



















**Worklist: 4728**

REVIEWED

By Anthony Kyte at 10:44 am, Jan 15, 2021

| <u>LAB CASE</u> | <u>ITEM</u> | <u>ITEM TYPE</u> | <u>DESCRIPTION</u> | |
|-----------------|-------------|------------------|--|---|
| C2020-2566 | 2 | UCK | AM 25/AM 26 Urine MultiDrug/THC Screen by LC-QQQ |  |
| C2020-2572 | 3 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2020-2609 | 1 | UCK | AM 25/AM 26 Urine MultiDrug/THC Screen by LC-QQQ |  |
| C2020-2611 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2020-2615 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2020-2618 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2020-2618 | 4 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0009 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0023 | 2 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0037 | 1 | UCK | AM 25/AM 26 Urine MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0047 | 3 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0048 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0049 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0050 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0056 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0066 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0068 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2021-0070 | 1 | BCK | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |

AM# 25: Multi-Drug Screen in Blood and Urine by LC-MS/MS

Extraction Date: 1/11/21 Analyst: Anne Nord
Plate lot#: 200511 Plate Expiration: 11/11/2020

Mobile phase A: 10mM Amm Form
0.5M Ammonium Hydroxide

Mobile phase B: 0.1% Formic Acid in MeOH
Ethyl Acetate LC Methanol

Blank Blood Lot: 20J20793 **Blank Urine lot:** 10120 **Column:** Phenomenex Phenyl Hexyl (4.6x50mm, 2.6um)
LCMS-QQQ ID: 69679

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 pipette: 250 ul urine in blank well, add 40 ul BG Turbo, add 100 ul 500 mm sodium phosphate buffer mix for at least five minutes ambient temperature.
Pipette 250 µL blood (calibrated pipette) or 250 ul urine in wells of analytical (standards) plate. **Pipette ID: 1926134**
- 3. Place on shaking incubator at ambient temp., 900 rpm for 15 minutes.
- 4. Pipette 250 µL of 0.5 M ammonium hydroxide in wells of analytical plate.
- 5. Place on shaking incubator at ambient temp., 900 rpm for 15 minutes.
- 6. Transfer 300 µL of blood or urine+base 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 900 µL ethyl acetate.
- 10. Wait 5 minutes.
- 11. Apply positive pressure for approx. 10-15 seconds. *(12-15 PSI- Selector to the left).*
- 12. Add 900 µL ethyl acetate.
- 13. Wait 5 minutes.
- 14. Apply positive pressure for approx. 10-15 seconds. *(12-15 PSI- Selector to the left).*
- 15. Remove plate containing eluate. add 50 ul 1% HCl in MeOH Place on SPE Dry and evaporate to dryness at approx. 35°C.
SPE Dry ID: 66819
- 16. Reconstitute in 100 µL 20% LC MeOH in LC Water and heat seal plate with foil. Place in autosampler and run worklist.

Post-Analytic

- 1. Open quantitation software and create a new quantitation batch.
- 2. Make necessary changes to integration limits
- 3. Evaluate samples, S/N of primary transition >5 and S/N of secondary transition >3 or evaluation of peak symmetry and resolution. Within +/- 2% or 0.1 min RT of administrative control. Calculated concentration 5 or greater or 2-5 for discretionary range.
- 4. Did all QCs pass for each analyte? (If no is it described in comments?)
- 5. Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS: Due to the extraction occurring after the expiration of the analytical plate, an external control was included with this run as specified in the analytical method.

Toxicology AM method 25/28 urine external control prep
working solution 10000 ng/ml in meoh methamphetamine, temazepam, midazolam
Stock solution 1mg/ml 50 ul each in 4850 ul MeOH (fisher 195629)

ppd 8/6/20: Exp: 4/1/2021 lot 4121 by baw

| Drug | lot | expiration |
|-----------------|------------|------------|
| Methamphetamine | FE08101708 | 10/1/2022 |
| midazolam | FE01221602 | 4/1/2021 |
| temazepam | FE04261601 | 5/1/2021 |

AM 25/28 control 500 ul working solution (4121) in 4500 ul negative urine (1000ng/mL Expected concentration)

ppd 8/6/20, exp 4/1/2021 lot u4121 negative urine 73020 by AMN

AM 25/28 Blood Control: 50ul working solution (4121) in 4950 ul neg blood (100ng/mL Expected concentration)

ppp 8/6/20, exp 4/1/21 lot b4121 neg blood 20G20792 by AMN

A

| | 1 | 2 | 3 | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12 |
|---|---------------|-----------|-----------|---|---|----------------|---|---|---|----|----|-------|
| A | Cal 1 | 20-2615-1 | 21-0050-1 | | | negative urine | | | | | | |
| B | Cal 1 | 20-2618-1 | 21-0056-1 | | | urine control | | | | | | |
| C | Cal 2 | 20-2618-4 | 21-0066-1 | | | 20-2566-2 | | | | | | |
| D | Cal 2 | 21-0009-1 | 21-0068-1 | | | 20-2609-1 | | | | | | |
| E | neg blood | 21-0023-2 | 21-0070-1 | | | 21-0037-1 | | | | | | Cal 2 |
| F | blood control | 21-0047-3 | | | | | | | | | | Cal 2 |
| G | 20-2572-3 | 21-0048-1 | | | | | | | | | | Cal 1 |
| H | 20-2611-1 | 21-0049-1 | | | | | | | | | | Cal 1 |

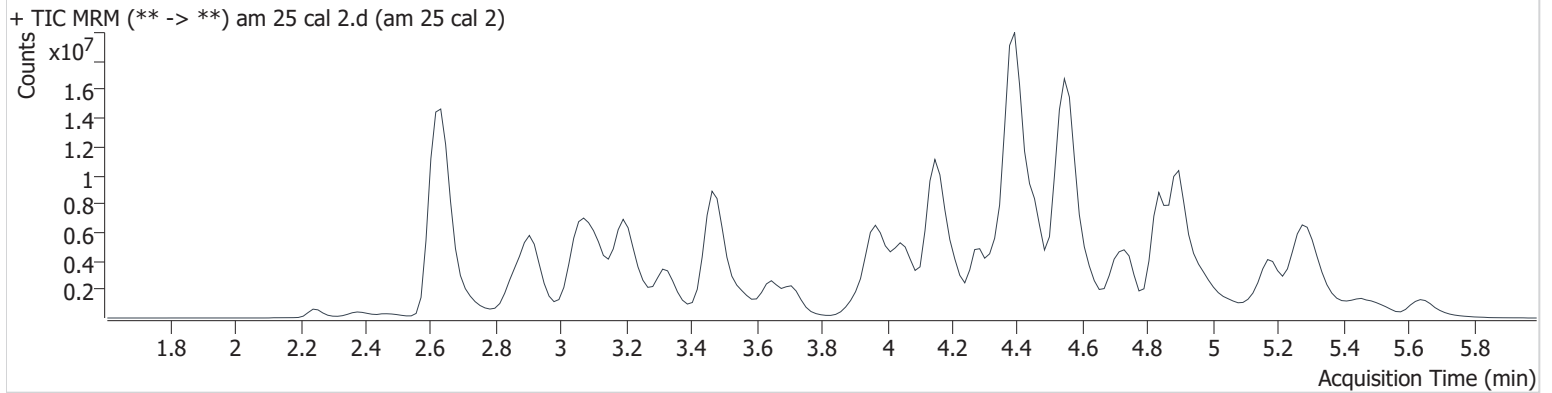
C20__-____-__

AM #25 Multi-Drug Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\mds.batch.bin
Calibration Last Update 1/11/2021 3:21:39 PM

| | | | |
|-------------------------|-----------------------|------------------|---------------|
| Instrument | 69679 | Data File | am 25 cal 2.d |
| Type | Cal | Sample | am 25 cal 2 |
| Acq. Method | mds 826.m | Operator | Anne Nord |
| Sample Position | P2-C1 | Comment | |
| Injection Volume | 2.5 | | |
| Acq. Date-Time | 1/11/2021 12:07:33 PM | | |

Sample Chromatogram



| Name | RT | Resp. | S/N | S/N | ISTD Resp. | Calc. Conc. |
|------------------------|-------|---------|----------|---------|------------|-------------|
| 6-MAM | 3.614 | 58759 | 49.5 | 50.4 | 1634732 | 10.000 |
| 7-aminoclonazepam | 3.331 | 965262 | 247.1 | 1013.5 | 3734768 | 10.000 |
| 7-aminoflunitrazepam | 3.558 | 1407590 | 1019.3 | 265.1 | 3734768 | 10.000 |
| Acetyl Fentanyl | 4.686 | 387222 | 171.4 | 94510.1 | 27694399 | 10.000 |
| Acetyl Norfentanyl | 2.910 | 256928 | 2924.5 | 354.5 | 27694399 | 10.000 |
| a-hydroxyalprazolam | 4.302 | 205631 | 600.4 | 63729.1 | 3734768 | 10.000 |
| alpha-hydroxymidazolam | 4.409 | 2518391 | 451.2 | 375.8 | 3734768 | 10.000 |
| alpha-PHP | 4.511 | 3049911 | 720.3 | 743.9 | 9684519 | 10.000 |
| alpha-PVP | 4.282 | 4222671 | 795.6 | 407.6 | 9684519 | 10.000 |
| Alprazolam | 4.429 | 1519992 | 635.5 | 829.0 | 10877549 | 10.000 |
| Amitriptyline | 5.285 | 2255839 | 127.8 | 544.0 | 10707862 | 10.000 |
| Amphetamine | 2.916 | 4439358 | 740.1 | 524.3 | 9684519 | 10.000 |
| Benzoylcegonine | 3.072 | 138894 | 732.1 | 51.0 | 293345 | 10.000 |
| Brompheniramine | 4.717 | 124042 | 56.4 | 57.7 | 52354648 | 10.000 |
| Buprenorphine | 5.258 | 675798 | 2530.0 | 60573.9 | 3037974 | 10.000 |
| Bupropion | 4.419 | 3920733 | 1586.7 | 719.9 | 15801926 | 10.000 |
| Carbamazepine | 3.991 | 5506094 | 5814.1 | 1288.1 | 116009 | 10.000 |
| Carisoprodol | 3.973 | 893583 | 1760.1 | 68.3 | 5217550 | 10.000 |
| Chlordiazepoxide | 4.538 | 882837 | 2025.0 | 210.5 | 10877549 | 10.000 |
| Chlorpheniramine | 4.570 | 6868419 | 746.6 | 73.3 | 52354648 | 10.000 |
| Citalopram | 4.624 | 3385876 | 421.8 | 196.8 | 52354648 | 10.000 |
| Clomipramine | 5.641 | 3434568 | 8130.5 | 1577.7 | 10707862 | 10.000 |
| Clonazepam | 4.242 | 505853 | 316.7 | 193.2 | 10877549 | 10.000 |
| Clonazolam | 4.162 | 601687 | 2639.8 | 1542.0 | 10877549 | 10.000 |
| Cocaethylene | 4.334 | 4226452 | 1440.8 | 477.4 | 29723519 | 10.000 |
| Cocaine | 4.183 | 5513108 | 166883.3 | 1147.1 | 29723519 | 10.000 |
| Codeine | 3.571 | 405141 | 1181.9 | 176.1 | 8292681 | 10.000 |
| Cyclobenzaprine | 5.149 | 4401608 | 1457.7 | 269.8 | 10707862 | 10.000 |
| Desipramine | 5.211 | 5167975 | ∞ | 282.7 | 10707862 | 10.000 |
| Dextromethorphan | 4.982 | 2873813 | 742.0 | 614.8 | 14124409 | 10.000 |
| Dextrorphan | 3.913 | 2712516 | 5237.6 | 1641.7 | 14124409 | 10.000 |
| Diazepam | 4.675 | 760709 | 326.8 | 372.4 | 10877549 | 10.000 |
| Dihydrocodeine | 3.115 | 998438 | 286.9 | 188.9 | 8292681 | 10.000 |
| Diphenhydramine | 4.571 | 9573299 | 1118.4 | 447.3 | 52354648 | 10.000 |

