



















**REVIEWED**  
By Sarah Collins at 11:16 am, Aug 23, 2023



**Worklist: 6471**

REVIEWED  
By Anthony Wylie at 2:21 pm, Aug 23, 2023

| <u>LAB_CASE</u> | <u>ITEM</u> | <u>ITEM_TYPE</u> | <u>DESCRIPTION</u>                               |   |
|-----------------|-------------|------------------|--|---|
| C2023-1725      | 1           | UCK              | AM 25/AM 26 Urine MultiDrug/THC Screen by LC-QQQ |    |
| C2023-1735      | 1           | UCK              | AM 25/AM 26 Urine MultiDrug/THC Screen by LC-QQQ |    |
| C2023-1739      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |    |
| C2023-1741      | 3           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |    |
| C2023-1748      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |    |
| C2023-1750      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |    |
| C2023-1766      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |    |
| C2023-1767      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |    |
| C2023-1779      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |    |
| C2023-1815      | 3           | UCK              | AM 25/AM 26 Urine MultiDrug/THC Screen by LC-QQQ |   |
| C2023-1816      | 1           | COBCK            | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2023-1821      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2023-1832      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2023-1874      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2023-1875      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2023-1890      | 1           | UCK              | AM 25/AM 26 Urine MultiDrug/THC Screen by LC-QQQ |  |
| C2023-1904      | 1           | BCK              | AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ |  |
| C2023-1906      | 1           | UCK              | AM 25/AM 26 Urine MultiDrug/THC Screen by LC-QQQ |  |

C2023-1449-1, C2023-1726-1 and M2023-2977-2 were also included with the batch for AM 25.

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

10/4/23  
\*

Extraction Date: 08/17/23 Analyst: Anne Nord

Plate lot#: 230712

Plate retest date: 1/12/2023<sup>4</sup>

**Mobile phase A:** 10mM Ammonium Formate  
0.5M Ammonium Hydroxide

**Mobile phase B:** 0.1% Formic Acid in MeOH  
Ethyl Acetate LC 20% Methanol

**Blank Blood Lot:** 23C57106 **Blank Urine lot:** 8423

**Column:** Agilent Phenyl Hexyl (4.6x50mm, 2.7um)

**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: 390993**
- 3. Pipette **250 µL of 0.5 M ammonium hydroxide** in wells of analytical plate.
- 4. Place on shaking incubator at ambient temp., 900 rpm for 15 minutes.
- 5. Transfer **300 µL of blood or urine+base** mixture to corresponding wells of SLE+ plate.
- 6. 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*
- 7. Wait 5 minutes.
- 8. Add **900 µL ethyl acetate**.
- 9. Wait 5 minutes.
- 10. Apply positive pressure for approx. 10-15 seconds. *(12-15 PSI- Selector to the left)*.
- 11. Add **900 µL ethyl acetate**.
- 12. Wait 5 minutes.
- 13. Apply positive pressure for approx. 10-15 seconds. *(12-15 PSI- Selector to the left)*.
- 14. 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*
- 15. 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:

*When the run was started some compounds were outside the acquisition window, the acquisition method was updated and the samples that had been run were re-injected and evaluated.*

|   | 1                | 2      | 3              | 4 | 5 | 6 | 7 | 8 | 9 | 10 | 11 | 12     |
|---|------------------|--------|----------------|---|---|---|---|---|---|----|----|--------|
| A | Cal 1            | 1741-3 | negative urine |   |   |   |   |   |   |    |    | 1725-1 |
| B |                  | 1748-1 | m2023-2977-2   |   |   |   |   |   |   |    |    | 1815-3 |
| C | internal control | 1750-1 | 1874-1         |   |   |   |   |   |   |    |    | 1890.1 |
| D |                  | 1766-1 | 1779-1         |   |   |   |   |   |   |    |    | 1735-1 |
| E | negative blood   | 1767-1 | 1875-1         |   |   |   |   |   |   |    |    |        |
| F | 1449-1           | 1816-1 | 1904-1         |   |   |   |   |   |   |    |    |        |
| G | 1726-1           | 1821-1 | 1906-1         |   |   |   |   |   |   |    |    |        |
| H | 1739-1           | 1832-1 |                |   |   |   |   |   |   |    |    |        |

