



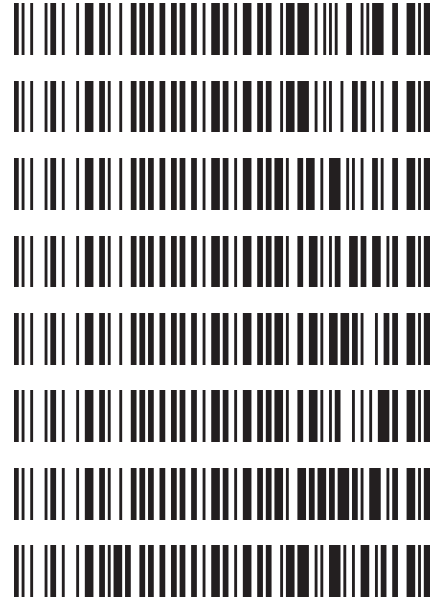
REVIEWED

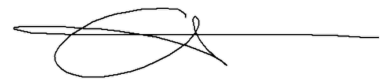
By Brittany Wylie at 8:04 am, Nov 14, 2019

11/13/2019

Worklist: 3825

<u>LAB CASE</u>	<u>ITEM</u>	<u>ITEM TYPE</u>	<u>DESCRIPTION</u>
C2019-2064	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2019-2065	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2019-2113	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2019-2119	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2019-2121	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2019-2125	1	BCK	AM 27 Blood THC Quant by LC-QQQ
C2019-2135	1	UCK	AM 27 Blood THC Quant by LC-QQQ
M2018-2893	3	UCK	AM 27 Blood THC Quant by LC-QQQ





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

Extraction Date: 11/8/19
Plate lot#: 190716

Analyst: Anne Nord
Plate Expiration: 1-16-2020

Mobile phase A: 0.1% Formic Acid in LCMS Water
MTBE LCMS Methanol

Mobile phase B: 0.1% Formic acid in Acetonitrile
Hexane

Blank Blood Lot: 19H52275-1 **Urine Blank:** 11719
LCMS-QQQ ID: 69679

Column: UCT Selectra DA 100 x 2.1mm 3um

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.

Analytic:

- 1. Remove standards, plate, controls, and samples from cold storage. Allow to reach room temperature.
- 2. Urine hydrolysis: add 1.5 ml urine to blank plate, add 250 ul 1N KOH mix and incubate at 40 degrees for 15 minutes.
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 blood sample, 500 ul saturated phosphate buffer in urine in wells of analytical plate.
- 5. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- 6. Transfer 800µL of blood+acid or urine 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.
- 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 blood), 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? (if not is it describe in comments section)
- 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 urine preparation information

Stock solution 8 ul (100 ug/ml) C-THC in 9.992 mls urine
Ppd 11/8/19 Exp: 3/1/20 lot 3120 by AMN

Drug	lot	expiration	lot
C-THC	FE03121501	3/1/2020	3120

concentration ~ 80 ng/ml Carboxy THC

AM #27 Cannabinoids

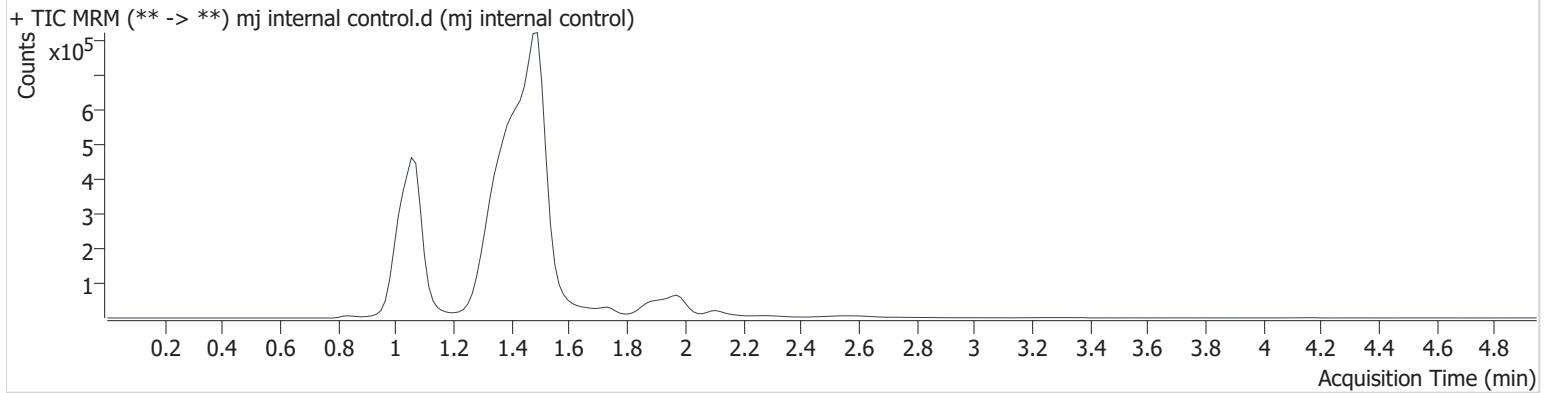


Batch results G:\TOX\CDA\Instrument 069679\2019\am28-27 110819\QuantResults\cann quant.batch.bin
Calibration Last Update 11/13/2019 2:07:48 PM

Instrument	69679	Data File	mj internal control.d
Type	QC	Sample	mj internal control
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-H1	Comment	
Injection Volume	10		
Acq. Date-Time	11/8/2019 3:10:25 PM		

Sample Info.