AM #25 Multi-Drug Screen Results

| Name | RT | Resp. | S/N | S/N | ISTD Resp. | Calc. Conc. |
|----------------------|-------|----------|-----------|----------|------------|-------------|
| Doxepin | 4.965 | 2395146 | 381.1 | 52.9 | 22176674 | 10.000 |
| Doxylamine | 4.049 | 8647837 | 3429.9 | 2108.6 | 14124409 | 10.000 |
| EDDP | 4.401 | 1634027 | 355.1 | 284.3 | 3464338 | 10.000 |
| Estazolam | 4.338 | 3181540 | 1234.9 | 1068.1 | 10877549 | 10.000 |
| Etizolam | 4.455 | 163057 | 43695.3 | 711.3 | 10877549 | 10.000 |
| Fentanyl | 4.853 | 324155 | 247.4 | 112123.5 | 18714730 | 10.000 |
| Flualprazolam | 4.286 | 601038 | 3487.3 | 594.2 | 10877549 | 10.000 |
| Flunitrazepam | 4.366 | 1426177 | 778.7 | 1375.4 | 10877549 | 10.000 |
| Fluoxetine | 4.734 | 2436011 | 635.9 | 154.7 | 5659390 | 10.000 |
| Flurazepam | 4.804 | 3239915 | 4165406.3 | 834388.1 | 10877549 | 10.000 |
| Hydrocodone | 4.092 | 1426487 | 222.8 | 215.7 | 8292681 | 10.000 |
| Hydromorphone | 2.876 | 1132060 | 185.2 | 712.1 | 202726 | 10.000 |
| Imipramine | 5.298 | 8099740 | 3016.9 | ∞ | 10707862 | 10.000 |
| Ketamine | 4.220 | 3178293 | 1477.4 | 130.8 | 10582269 | 10.000 |
| Lamotrigine | 3.409 | 259180 | 461.9 | 9322.2 | 52354648 | 10.000 |
| Levamisole | 3.642 | 2261222 | 2766.8 | 34.1 | 29723519 | 10.000 |
| Levetireacetam | 2.247 | 756191 | 430.8 | 1764.0 | 5217550 | 10.000 |
| Lorazepam | 4.210 | 284058 | ∞ | 320.6 | 10877549 | 10.000 |
| Maprotiline | 5.285 | 1656425 | 1618.0 | 638.7 | 10707862 | 10.000 |
| MDA | 3.141 | 2761135 | 784.3 | 1053.1 | 24771717 | 10.000 |
| MDEA | 3.489 | 3840862 | 1490.1 | 402.2 | 24771717 | 10.000 |
| MDMA | 3.322 | 4327077 | 622.2 | 313.1 | 24771717 | 10.000 |
| Meperidine | 4.280 | 2660353 | 1785.4 | 645.4 | 14124409 | 10.000 |
| Meprobamate | 3.351 | 320082 | 426.7 | 74.3 | 5217550 | 10.000 |
| Methadone | 4.840 | 5859916 | 677.5 | 514.9 | 3464338 | 10.000 |
| Methamphetamine | 3.112 | 7379550 | 203.8 | 391.7 | 24771717 | 10.000 |
| Methocarbamol | 3.273 | 241156 | 161.8 | 6850.2 | 3464338 | 10.000 |
| Methylphenidate | 3.976 | 8640345 | 330.4 | 2649.4 | 19527148 | 10.000 |
| Metoprolol | 3.651 | 670546 | ∞ | 17092.8 | 14124409 | 10.000 |
| Midazolam | 4.624 | 555562 | 898.4 | 13423.5 | 10877549 | 10.000 |
| Mirtazapine | 4.708 | 3526726 | 1659.4 | 696.0 | 14124409 | 10.000 |
| Mitragynine | 4.864 | 450043 | 209834.8 | 525.2 | 14124409 | 10.000 |
| Morphine | 2.485 | 318708 | ∞ | 856.5 | 202726 | 10.000 |
| Norbuprenorphine | 4.528 | 72540 | 183.9 | 22148.9 | 3037974 | 10.000 |
| Nordiazepam | 4.510 | 815683 | 644.9 | 1297.0 | 10877549 | 10.000 |
| Norfentanyl | 3.487 | 5119307 | 330.3 | 176.4 | 27694399 | 10.000 |
| Norhydrocodone | 3.269 | 64148 | 30.7 | 59.5 | 8292681 | 10.000 |
| norketamine | 4.023 | 524046 | 1316.9 | 3800.6 | 52354648 | 10.000 |
| Normeperidine | 3.946 | 2767586 | 155.9 | 357.4 | 52354648 | 10.000 |
| Noroxycodone | 3.070 | 1009760 | 84.6 | 98.3 | 10582269 | 10.000 |
| Nortriptyline | 5.332 | 1881170 | 265.8 | 55.5 | 10707862 | 10.000 |
| O-desmethyl-tramadol | 2.924 | 6236591 | 10219.4 | 359.6 | 52354648 | 10.000 |
| Olanzapine | 4.566 | 816248 | 999.4 | 268.3 | 116009 | 10.000 |
| Oxazepam | 4.292 | 501923 | 414.5 | 66.8 | 3266861 | 10.000 |
| Oxycodone | 3.326 | 1889424 | 195.7 | 150.5 | 10582269 | 10.000 |
| Oxymorphone | 2.375 | 1311454 | 191.6 | 4143.8 | 202726 | 10.000 |
| Paroxetine | 5.593 | 96695 | 53.8 | 66.9 | 5659390 | 10.000 |
| Phenazepam | 4.454 | 1054216 | 1153.5 | 2048.9 | 10877549 | 10.000 |
| Phencyclidine | 4.373 | 4313224 | 5425.6 | 368.3 | 14124409 | 10.000 |
| Phentermine | 3.233 | 2088082 | 686.6 | 22.4 | 19527148 | 10.000 |
| Phenytoin | 3.882 | 195247 | 272.6 | 134.5 | 116009 | 10.000 |
| Promethazine | 5.191 | 9883957 | 1119.8 | 687.5 | 52354648 | 10.000 |
| Pseudoephedrine | 2.642 | 62007184 | ∞ | ∞ | 24771717 | 10.000 |
| Quetiapine | 4.744 | 4814643 | 1635809.6 | 1405.9 | 31916733 | 10.000 |
| Sertraline | 5.460 | 1066935 | 865.3 | 490.5 | 5659390 | 10.000 |
| Sufentanil | 5.048 | 258910 | 1557.3 | 333.7 | 27694399 | 10.000 |
| Tapentadol | 3.657 | 4142609 | 4741.7 | 543.3 | 10582269 | 10.000 |
| Temazepam | 4.476 | 2072745 | 781.5 | 94.1 | 10877549 | 10.000 |
| Tramadol | 3.712 | 7014231 | ∞ | 85.7 | 52354648 | 10.000 |
| Trazodone | 4.897 | 4496230 | 2116338.1 | 1357.3 | 22176674 | 10.000 |

GA

AM #25 Multi-Drug Screen Results

| Name | RT | Resp. | S/N | S/N | ISTD Resp. | Calc. Conc. |
|-------------|-------|---------|---------|--------|------------|-------------|
| Venlafaxine | 4.171 | 5765394 | 993.3 | 140.3 | 5659390 | 10.000 |
| Zaleplon | 4.152 | 1139829 | 819.8 | 387.4 | 31916733 | 10.000 |
| Zolpidem | 4.398 | 6112353 | 30154.9 | 1073.5 | 31916733 | 10.000 |
| Zopiclone | 4.437 | 711587 | 687.4 | 323.2 | 3611194 | 10.000 |

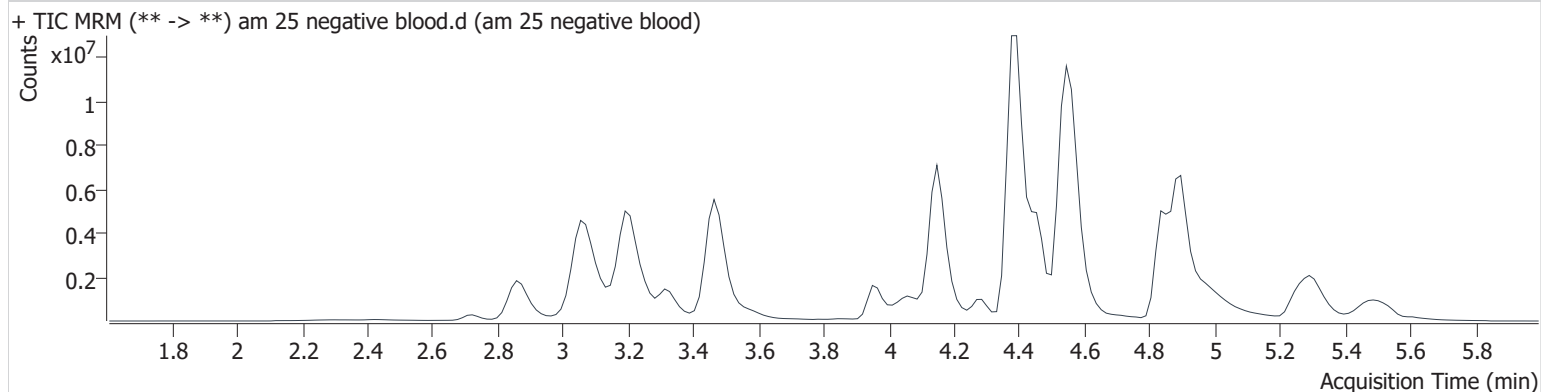
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AM #25 Multi-Drug Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\mds.batch.bin
Calibration Last Update 1/11/2021 3:21:39 PM

| | | | |
|-------------------------|-----------------------|------------------|------------------------|
| Instrument | 69679 | Data File | am 25 negative blood.d |
| Type | Sample | Sample | am 25 negative blood |
| Acq. Method | mds 826.m | Operator | Anne Nord |
| Sample Position | P2-E1 | Comment | |
| Injection Volume | 2.5 | | |
| Acq. Date-Time | 1/11/2021 12:14:16 PM | | |
| Sample Info. | | | |

