using appropriate GCMS method.

Post-Analytic

☒ 1. Complete Data analysis on all samples and corresponding sample blanks

☒ 2. Did positive and negative control samples provide intended response? yes

☒ 3. **Criteria for ID**: RT +/1 0.1 min., Ion Ratio of 347:473 & 371:488 within +/- 20%.
Sample response greater than Min Corrected Area, Diluted samples

☒ 4. Central File Packet to include: LIMS Worklist, Method Checklist, Working solution prep sheet, and Control sample GCMS data printouts

Comments:

Toxicology AM 3 Method 3 spiked 60 ng/ml

Stock solution 3.6 ul (100 ug/ml) C-THC in 5.996 mls neg urine lot 2121

Ppd 5/6/21 expires 11/6/21 lot 2621

Drug	Lot	expiration
C-THC	FE01061702	3/1/2022

Toxicology AM 3 Commercial control

Pinpoint THC quantitation plate (IDP-108-2) lot 201206 exp 06/06/21

Added 3 mls negative urine (2121) to Cal 5 (approximate concentration 25 ng/ml THC-COOH)

Run Date:	5/6/2021
Worklist #:	4955
Laboratory Case #:	Positive control PinPoint

Case Calculations:

EIC Ion 371 Corr. Area:	*1027094*
EIC Ion 473 Corr. Area:	*486085*
EIC Ion 488 Corr. Area:	*289348*

473:371 Ratio: **0.473**
488:371 Ratio: **0.282**

Quality Control Calculations:

	1st Control	2nd Control
EIC Ion 371 Corr. Area:	*1313508*	*1524670*
EIC Ion 473 Corr. Area:	*612754*	*716147*
EIC Ion 488 Corr. Area:	*388564*	*452457*
Ratio of 473:371:	*0.467*	*0.470*
		Average: *0.468*
Ratio of 488:371:	*0.296*	*0.297*
		Average: *0.296*

Acceptable Retention Time Range:	*4.404*	to	*4.594*
Approximate Minimum Corrected Area of 371 Ion:			*262702*
Acceptable 473:371 Ratio Range:	*0.374*	to	*0.562*
Acceptable 488:371 Ratio Range:	*0.237*	to	*0.356*

Area Percent Report

Data Path : D:\DATA\2021\am 2\050621\
Data File : 01401012.D
Acq On : 06 May 2021 18:03
Operator : Instrument 65198
Sample : C-THC control pinpoint
Misc : am 3
ALS Vial : 14 Sample Multiplier: 1

Integration Parameters: autoint1.e
Integrator: ChemStation

Method : D:\MassHunter\GCMS\1\methods\TOXI-A 10115.M
Title :

Signal : EIC Ion 371.00 (370.70 to 371.70): 01401012.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total
1	4.241	3	11	35	BB 2	3469	60280	5.87%	5.544%
2	4.493	51	58	81	VB	60883	1027094	100.00%	94.456%

Sum of corrected areas: 1087374

Signal : EIC Ion 473.00 (472.70 to 473.70): 01401012.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total
1	4.495	36	58	81	BB	28422	486085	100.00%	84.480%
2	4.931	128	139	154	BB	3832	89302	18.37%	15.520%