Sample Chromatogram



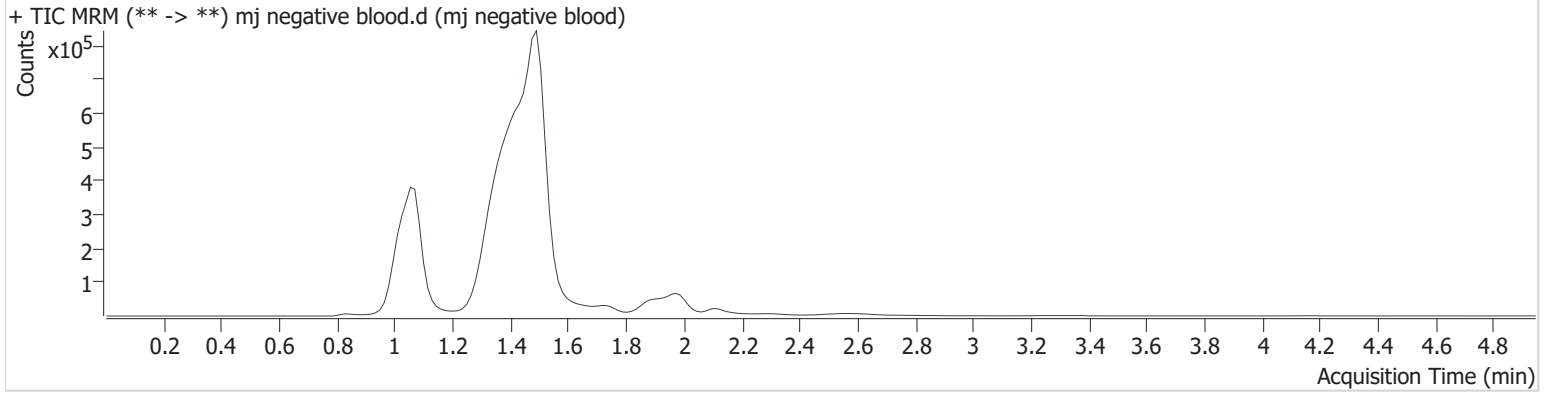
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.070	108197	227.7	37.3	1987.2	651025	14.949 ng/ml
THC-OH	1.074	137881	∞	8.4	∞	1613988	5.055 ng/ml
THC	1.980	13526	59.6	24.7	29.9	468558	4.368 ng/ml

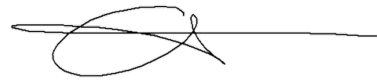
AM #27 Cannabinoids

Batch results G:\TOX\CDA\Instrument 069679\2019\am28-27 110819\QuantResults\cann quant.batch.bin
Calibration Last Update 11/13/2019 2:07:48 PM

Instrument	69679	Data File	mj negative blood.d
Type	Sample	Sample	mj negative blood
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-A2	Comment	
Injection Volume	10		
Acq. Date-Time	11/8/2019 3:18:06 PM		
Sample Info.			

Sample Chromatogram



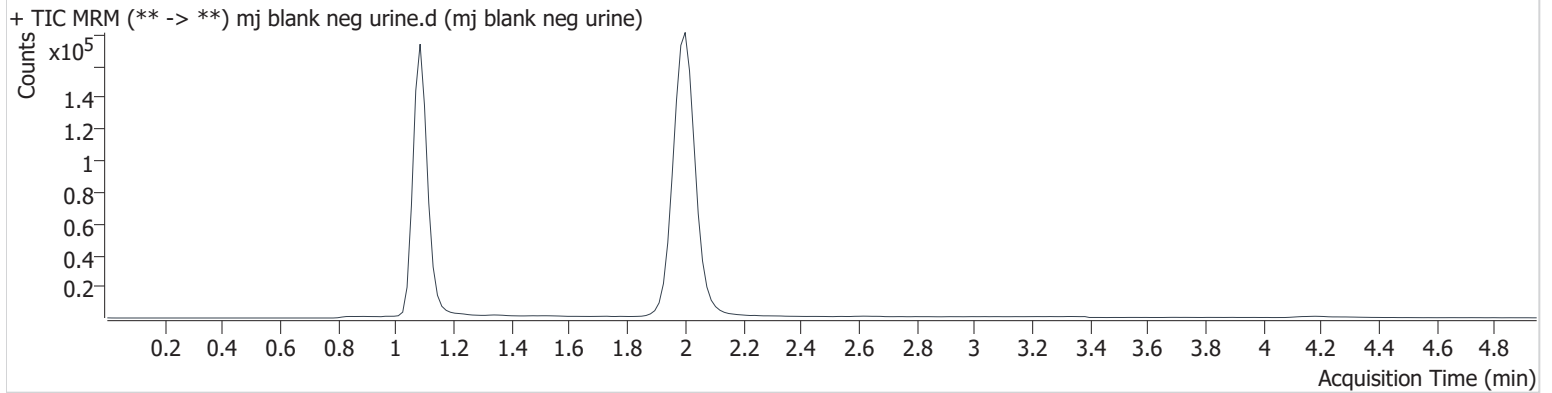


AM #27 Cannabinoids

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Calibration Last Update 11/13/2019 2:07:48 PM