Sample Chromatogram



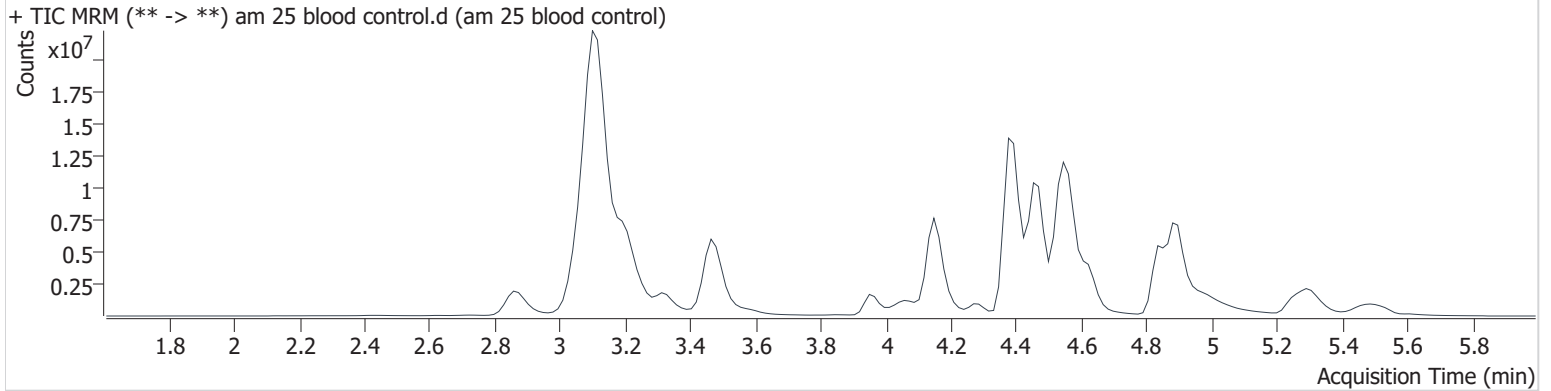
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AM #25 Multi-Drug Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\mds.batch.bin
Calibration Last Update 1/11/2021 3:21:39 PM

| | | | |
|-------------------------|-----------------------|------------------|-----------------------|
| Instrument | 69679 | Data File | am 25 blood control.d |
| Type | Sample | Sample | am 25 blood control |
| Acq. Method | mds 826.m | Operator | Anne Nord |
| Sample Position | P2-F1 | Comment | |
| Injection Volume | 2.5 | | |
| Acq. Date-Time | 1/11/2021 12:20:57 PM | | |
| Sample Info. | | | |

Sample Chromatogram



| Name | RT | Resp. | S/N | S/N | ISTD Resp. | Calc. Conc. |
|-----------------|-------|----------|-----------|--------|------------|-------------|
| Methamphetamine | 3.127 | 46537353 | 36152.8 | 725.1 | 21894665 | 71.349 |
| Midazolam | 4.624 | 4336804 | 3828519.6 | 8905.1 | 10168188 | 83.507 |
| Temazepam | 4.476 | 17105684 | ∞ | 823.2 | 10168188 | 88.284 |

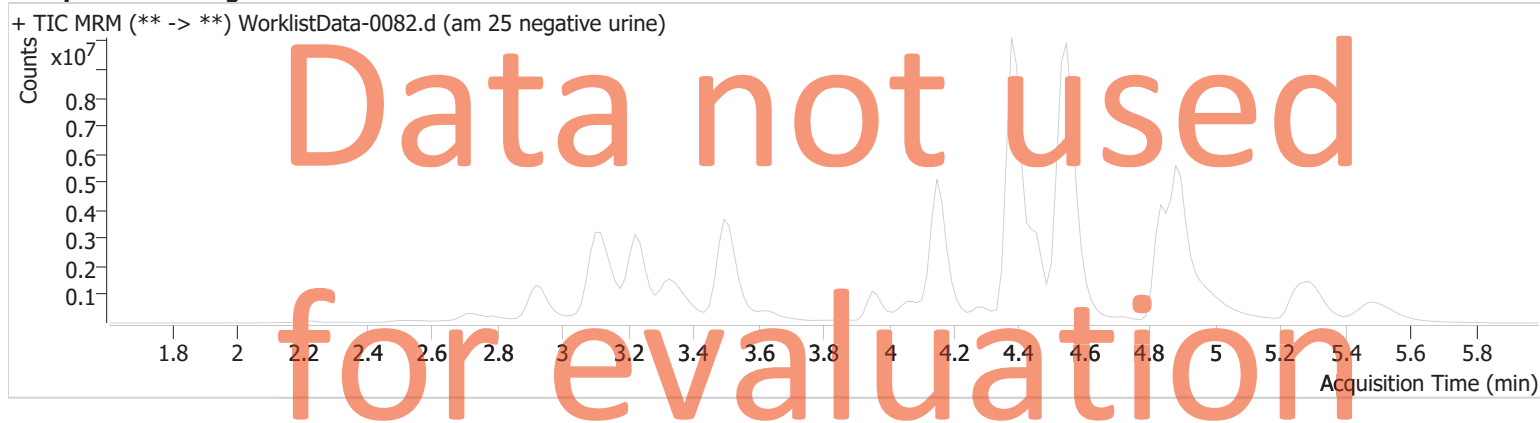
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AM #25 Multi-Drug Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\mds.batch.bin
Calibration Last Update 1/11/2021 3:21:39 PM

| | | | |
|-------------------------|----------------------|------------------|----------------------|
| Instrument | 69679 | Data File | WorklistData-0082.d |
| Type | Sample | Sample | am 25 negative urine |
| Acq. Method | mds 826.m | Operator | Anne Nord |
| Sample Position | P2-A6 | Comment | |
| Injection Volume | 2.5 | | |
| Acq. Date-Time | 1/11/2021 2:10:57 PM | | |
| Sample Info. | | | |

Sample Chromatogram



This sample injected before the mobile phases equilibrated sample was re-injected and evaluated.

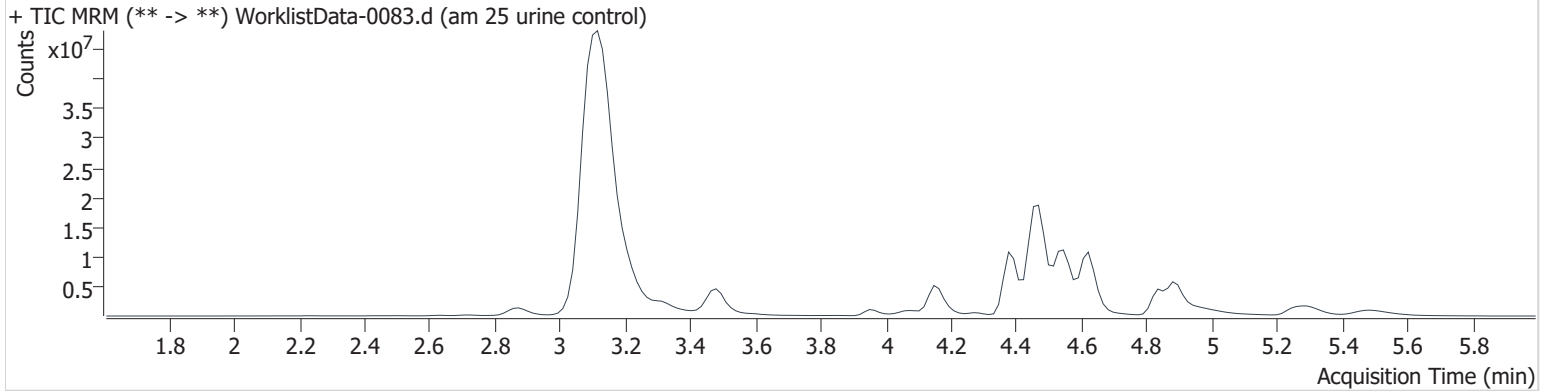
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AM #25 Multi-Drug Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\mds.batch.bin
Calibration Last Update 1/11/2021 3:21:39 PM

| | | | |
|-------------------------|----------------------|------------------|---------------------|
| Instrument | 69679 | Data File | WorklistData-0083.d |
| Type | Sample | Sample | am 25 urine control |
| Acq. Method | mds 826.m | Operator | Anne Nord |
| Sample Position | P2-B6 | Comment | |
| Injection Volume | 2.5 | | |
| Acq. Date-Time | 1/11/2021 2:17:37 PM | | |

Sample Chromatogram



| Name | RT | Resp. | S/N | S/N | ISTD Resp. | Calc. Conc. |
|-----------------|-------|-----------|---------|--------|------------|-------------|
| Methamphetamine | 3.127 | 111290101 | 10834.9 | 459.6 | 14812170 | 252.211 |
| Midazolam | 4.624 | 17232316 | 7253.9 | 5713.8 | 5786012 | 583.127 |
| Temazepam | 4.476 | 47270561 | 9566.0 | 1227.7 | 5786012 | 428.743 |