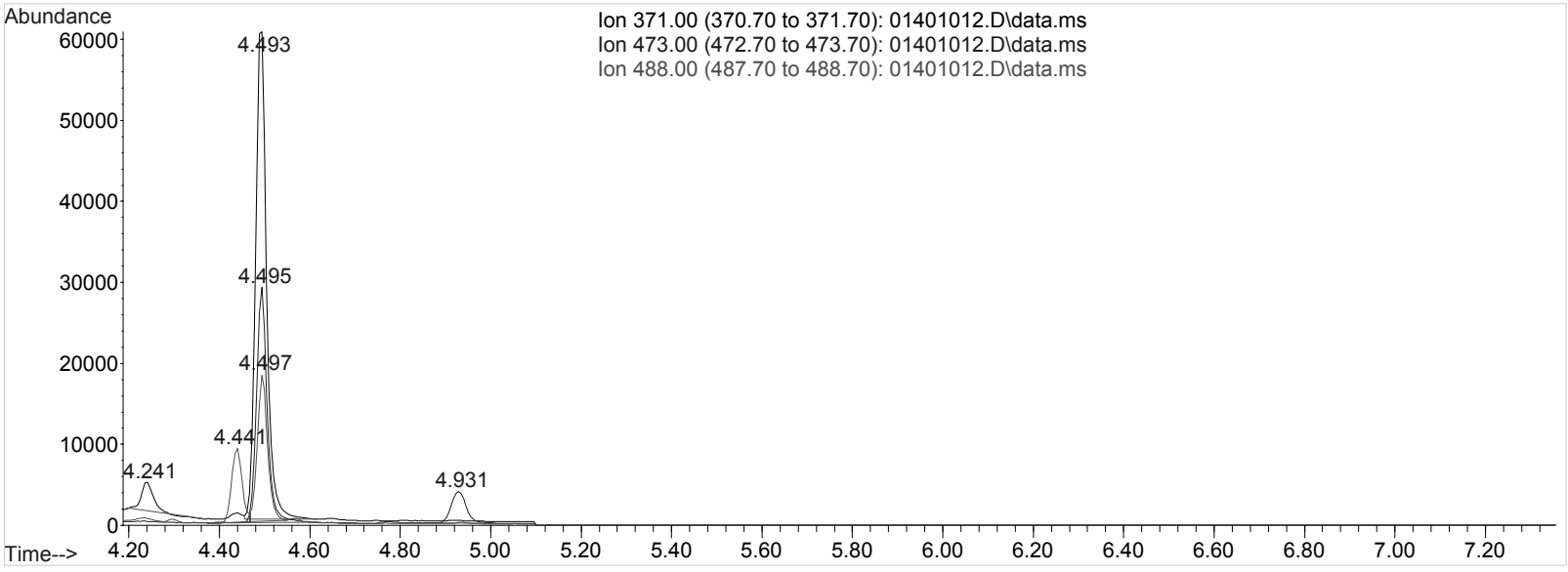
Sum of corrected areas: 575387

Signal : EIC Ion 488.00 (487.70 to 488.70): 01401012.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total
1	4.441	36	48	53	BV	8943	143464	49.58%	33.147%
2	4.497	53	59	69	VV	17683	289348	100.00%	66.853%

Sum of corrected areas: 432812

TOXI-A 10115.M Mon May 10 10:09:55 2021



Area Percent Report

Data Path : D:\DATA\2021\am 2\050621\
Data File : 00801002.D
Acq On : 06 May 2021 16:29
Operator : Instrument 65198
Sample : 60ng C-THC
Misc : am3
ALS Vial : 8 Sample Multiplier: 1

Integration Parameters: autoint1.e
Integrator: ChemStation

Method : D:\MassHunter\GCMS\1\methods\TOXI-A 10115.M
Title :

Signal : EIC Ion 371.00 (370.70 to 371.70): 00801002.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total
1	4.504	41	60	83	BB	73156	1313508	100.00%	100.000%

Sum of corrected areas: 1313508

Signal : EIC Ion 473.00 (472.70 to 473.70): 00801002.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total
1	4.506	50	60	84	BB	33965	612754	100.00%	100.000%