C2023-\_\_\_\_-

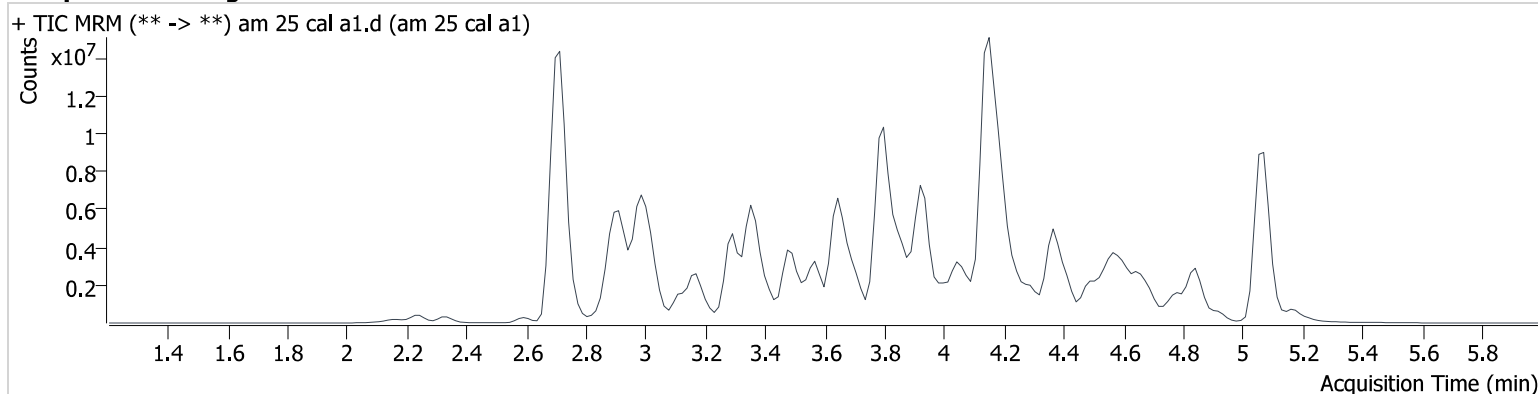
plate position 2

# AM #25 Multi-Drug Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\mds.batch.bin  
**Calibration Last Update** 8/22/2023 2:43:42 PM

**Instrument** 69679 **Data File** am 25 cal a1.d  
**Type** Cal **Sample** am 25 cal a1  
**Acq. Method** mds713.m **Operator** Anne Nord  
**Sample Position** P2-A1 **Comment**  
**Injection Volume** 2.5  
**Acq. Date-Time** 8/17/2023 4:50:12 PM  
**Sample Info.**

## Sample Chromatogram



| Name                   | RT    | Resp.   | S/N       | S/N      | ISTD Resp. | Calc. Conc. |
|------------------------|-------|---------|-----------|----------|------------|-------------|
| 10-OH-Carbamazepine    | 3.851 | 696801  | 7007.0    | 30.9     | 649102     | 10.000      |
| 6-MAM                  | 2.847 | 27926   | 40829.3   | 11698.0  | 1051678    | 10.000      |
| 7-aminoclonazepam      | 3.586 | 169731  | 77306.3   | 157459.7 | 1487527    | 10.000      |
| 7-aminoflunitrazepam   | 3.817 | 729732  | 601.8     | 100140.3 | 1487527    | 10.000      |
| 9-Hydroxyrisperidone   | 3.933 | 3454243 | 1143198.4 | 326948.1 | 1487527    | 10.000      |
| Acetyl Fentanyl        | 3.798 | 183640  | 938.8     | 28404.0  | 8707231    | 10.000      |
| Acetyl Norfentanyl     | 2.887 | 156281  | 243028.5  | 53.0     | 14140534   | 10.000      |
| a-hydroxyalprazolam    | 4.690 | 83290   | 72.6      | 17706.6  | 649102     | 10.000      |
| alpha-hydroxymidazolam | 4.643 | 822879  | 2462.2    | 294.9    | 3651250    | 10.000      |
| alpha-PHP              | 3.852 | 1983135 | 8365.6    | 728.8    | 3991151    | 10.000      |
| alpha-PVP              | 3.546 | 2416456 | 1867562.1 | 853.2    | 3991151    | 10.000      |
| Alprazolam             | 4.784 | 1282649 | 6445.2    | 670.7    | 3651250    | 10.000      |
| Amitriptyline          | 4.603 | 770421  | 301.6     | 206.4    | 2552751    | 10.000      |
| Amphetamine            | 2.922 | 1225714 | 328.0     | 1093.1   | 3991151    | 10.000      |
| Benzoylcegonine        | 3.447 | 31887   | 2200.9    | 298.8    | 111787     | 10.000      |
| Brompheniramine        | 4.199 | 67057   | 102730.9  | 5658.9   | 6022282    | 10.000      |
| Buprenorphine          | 4.132 | 2786    | 1828.7    | 483.6    | 1291070    | 10.000      |
| Bupropion              | 3.822 | 2426948 | 825.5     | 1166.8   | 10192212   | 10.000      |
| Carbamazepine          | 4.361 | 4218803 | 61407.8   | 1021.2   | 3630909    | 10.000      |
| Carisoprodol           | 4.298 | 540408  | 374.0     | 314.1    | 2809042    | 10.000      |
| Chlordiazepoxide       | 4.786 | 355431  | 162007.9  | 135.6    | 3651250    | 10.000      |
| Chlorpheniramine       | 4.079 | 3792793 | 57045.2   | 141.9    | 6022282    | 10.000      |
| Chlorpromazine         | 4.828 | 498156  | 7462.2    | 1096.6   | 1945573    | 10.000      |
| Citalopram             | 4.245 | 1943287 | 2868.2    | 801072.2 | 37188270   | 10.000      |
| Clomipramine           | 4.844 | 562220  | 148446.6  | 578.1    | 869726     | 10.000      |
| Clonazepam             | 4.630 | 212103  | 91673.5   | 232.7    | 38808      | 10.000      |
| Clonazepam             | 4.519 | 364159  | 91650.0   | 33126.2  | 649102     | 10.000      |
| clozapine              | 4.366 | 2082238 | 9223.8    | 367730.6 | 9052033    | 10.000      |
| Cocaehtylene           | 3.861 | 2539414 | 1211413.2 | 806.8    | 12391593   | 10.000      |
| Cocaine                | 3.646 | 2540463 | 1433.2    | 251.9    | 12391593   | 10.000      |
| Codeine                | 2.713 | 183762  | 26956.0   | 57934.4  | 3630909    | 10.000      |
| Cyclobenzaprine        | 4.527 | 1361536 | 3129.3    | 169.1    | 2552751    | 10.000      |
| Desipramine            | 4.574 | 1777570 | 707592.5  | 1692.5   | 2552751    | 10.000      |
| Dextromethorphan       | 4.188 | 1120554 | 246216.8  | 230519.4 | 6022282    | 10.000      |