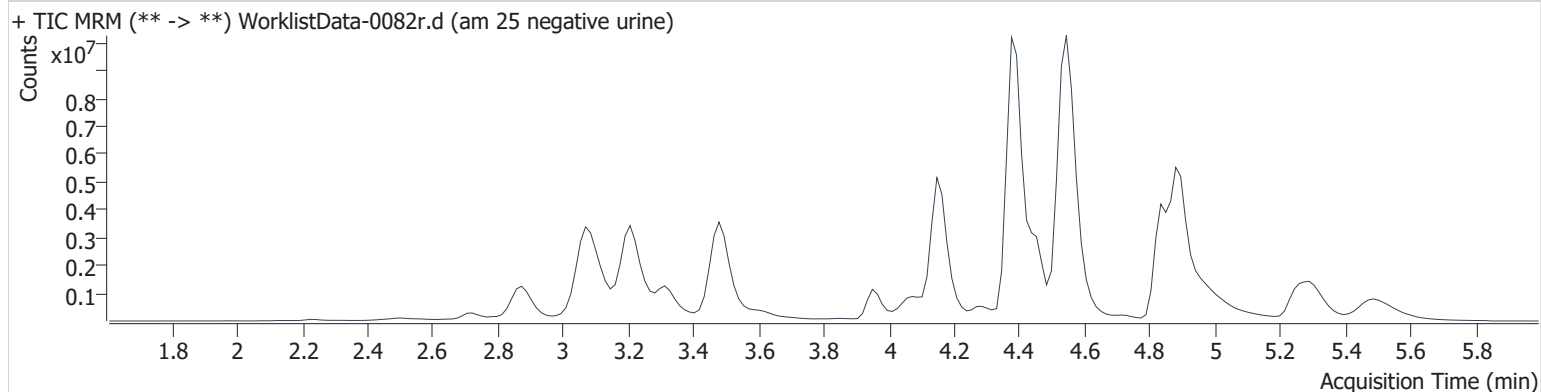
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AM #25 Multi-Drug Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\mds.batch.bin
Calibration Last Update 1/11/2021 3:21:39 PM

| | | | |
|-------------------------|----------------------|------------------|----------------------|
| Instrument | 69679 | Data File | WorklistData-0082r.d |
| Type | Sample | Sample | am 25 negative urine |
| Acq. Method | mds 826.m | Operator | Anne Nord |
| Sample Position | P2-A6 | Comment | |
| Injection Volume | 2.5 | | |
| Acq. Date-Time | 1/11/2021 2:50:55 PM | | |
| Sample Info. | | | |

Sample Chromatogram



AM# 26: THC and Metabolites Screen in Blood by LC-MS/MS

Extraction Date: 1/11/21 Analyst: Anne Nord

Plate lot#: 200723 Plate Expiration: 1/23/2021

Mobile phase A: 10mM Ammonium Formate
0.1% Formic Acid in Water

Mobile phase B: 0.1% Formic acid in MeOH
MTBE Hexane

Blank Blood Lot: 20J20793 **Urine Blank:** 10120 **Column:** Phenomenex Phenyl Hexyl (4.6x50mm: 2.6 um)
LCMS-QQQ ID: 69679

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 1000 µL blood (calibrated pipette) in wells of analytical (standards) plate. Pipette ID: K52558g
Pipette 1000 ul urine to analytical (standards) plate.
- 3. Place on shaking incubator at ambient temp., 900 rpm for 15 minutes.
- 4. Pipette 500 µL 0.1% formic acid in blood wells 500 ul saturated phosphate buffer in urine wells of analytical plate.
- 5. Place on shaking incubator at ambient temp., 900 rpm for 15 minutes.
- 6. Transfer 800 µL 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 2.25 mL MTBE (add in 3 increments of 750 µL).
- 10. Wait 5 minutes.
- 11. Apply positive pressure for approx. 10-15 seconds. (12-15 PSI- Selector to the left).
- 12. Add 2.25 mL hexane (add in 3 increments of 750 µL).
- 13. Wait 5 minutes.
- 14. Apply positive pressure for approx. 10-15 seconds. (12-15 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 100 µL 100% LCMS MeOH and heat seal plate with foil. Place in autosampler and run worklist.

Post-Analytic

- 1. Create batch and process data.
- 2. Calculated sample concentration of 3 ng/mL or greater for THC and THC-OH, a calculated sample concentration of 10 ng/mL or greater for Carboxy-THC.
- 3. Retention time within +/- 2% or +/-0.100 min whichever is greater of the average retention time of the calibrators.
- 4. Did all QCs pass for each analyte? Yes
- 5. Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports

COMMENTS: Extracts run on both am 26 and am 30, for am 26 samples were reconstituted and run 1/14/20
Urine did not evaluate THC control too low.

Toxicology AM method 27/26 external prep information GA

working solution 15 ug/ml in meoh C-THC, THC-OH, 7.5 ug/ml THC

Stock solution 1mg/ml 7.5 ul each THC, 100 ug/ml 150 ul C-THC, 150 ul THC-OH in 9692.5 ul meOH

Ppd 8/26/20 Exp: 7/1/21 lot 82620 by AMN

| Drug | lot | expiration |
|--------|------------|------------|
| C-THC | FE01061702 | 3/1/2022 |
| THC-OH | FE07221601 | 7/1/2021 |
| THC | FE01041701 | 3/1/2022 |

AM 27/26 blood control 100 ul working solution lot () in 9900 ul blood lot ()

| | | | |
|--|--|--|--|
| | | Concentration 7.5 ng/ml THC, 15 ng/ml C-THC, THC-OH | |
|--|--|--|--|

AM 27/26 urine control 400 ul working solution lot (82620) in 9600 ul urine

out of use

| | | | | |
|---|------------|---|--------|-----------|
| ppd 8/26/20 Exp 7/1/21 neg urine lot 73020 | lot u82620 | Concentration 30 ng/ml THC, and 60 ng/ml C-THC, THC-OH | by amn | 10/4/2020 |
| ppd 10/5/20 Exp 7/1/21 neg urine lot 10120 | lot 10520 | Concentration 30 ng/ml THC, and 60 ng/ml C-THC, THC-OH | by amn | 1/12/2021 |
| ppd 1/13/21 Exp 7/1/21 neg urine lot 10120 | lot 11321 | Concentration 30 ng/ml THC, and 60 ng/ml C-THC, THC-OH | by amn | |
| | | | | |

| | 1 | 2 | 3 | 4 | 5 | 6 |
|---|----------|-----------|---------------------------------|----------------|---|------------|
| a | cal 1ng | neg blood | 21-0047-3 | negative urine | | QC 1 |
| b | cal 3 ng | 20-2615-1 | 21-0048-1 1-11-21 | urine control | | cal 100 ng |
| c | cal 5 ng | 20-2611-1 | 21-0049-1 | 20-2566-2 | | cal 50 ng |
| d | cal 10ng | 20-2615-1 | 21-0050-1 | 20-2609-1 | | cal 25 ng |
| e | 25 | 20-2618-1 | 21-0056-1 | 21-0037-1 | | cal 10ng |
| f | 50 | 20-2618-4 | 21-0066-1 | 21-0048-1 | | cal 5 ng |
| g | 100 | 21-0009-1 | 21-0068-1 | | | cal 3 ng |
| h | QC 1 | 21-0023-2 | 21-0070-1 | | | cal 1ng |

C20_--_--_--

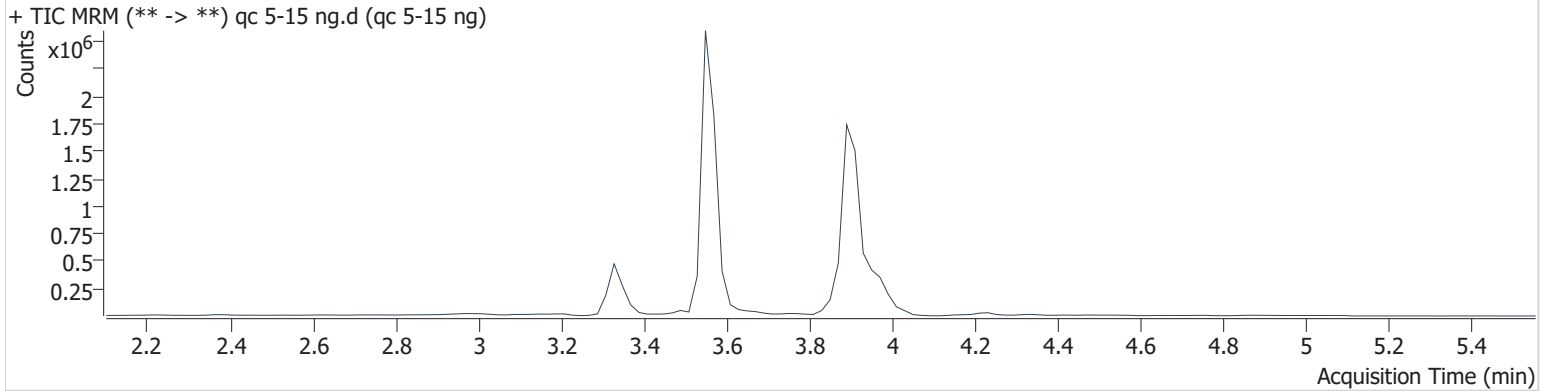
Q

AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|--------------|
| Instrument | 69679 | Data File | qc 5-15 ng.d |
| Type | QC | Sample | qc 5-15 ng |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-H1 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 10:24:56 AM | | |
| Sample Info. | | | |

Sample Chromatogram



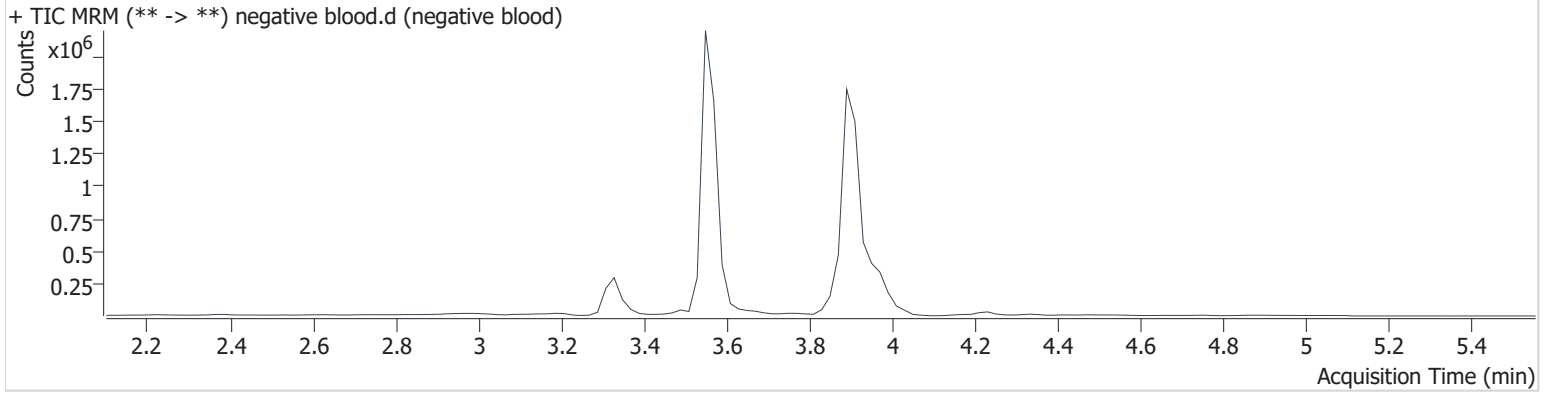
| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|-------|--------|------------|--------------|
| THC | 3.984 | 26765 | 669213 | 4.639 ng/ml |
| THC-COOH | 3.331 | 166189 | 738550 | 17.008 ng/ml |
| THC-OH | 3.558 | 55639 | 5984262 | 4.661 ng/ml |

AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|------------------|
| Instrument | 69679 | Data File | negative blood.d |
| Type | Sample | Sample | negative blood |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-A2 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 10:31:32 AM | | |
| Sample Info. | | | |

Sample Chromatogram

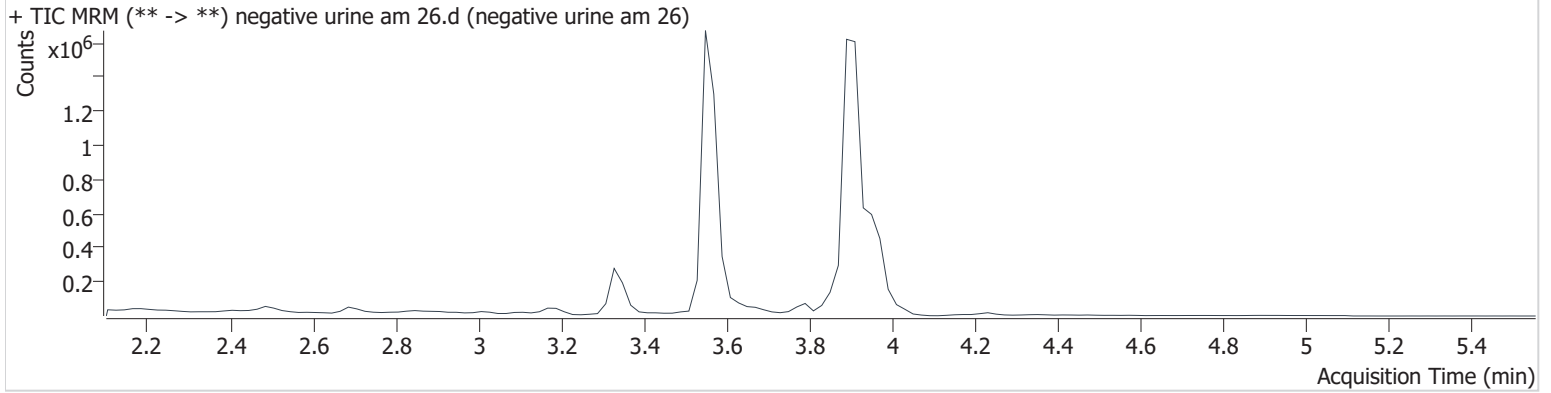


AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|------------------------|
| Instrument | 69679 | Data File | negative urine am 26.d |
| Type | Sample | Sample | negative urine am 26 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-A4 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 12:10:30 PM | | |
| Sample Info. | | | |

Sample Chromatogram



| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|------------------|-------|------------|------------------------|
| THC-COOH | 3.171 Low | 11214 | 695315 | 1.217 ng/ml Low |

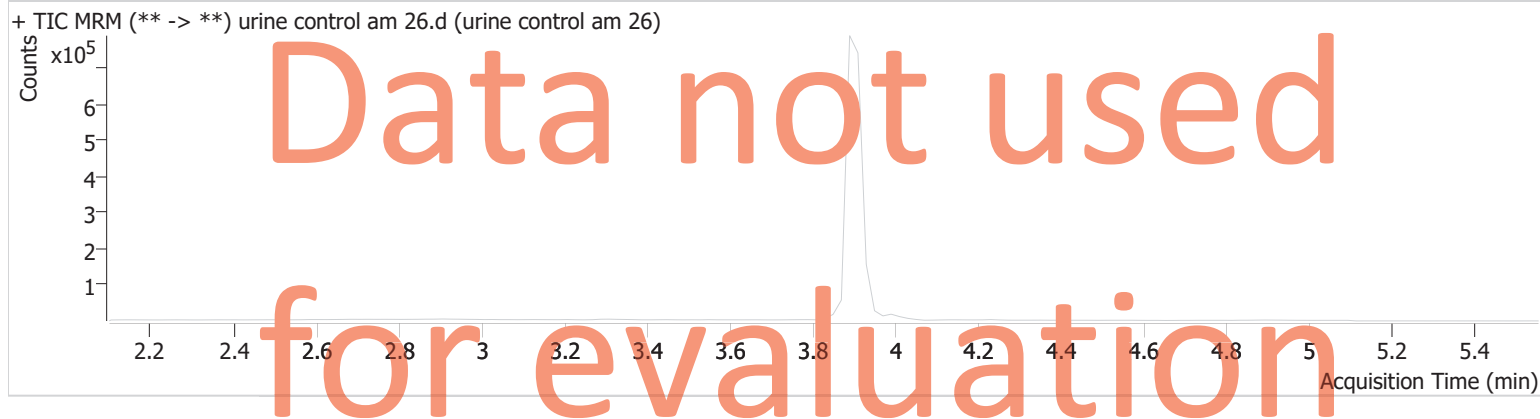
A

AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|-----------------------|
| Instrument | 69679 | Data File | urine control am 26.d |
| Type | Sample | Sample | urine control am 26 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-B3 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 12:17:06 PM | | |
| Sample Info. | | | |

Sample Chromatogram



wrong plate position injected. The correct position was injected and evaluated.

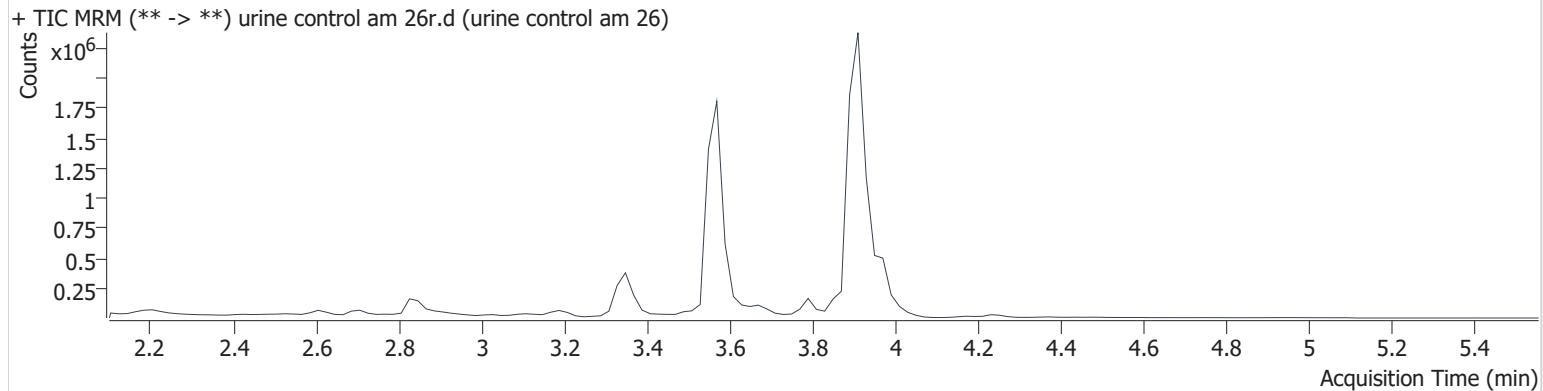
GA

AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|------------------------|
| Instrument | 69679 | Data File | urine control am 26r.d |
| Type | Sample | Sample | urine control am 26 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-B4 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 1:18:41 PM | | |
| Sample Info. | | | |

Sample Chromatogram



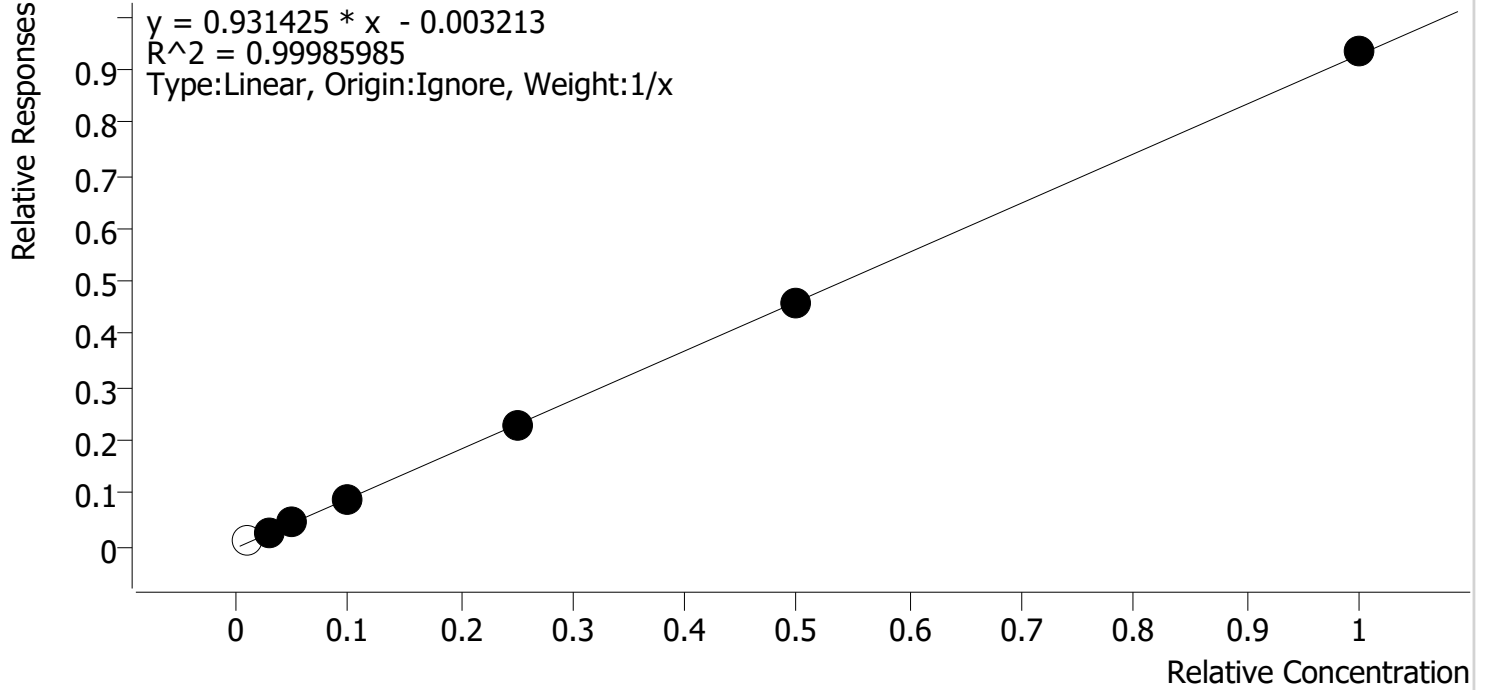
| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|-------|--------|------------|------------------------|
| THC | 3.984 | 21440 | 884785 | 2.947 ng/ml Low |
| THC-COOH | 3.351 | 154725 | 661399 | 17.682 ng/ml |
| THC-OH | 3.578 | 123573 | 3785159 | 15.908 ng/ml |