Instrument	69679	Data File	mj blank neg urine.d
Type	Sample	Sample	mj blank neg urine
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	Vial 2	Comment	
Injection Volume	10		
Acq. Date-Time	11/8/2019 5:05:43 PM		
Sample Info.			

Sample Chromatogram



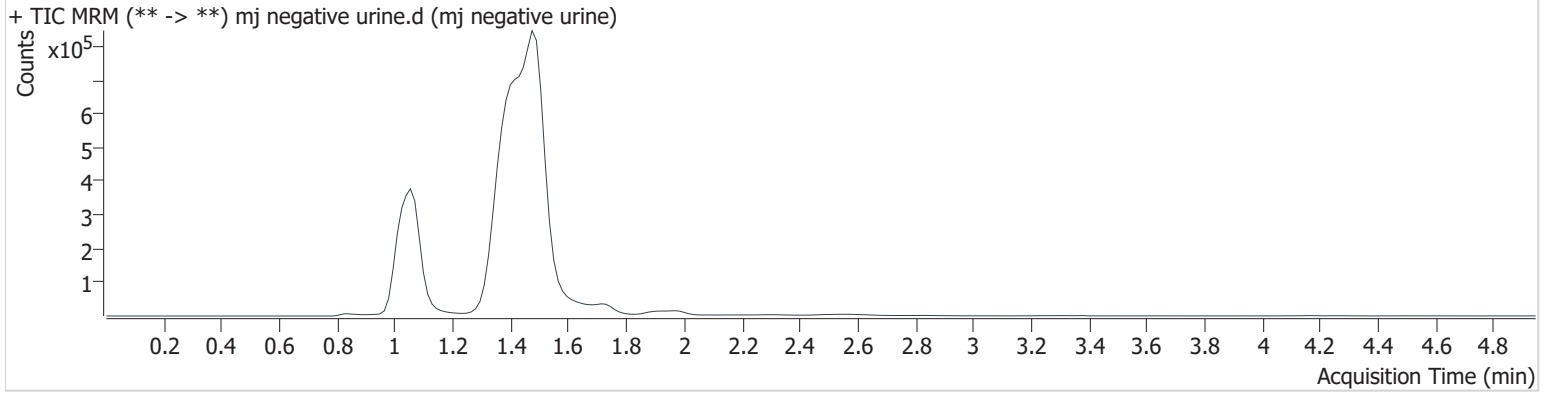
AM #27 Cannabinoids



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Calibration Last Update 11/13/2019 2:07:48 PM

Instrument	69679	Data File	mj negative urine.d
Type	Sample	Sample	mj negative urine
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-H2	Comment	
Injection Volume	10		
Acq. Date-Time	11/8/2019 5:13:19 PM		
Sample Info.			

Sample Chromatogram

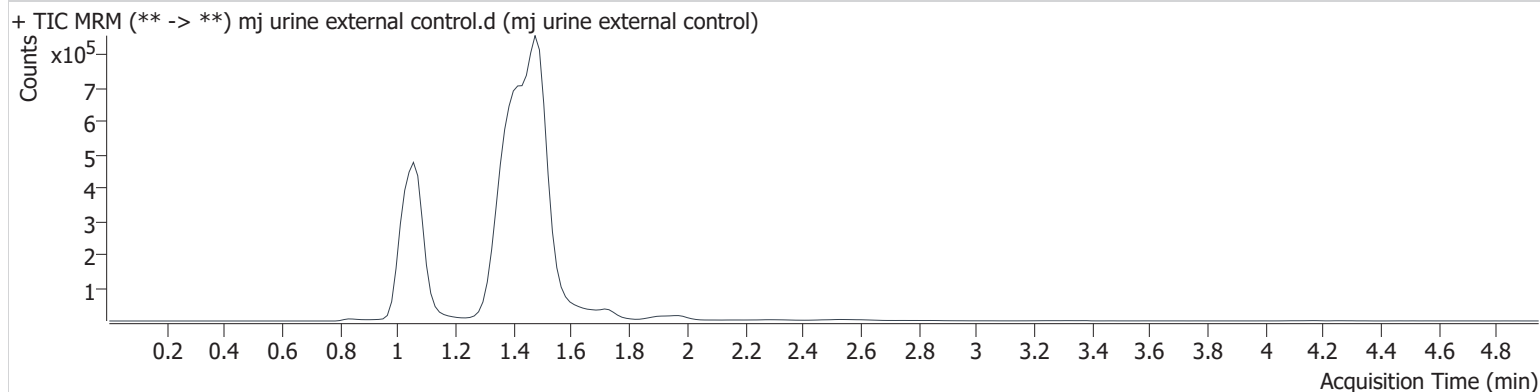


AM #27 Cannabinoids

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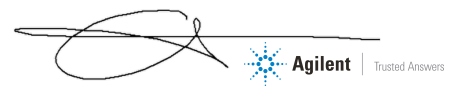
Instrument	69679	Data File	mj urine external control.d
Type	Sample	Sample	mj urine external control
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-A3	Comment	
Injection Volume	10		
Acq. Date-Time	11/8/2019 5:28:39 PM		
Sample Info.			