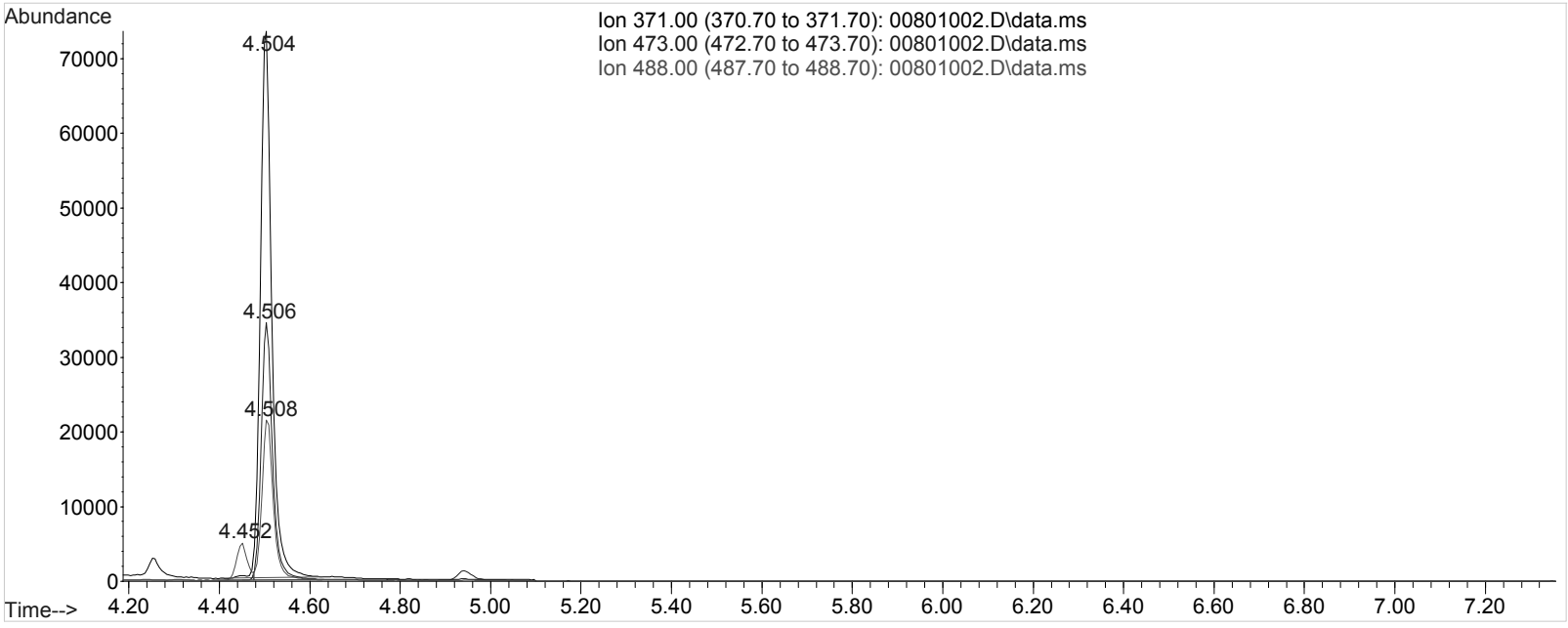
Sum of corrected areas: 612754

Signal : EIC Ion 488.00 (487.70 to 488.70): 00801002.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total
1	4.452	40	50	55	BV	4911	82258	21.17%	17.471%
2	4.508	55	61	83	VB	21360	388564	100.00%	82.529%

Sum of corrected areas: 470823

TOXI-A 10115.M Mon May 10 10:02:39 2021



Area Percent Report

Data Path : D:\DATA\2021\am 2\050621\
Data File : 00801015.D
Acq On : 06 May 2021 18:31
Operator : Instrument 65198
Sample : 60 ng CTHC control
Misc : am 3 end of run control
ALS Vial : 8 Sample Multiplier: 1

Integration Parameters: autoint1.e
Integrator: ChemStation

Method : D:\MassHunter\GCMS\1\methods\TOXI-A 10115.M
Title :

Signal : EIC Ion 371.00 (370.70 to 371.70): 00801015.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total
1	4.242	3	11	35	BB	2953	55156	3.62%	3.491%
2	4.494	51	58	92	VB	90164	1524670	100.00%	96.509%

Sum of corrected areas: 1579826

Signal : EIC Ion 473.00 (472.70 to 473.70): 00801015.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total
1	4.495	49	58	80	BB	42372	716147	100.00%	92.398%
2	4.932	126	139	153	BB	2532	58923	8.23%	7.602%