Compound Calibration Report



Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Last Cal. Update 1/14/2021 1:52 PM
Analyst Name ISP\datastor
Analyte THC **Internal Standard** THC-d3

THC - 7 Levels, 6 Levels Used, 7 Points, 6 Points Used, 0 QCs



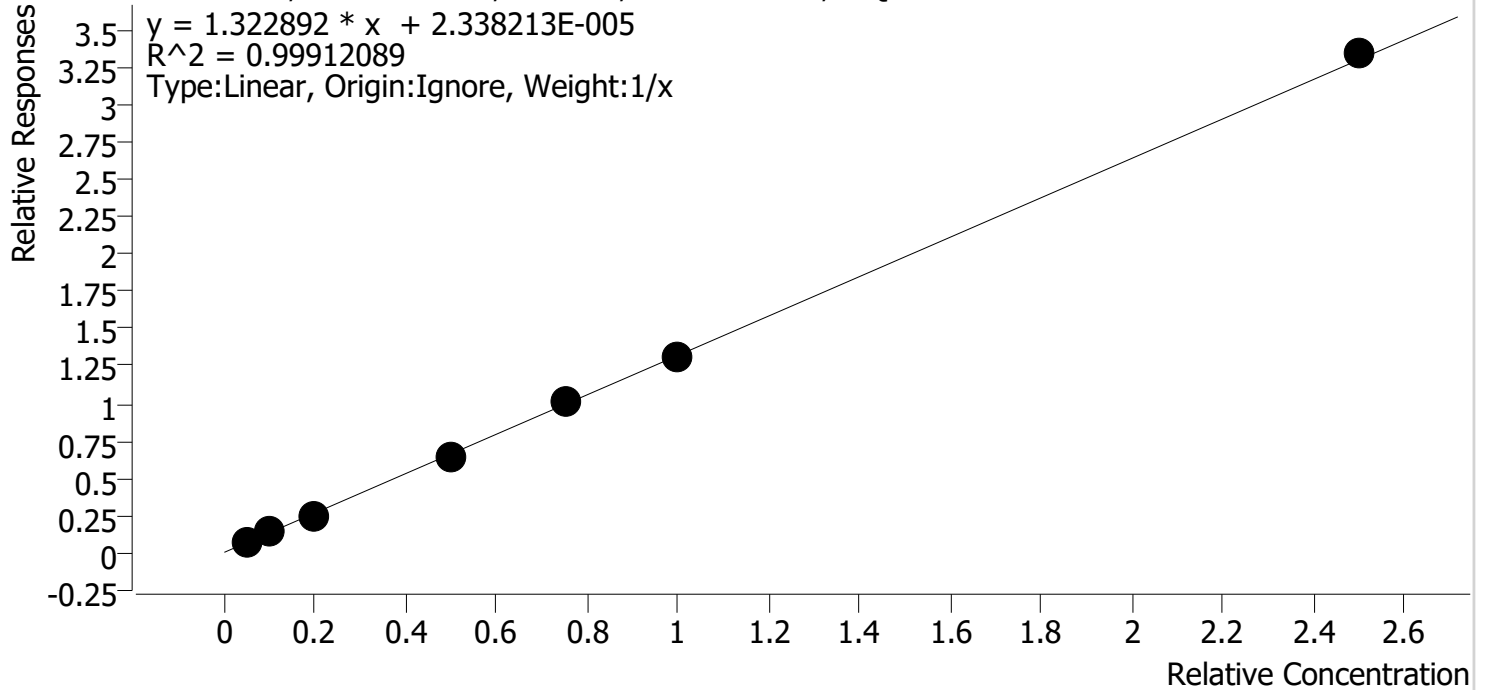
| Sample | Level | Enabled | Expected Concentration | Final Concentration | Accuracy |
|--------|-------|---------|------------------------|---------------------|----------|
| cal 1 | 1 | x | 1.0 | 1.7 | 167.9 |
| cal 2 | 2 | ✓ | 3.0 | 3.0 | 101.0 |
| cal 3 | 3 | ✓ | 5.0 | 5.1 | 102.6 |
| cal 4 | 4 | ✓ | 10.0 | 9.8 | 97.7 |
| cal 5 | 5 | ✓ | 25.0 | 24.6 | 98.5 |
| cal-6 | 6 | ✓ | 50.0 | 49.8 | 99.5 |
| cal-7 | 7 | ✓ | 100.0 | 100.7 | 100.7 |

Compound Calibration Report



| | | | |
|-------------------------|--|--------------------------|-------------|
| Batch results | D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin | | |
| Last Cal. Update | 1/14/2021 1:52 PM | | |
| Analyst Name | ISP\datastor | | |
| Analyte | THC-COOH | Internal Standard | THC-COOH-d9 |

THC-COOH - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs



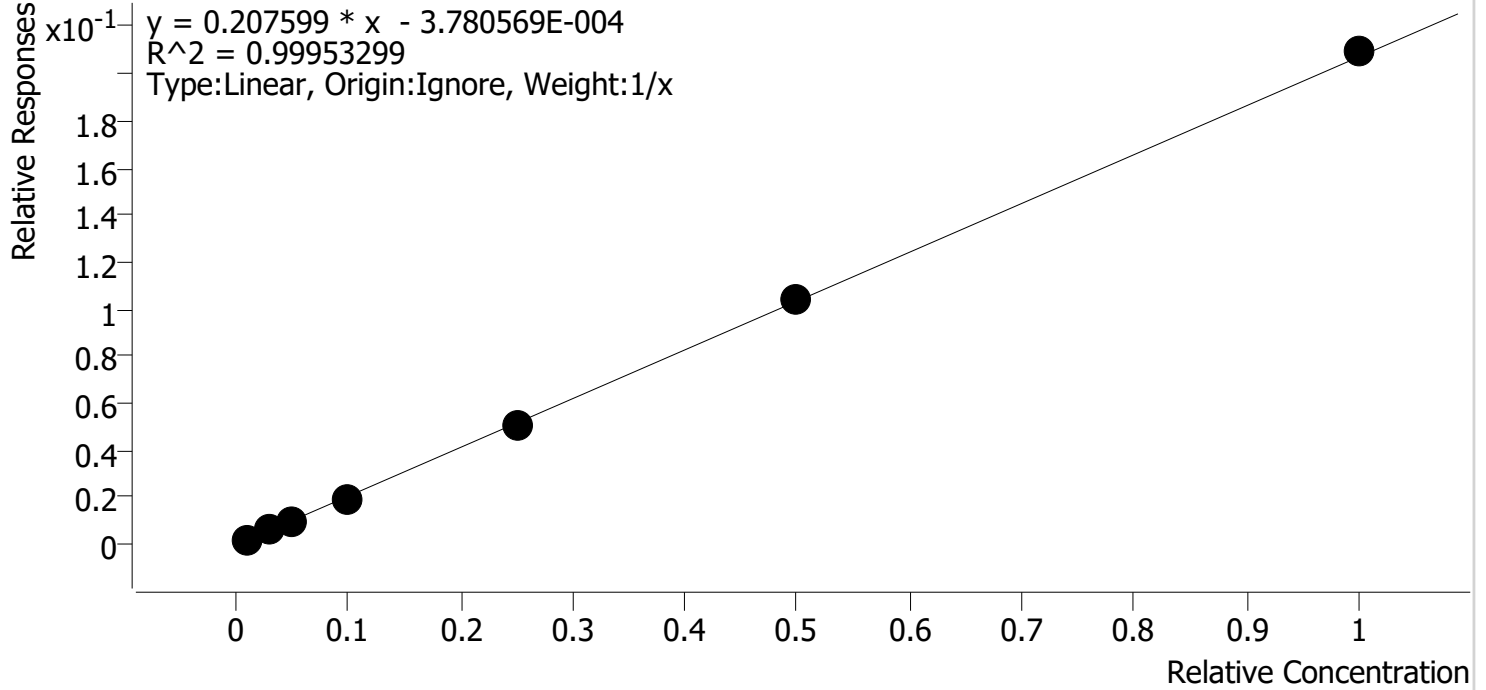
| Sample | Level | Enabled | Expected Concentration | Final Concentration | Accuracy |
|--------|-------|---------|------------------------|---------------------|----------|
| cal 1 | 1 | ✓ | 5.0 | 5.4 | 107.8 |
| cal 2 | 2 | ✓ | 10.0 | 10.4 | 103.6 |
| cal 3 | 3 | ✓ | 20.0 | 18.1 | 90.4 |
| cal 4 | 4 | ✓ | 50.0 | 48.5 | 97.1 |
| cal 5 | 5 | ✓ | 75.0 | 75.9 | 101.2 |
| cal-6 | 6 | ✓ | 100.0 | 98.6 | 98.6 |
| cal-7 | 7 | ✓ | 250.0 | 253.1 | 101.2 |

Compound Calibration Report



Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Last Cal. Update 1/14/2021 1:52 PM
Analyst Name ISP\datastor
Analyte THC-OH **Internal Standard** THC-OH-d3