Sample Chromatogram



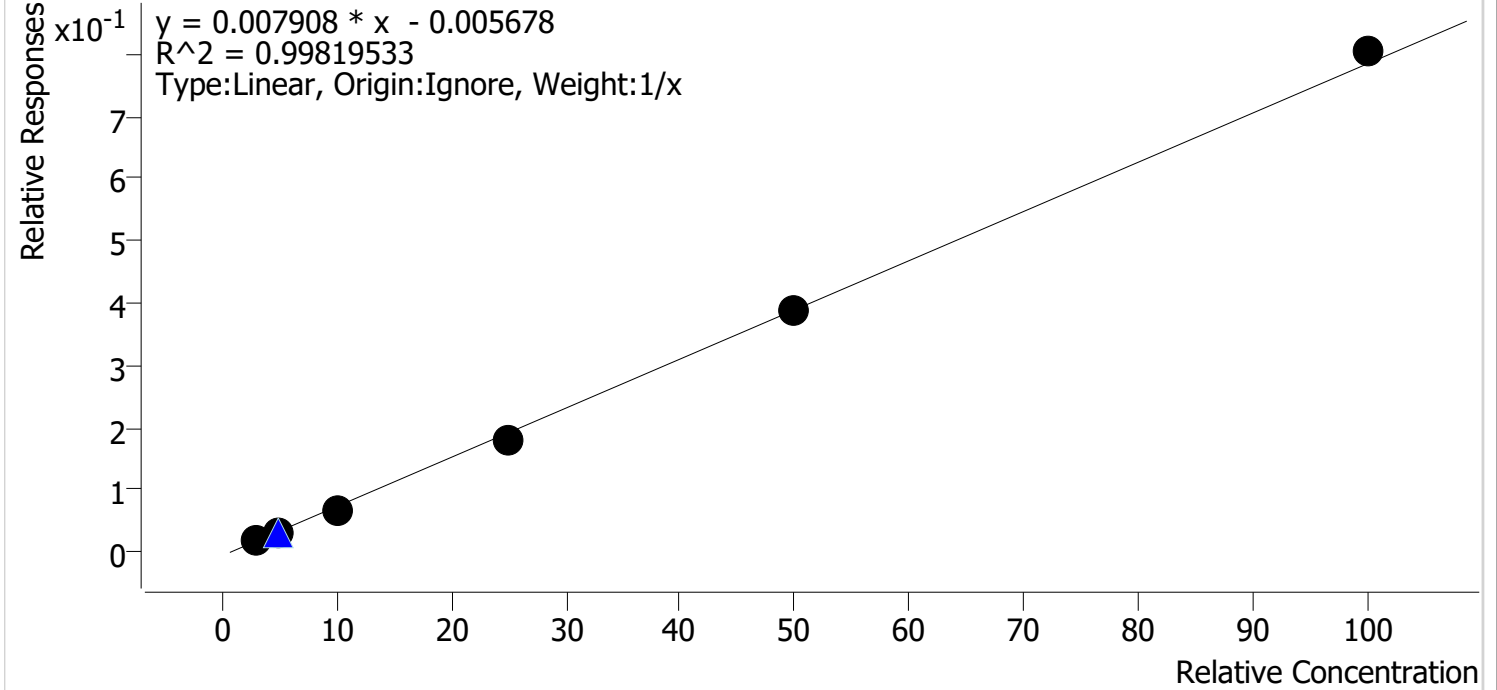
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.070	205917	3884.2	37.6	402.7	430999	40.128 ng/ml

Compound Calibration Report



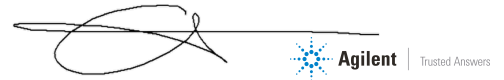
Batch results G:\TOX\CDA\Instrument 069679\2019\am28-27 110819\QuantResults\cann quant.batch.bin
Last Cal. Update 11/13/2019 2:07 PM
Analyst Name ISP\anord
Analyte THC **Internal Standard** THC-d3