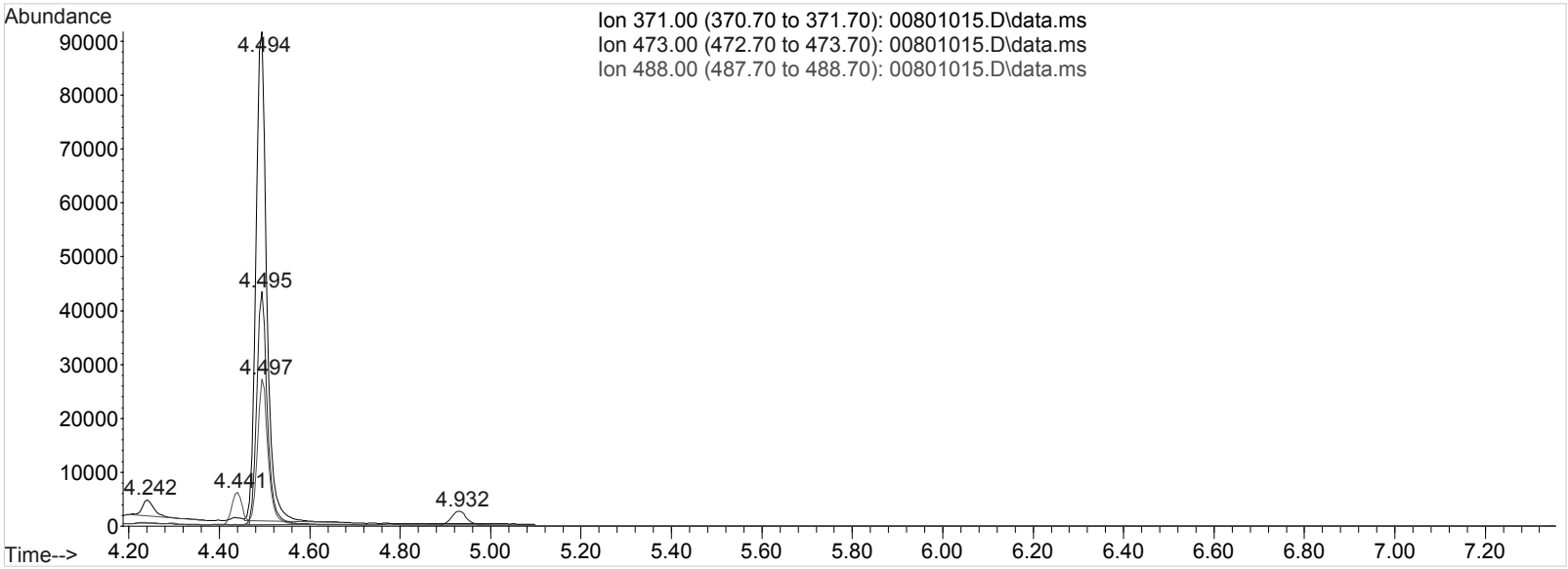
Sum of corrected areas: 775070

Signal : EIC Ion 488.00 (487.70 to 488.70): 00801015.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total
1	4.441	39	48	53	BV	5939	96909	21.42%	17.640%
2	4.497	53	59	79	VB	26671	452457	100.00%	82.360%

Sum of corrected areas: 549366

TOXI-A 10115.M Mon May 10 10:03:13 2021



Area Percent Report

Data Path : D:\DATA\2021\am 2\050621\
Data File : 00901003.D
Acq On : 06 May 2021 16:39
Operator : Instrument 65198
Sample : negative control am 3
Misc : am 3
ALS Vial : 9 Sample Multiplier: 1

Integration Parameters: autoint1.e
Integrator: ChemStation

Method : D:\MassHunter\GCMS\1\methods\TOXI-A 10115.M
Title :

Signal : EIC Ion 371.00 (370.70 to 371.70): 00901003.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total

No peaks were detected using the method integration parameters!

Signal : EIC Ion 473.00 (472.70 to 473.70): 00901003.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total

No peaks were detected using the method integration parameters!

Signal : EIC Ion 488.00 (487.70 to 488.70): 00901003.D\data.ms

peak #	R.T. min	first scan	max scan	last scan	PK TY	peak height	corr. area	corr. % max.	% of total
1	4.445	40	49	64	BB	5314	91574	100.00%	100.000%

Sum of corrected areas: 91574

TOXI-A 10115.M Mon May 10 10:04:01 2021