THC-OH - 7 Levels, 7 Levels Used, 7 Points, 7 Points Used, 0 QCs



| Sample | Level | Enabled | Expected Concentration | Final Concentration | Accuracy |
|--------|-------|---------|------------------------|---------------------|----------|
| cal 1 | 1 | ✓ | 1.0 | 1.1 | 110.1 |
| cal 2 | 2 | ✓ | 3.0 | 2.9 | 98.1 |
| cal 3 | 3 | ✓ | 5.0 | 4.9 | 98.7 |
| cal 4 | 4 | ✓ | 10.0 | 9.5 | 94.5 |
| cal 5 | 5 | ✓ | 25.0 | 24.3 | 97.0 |
| cal-6 | 6 | ✓ | 50.0 | 50.3 | 100.5 |
| cal-7 | 7 | ✓ | 100.0 | 101.1 | 101.1 |

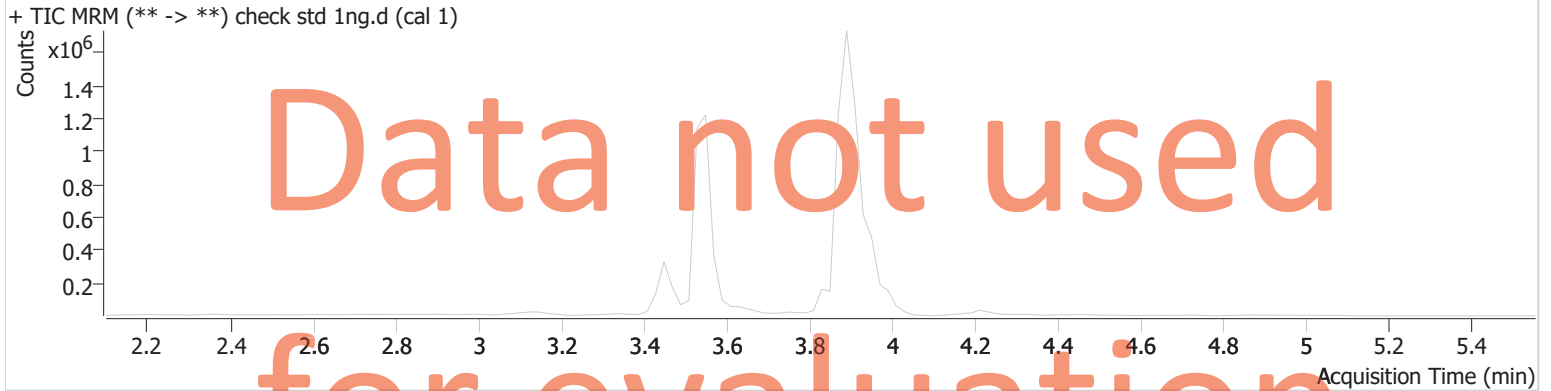
GA

AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|-----------------|
| Instrument | 69679 | Data File | check std 1ng.d |
| Type | Sample | Sample | cal 1 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-A1 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 9:38:42 AM | | |
| Sample Info. | | | |

Sample Chromatogram



| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|-------------------|-------|------------|------------------------|
| THC | 3.964 | 4886 | 605624 | 1.211 ng/ml Low |
| THC-COOH | 3.451 High | 40681 | 638335 | 4.816 ng/ml Low |
| THC-OH | 3.558 | 7076 | 3538109 | 1.145 ng/ml Low |

system had no equilibrated, sample was re-injected and evaluated.

GA

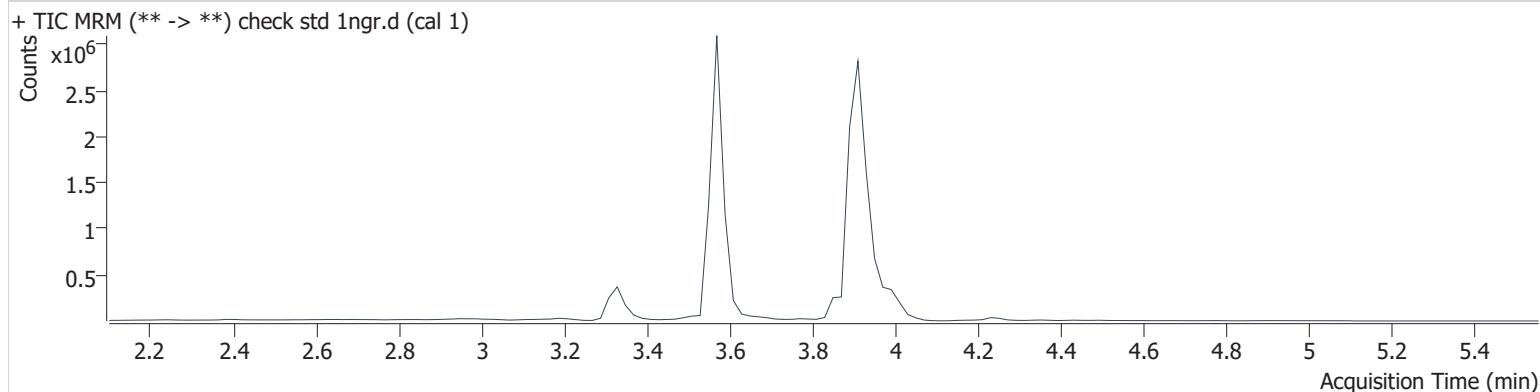
AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|------------------|
| Instrument | 69679 | Data File | check std 1ngr.d |
| Type | Cal | Sample | cal 1 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-A1 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 1:12:05 PM | | |

Sample Info.

Sample Chromatogram



| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|-------|-------|------------|------------------------|
| THC | 3.984 | 6187 | 497908 | 1.679 ng/ml Low |
| THC-COOH | 3.331 | 56841 | 796790 | 5.391 ng/ml Low |
| THC-OH | 3.578 | 13219 | 6926415 | 1.101 ng/ml Low |

GA

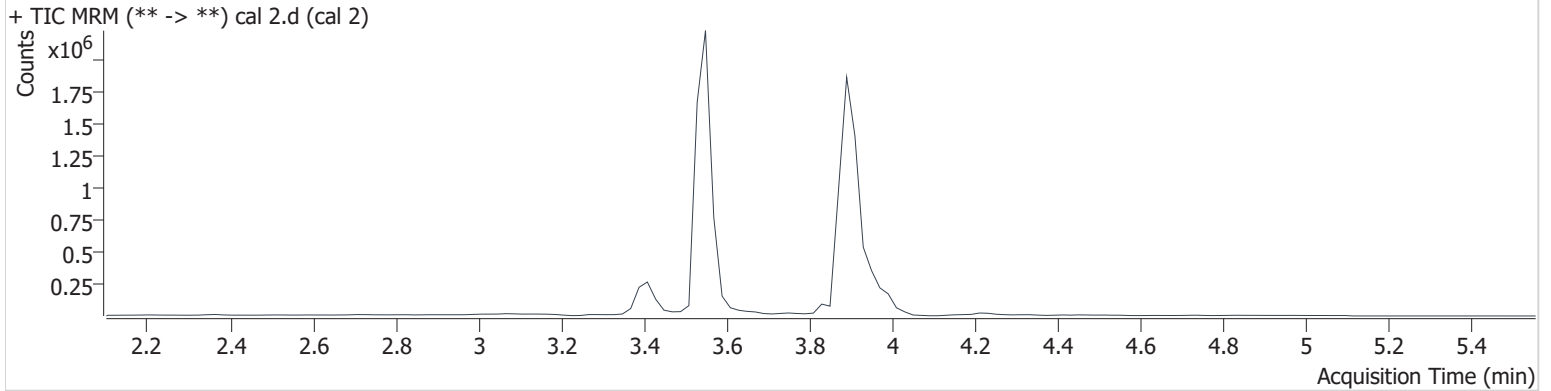
AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|-----------|
| Instrument | 69679 | Data File | cal 2.d |
| Type | Cal | Sample | cal 2 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-B1 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 9:45:20 AM | | |

Sample Info.

Sample Chromatogram



| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|-------|-------|------------|------------------------|
| THC | 3.964 | 12423 | 496697 | 3.030 ng/ml |
| THC-COOH | 3.411 | 81088 | 591305 | 10.364 ng/ml |
| THC-OH | 3.558 | 32462 | 5665179 | 2.942 ng/ml Low |

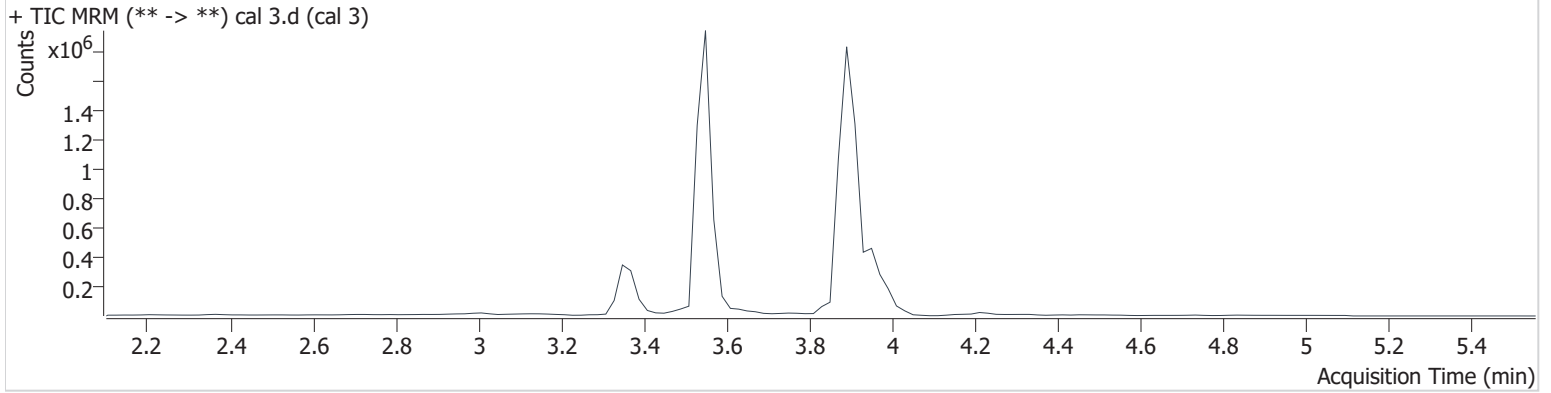
GA

AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|-----------|
| Instrument | 69679 | Data File | cal 3.d |
| Type | Cal | Sample | cal 3 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-C1 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 9:51:56 AM | | |
| Sample Info. | | | |

Sample Chromatogram



| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|-------|--------|------------|--------------|
| THC | 3.964 | 28751 | 645265 | 5.129 ng/ml |
| THC-COOH | 3.371 | 147244 | 615652 | 18.077 ng/ml |
| THC-OH | 3.558 | 45163 | 4577819 | 4.934 ng/ml |