THC - 6 Levels, 6 Levels Used, 6 Points, 6 Points Used, 1 QCs



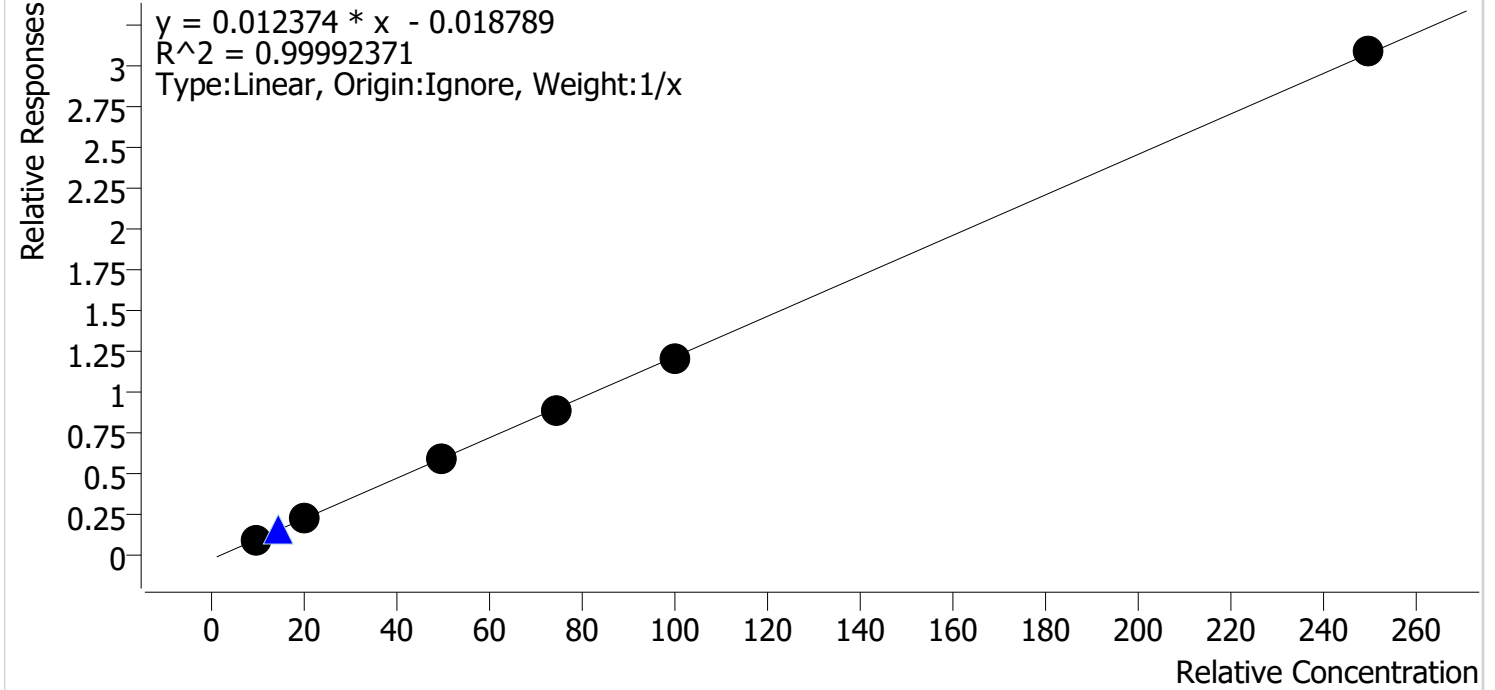
Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal2	2	✓	3.0	3.3	110.2
mj cal 3	3	✓	5.0	5.1	101.2
mj cal 4	4	✓	10.0	9.4	94.3
mj cal 5	5	✓	25.0	23.2	92.8
mj cal 6	6	✓	50.0	49.6	99.2
mj cal 7	7	✓	100.0	102.4	102.4

Compound Calibration Report



Batch results G:\TOX\CDA\Instrument 069679\2019\am28-27 110819\QuantResults\cann quant.batch.bin
Last Cal. Update 11/13/2019 2:07 PM
Analyst Name ISP\anord
Analyte THC-COOH **Internal Standard** THC-COOH-d9

THC-COOH - 6 Levels, 6 Levels Used, 6 Points, 6 Points Used, 1 QCs



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
mj cal2	2	✓	10.0	10.0	100.0
mj cal 3	3	✓	20.0	20.1	100.7
mj cal 4	4	✓	50.0	50.4	100.8
mj cal 5	5	✓	75.0	74.3	99.1
mj cal 6	6	✓	100.0	99.0	99.0
mj cal 7	7	✓	250.0	251.1	100.5

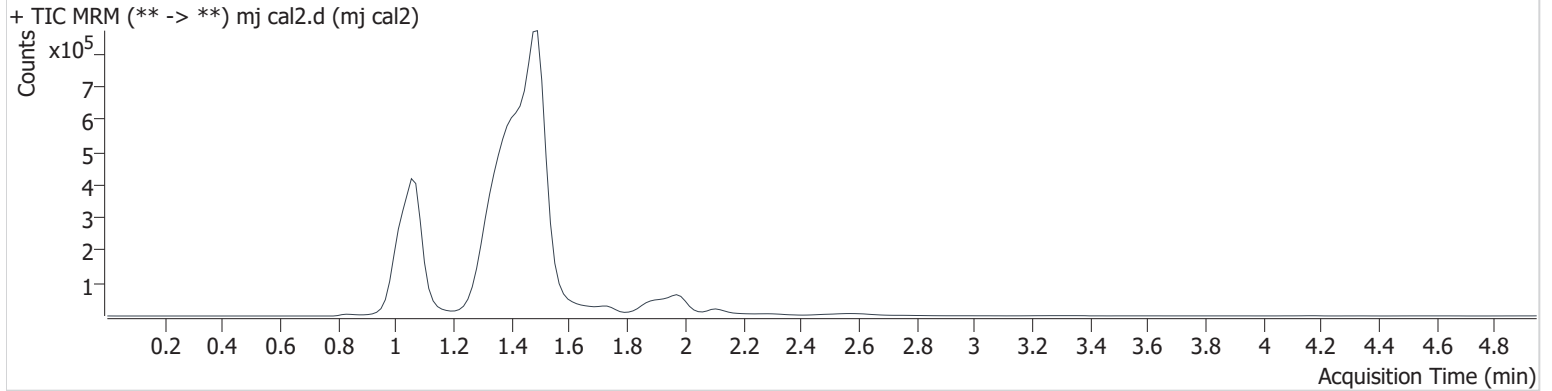
AM #27 Cannabinoids

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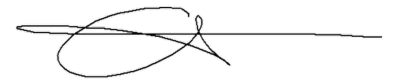
Instrument 69679
Type Cal
Acq. Method AM 27 THC quant.m
Sample Position P3-B1
Injection Volume 10
Acq. Date-Time 11/8/2019 2:24:11 PM
Sample Info.

Data File mj cal2.d
Sample mj cal2
Operator Anne Nord
Comment

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.	
THC-COOH	1.070	63762	1886.6	40.8	2347.8	607418	10.002 ng/ml	
THC-OH	1.074	77739	∞	10.0	∞	1563420	2.521 ng/ml	Low
THC	1.995	9586	134.8	25.9	∞	468628	3.305 ng/ml	



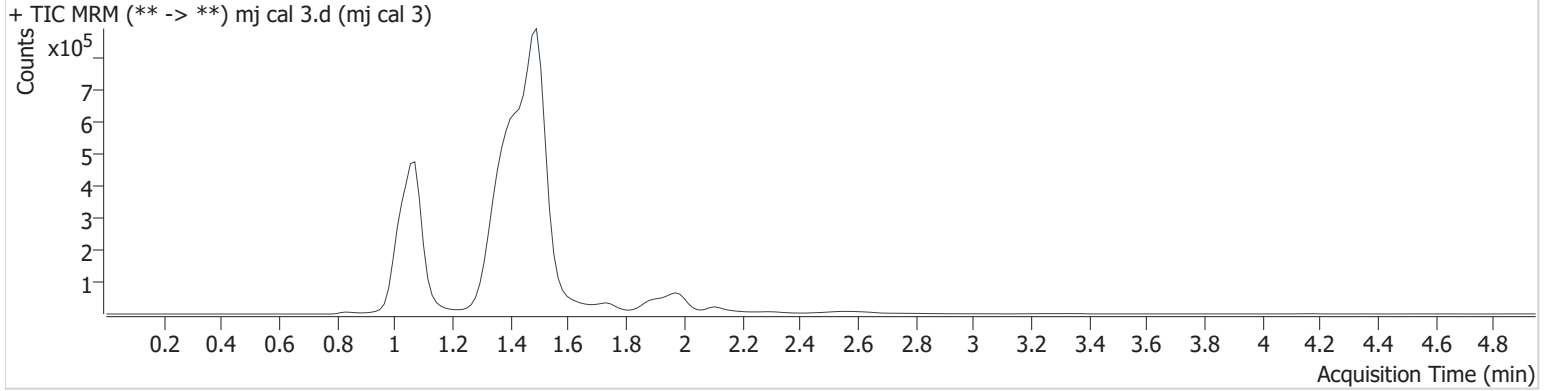
AM #27 Cannabinoids

Batch results G:\TOX\CDA\Instrument 069679\2019\am28-27 110819\QuantResults\cann quant.batch.bin
Calibration Last Update 11/13/2019 2:07:48 PM

Instrument	69679	Data File	mj cal 3.d
Type	Cal	Sample	mj cal 3
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-C1	Comment	
Injection Volume	10		
Acq. Date-Time	11/8/2019 2:31:53 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.070	138802	254.1	37.3	402.3	602579	20.133 ng/ml
THC-OH	1.074	145097	∞	9.7	∞	1567315	5.562 ng/ml
THC	1.995	15355	768.8	27.3	∞	447391	5.058 ng/ml