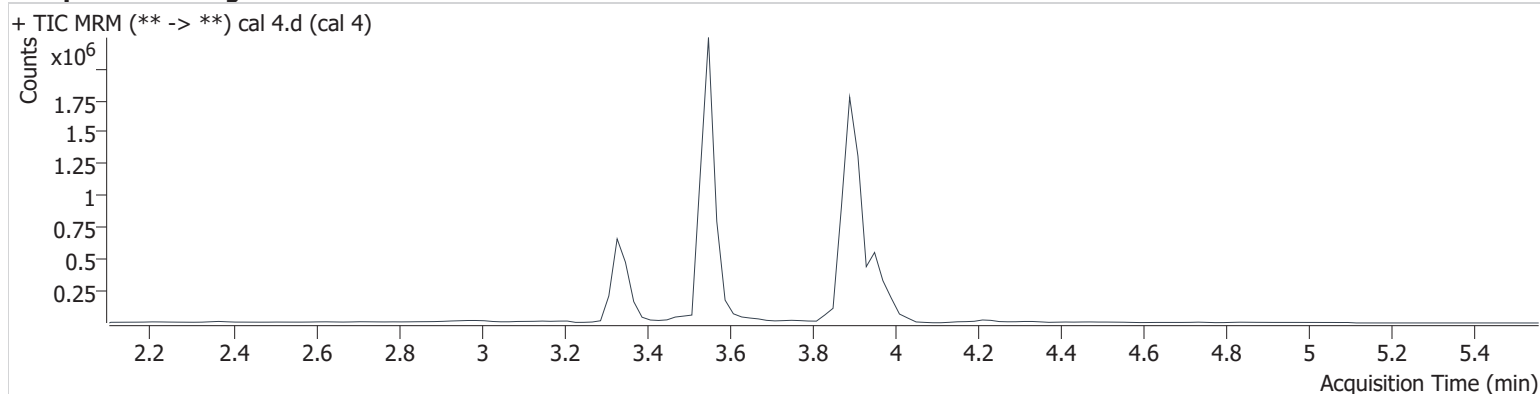
GA

AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|-----------|
| Instrument | 69679 | Data File | cal 4.d |
| Type | Cal | Sample | cal 4 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-D1 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 9:58:32 AM | | |
| Sample Info. | | | |

Sample Chromatogram



| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|-------|--------|------------|--------------|
| THC | 3.964 | 67677 | 771197 | 9.767 ng/ml |
| THC-COOH | 3.331 | 431632 | 672354 | 48.526 ng/ml |
| THC-OH | 3.558 | 88631 | 4606461 | 9.450 ng/ml |

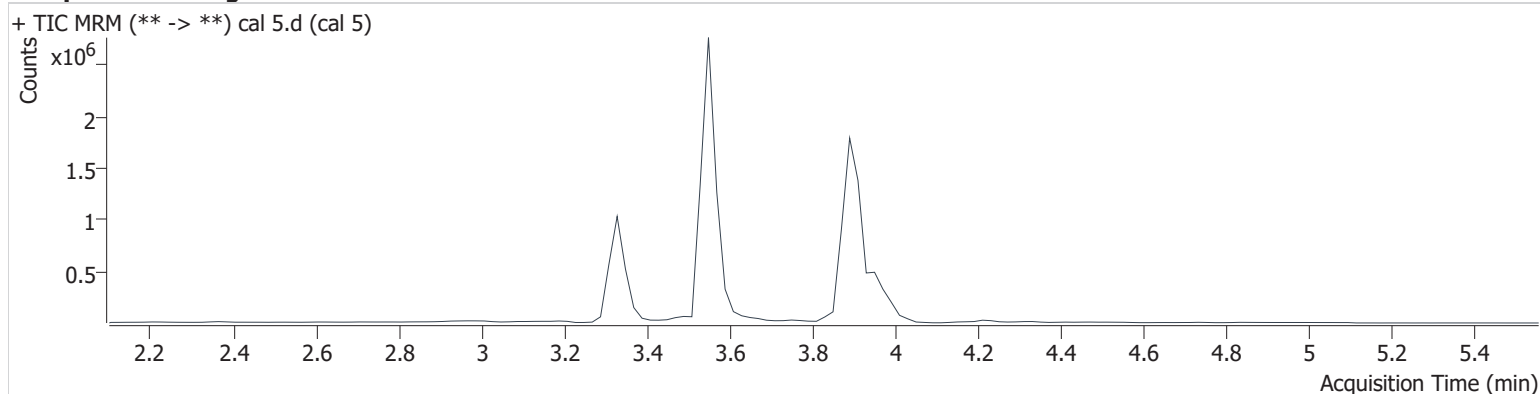
GA

AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|-----------|
| Instrument | 69679 | Data File | cal 5.d |
| Type | Cal | Sample | cal 5 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-E1 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 10:05:08 AM | | |
| Sample Info. | | | |

Sample Chromatogram



| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|-------|--------|------------|--------------|
| THC | 3.964 | 137579 | 608055 | 24.637 ng/ml |
| THC-COOH | 3.331 | 750019 | 746696 | 75.927 ng/ml |
| THC-OH | 3.558 | 242636 | 4854927 | 24.256 ng/ml |

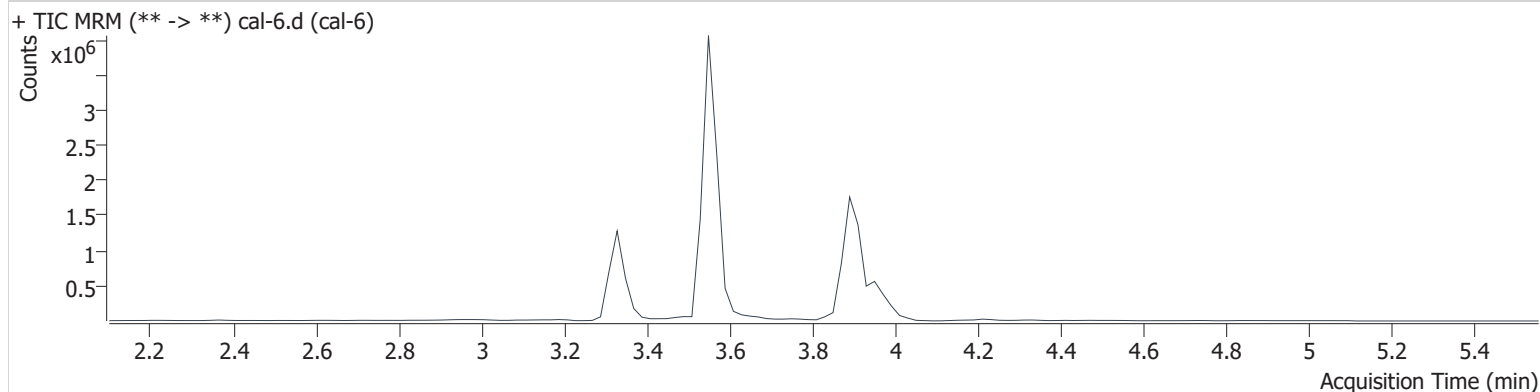
GA

AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|-----------|
| Instrument | 69679 | Data File | cal-6.d |
| Type | Cal | Sample | cal-6 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-F1 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 10:11:44 AM | | |
| Sample Info. | | | |

Sample Chromatogram



| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|-------|--------|------------|--------------|
| THC | 3.964 | 279250 | 606608 | 49.769 ng/ml |
| THC-COOH | 3.331 | 993964 | 761759 | 98.633 ng/ml |
| THC-OH | 3.558 | 555193 | 5341199 | 50.252 ng/ml |

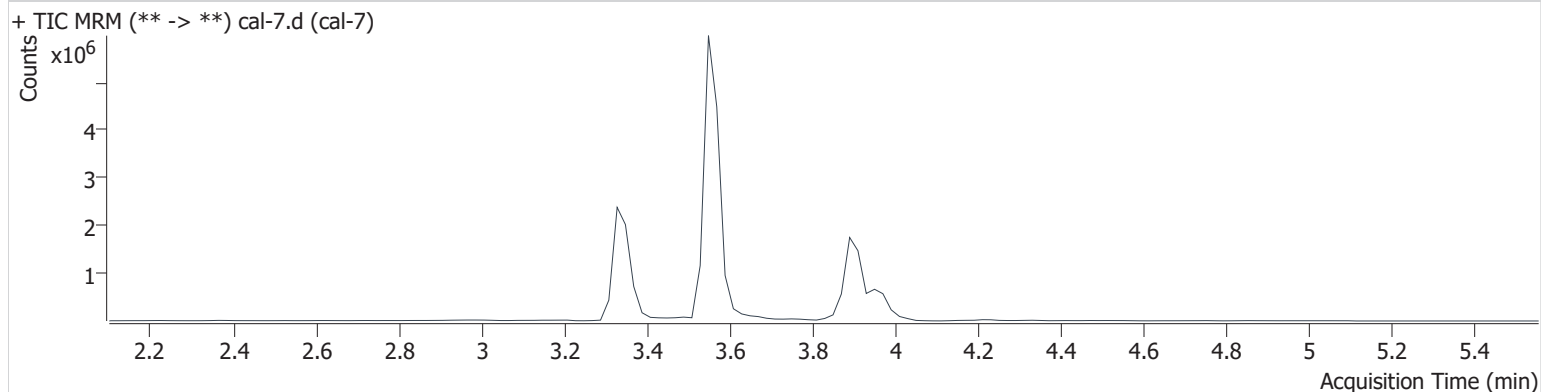
GA

AM #26 Cannabinoids Screen Results

Batch results D:\MassHunter\Data\2021\am 25-26\011121\QuantResults\cann screen.batch.bin
Calibration Last Update 1/14/2021 1:52:59 PM

| | | | |
|-------------------------|-------------------------|------------------|-----------|
| Instrument | 69679 | Data File | cal-7.d |
| Type | Cal | Sample | cal-7 |
| Acq. Method | am 26 cann scr 5-5-20.m | Operator | Anne Nord |
| Sample Position | P3-G1 | Comment | |
| Injection Volume | 5 | | |
| Acq. Date-Time | 1/14/2021 10:18:20 AM | | |
| Sample Info. | | | |

Sample Chromatogram



| Name | RT | Resp. | ISTD Resp. | Final Conc. |
|----------|-------|---------|------------|---------------|
| THC | 3.964 | 580844 | 621595 | 100.669 ng/ml |
| THC-COOH | 3.331 | 2378045 | 710283 | 253.082 ng/ml |
| THC-OH | 3.558 | 1120416 | 5349897 | 101.063 ng/ml |