AM #27 Cannabinoids

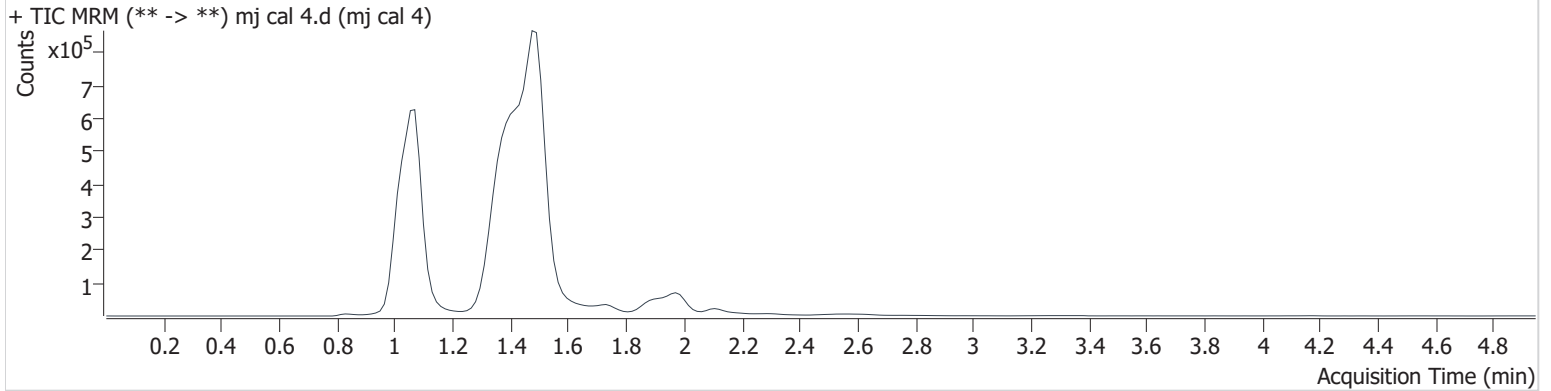


Batch results G:\TOX\CDA\Instrument 069679\2019\am28-27 110819\QuantResults\cann quant.batch.bin
Calibration Last Update 11/13/2019 2:07:48 PM

Instrument	69679	Data File	mj cal 4.d
Type	Cal	Sample	mj cal 4
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-D1	Comment	
Injection Volume	10		
Acq. Date-Time	11/8/2019 2:39:36 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.070	371335	1395.2	37.9	85643.8	614219	50.375 ng/ml
THC-OH	1.074	262267	∞	9.1	∞	1604070	10.596 ng/ml
THC	1.980	32200	∞	26.2	∞	467402	9.430 ng/ml

AM #27 Cannabinoids

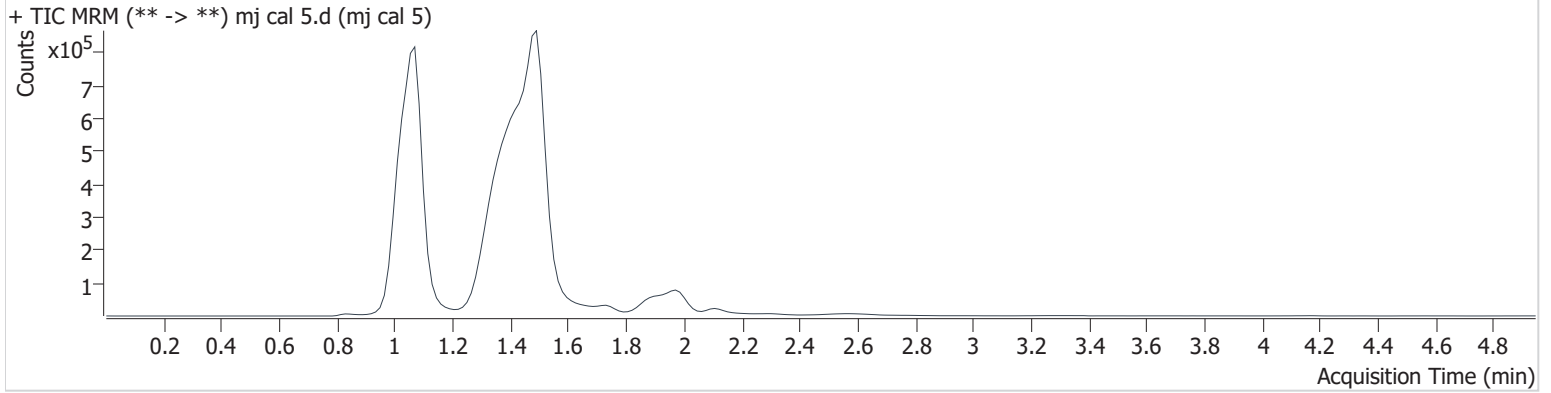


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Calibration Last Update 11/13/2019 2:07:48 PM

Instrument	69679	Data File	mj cal 5.d
Type	Cal	Sample	mj cal 5
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-E1	Comment	
Injection Volume	10		
Acq. Date-Time	11/8/2019 2:47:18 PM		

Sample Info.

Sample Chromatogram



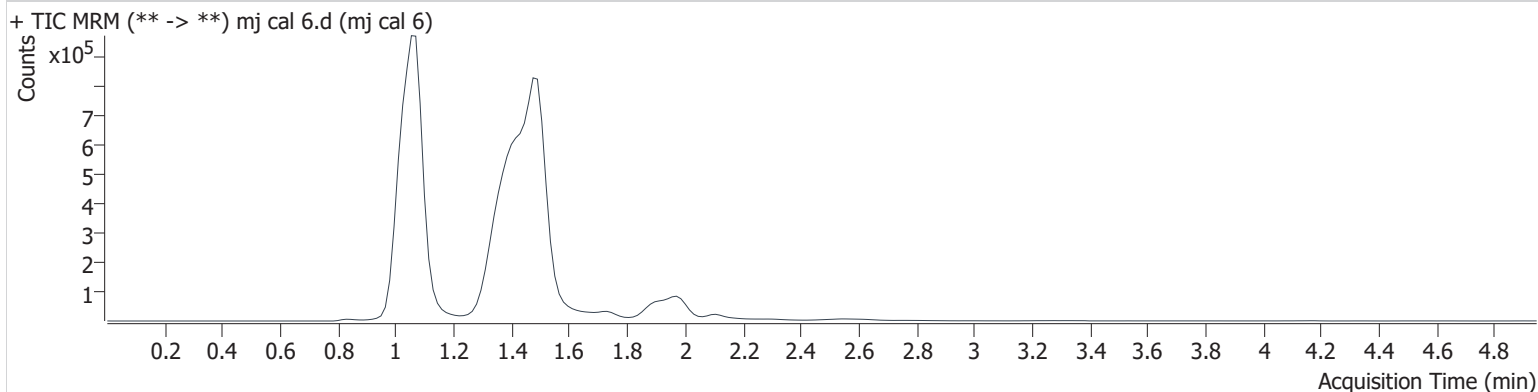
Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.070	600795	2045.4	38.2	3819.7	667137	74.295 ng/ml
THC-OH	1.074	592223	∞	9.9	∞	1631171	24.759 ng/ml
THC	1.980	86762	1313.8	22.2	70.3	488230	23.190 ng/ml

AM #27 Cannabinoids

Batch results G:\TOX\CDA\Instrument 069679\2019\am28-27 110819\QuantResults\cann quant.batch.bin
Calibration Last Update 11/13/2019 2:07:48 PM


Instrument	69679	Data File	mj cal 6.d
Type	Cal	Sample	mj cal 6
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-F1	Comment	
Injection Volume	10		
Acq. Date-Time	11/8/2019 2:55:01 PM		
Sample Info.			

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.070	722633	3487.8	38.7	4665.2	598764	99.049 ng/ml
THC-OH	1.059	1109073	∞	11.2	∞	1537915	50.173 ng/ml
THC	1.980	167476	1127.3	25.4	∞	433192	49.607 ng/ml

AM #27 Cannabinoids

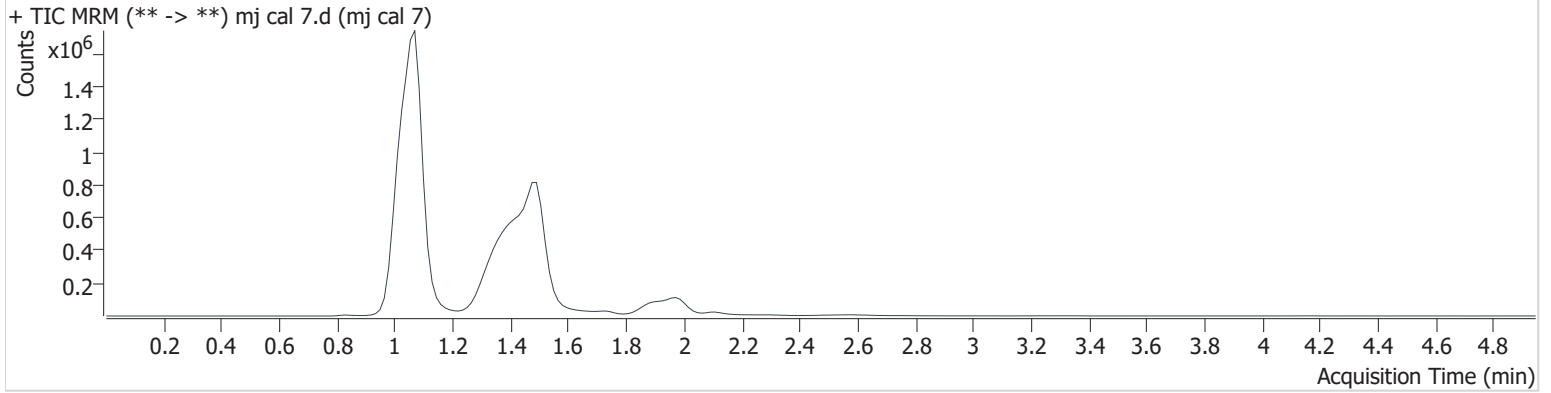


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Calibration Last Update 11/13/2019 2:07:48 PM

Instrument	69679	Data File	mj cal 7.d
Type	Cal	Sample	mj cal 7
Acq. Method	AM 27 THC quant.m	Operator	Anne Nord
Sample Position	P3-G1	Comment	
Injection Volume	10		
Acq. Date-Time	11/8/2019 3:02:43 PM		

Sample Info.

Sample Chromatogram



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
THC-COOH	1.070	1823119	19899.0	38.4	56514 0.8	590207	251.145 ng/ml
THC-OH	1.074	2197517	∞	11.8	∞	1553450	99.389 ng/ml
THC	1.980	370129	3759.2	24.2	1503.1	460256	102.411 ng/ml