# AM #25 Multi-Drug Screen Results

| Name                   | RT    | Resp.   | S/N           | S/N       | ISTD Resp. | Calc. Conc. |
|------------------------|-------|---------|---------------|-----------|------------|-------------|
| Dextrorphan            | 3.420 | 1790225 | 1340.1        | 376232.0  | 3991151    | 10.000      |
| Diazepam               | 5.047 | 581873  | 487.3         | 394.0     | 3651250    | 10.000      |
| Dihydrocodeine         | 2.682 | 659445  | 3150.9        | 195534.5  | 3630909    | 10.000      |
| Dimethyltryptamine     | 2.995 | 1153141 | 633.6         | 11698.0   | 3991151    | 10.000      |
| Diphenhydramine        | 4.159 | 6980912 | 1078.4        | 768.5     | 37188270   | 10.000      |
| Doxepin                | 4.311 | 1032073 | 500.2         | 49.8      | 9052033    | 10.000      |
| Doxylamine             | 3.665 | 4458231 | 565.2         | 183.4     | 3991151    | 10.000      |
| Duloxetine             | 4.525 | 22337   | 11312.3       | 85747.7   | 869726     | 10.000      |
| EDDP                   | 4.203 | 385021  | 85176.5       | 44339.5   | 1616248    | 10.000      |
| Estazolam              | 4.694 | 2018169 | 2734864.8     | 358890.0  | 3651250    | 10.000      |
| Etizolam               | 4.765 | 99853   | 65885.9       | 165727.3  | 3651250    | 10.000      |
| Fentanyl               | 4.059 | 128459  | 76.2          | 83.0      | 8269941    | 10.000      |
| Flualprazolam          | 4.597 | 447401  | 123121.5      | 2433096.2 | 3651250    | 10.000      |
| Flunitrazepam          | 4.737 | 978995  | 359251.5      | 102258.8  | 649102     | 10.000      |
| Fluorofentanyl         | 4.073 | 103780  | 48689.3       | 135.1     | 8269941    | 10.000      |
| Fluoxetine             | 4.508 | 936617  | 1032.7        | 154.6     | 869726     | 10.000      |
| Flurazepam             | 4.225 | 1683019 | 311751.6      | 243178.5  | 1291070    | 10.000      |
| Hydrocodone            | 2.942 | 592980  | 779.7         | 221.4     | 3630909    | 10.000      |
| Hydromorphone          | 2.337 | 606665  | 613.4         | 665.3     | 137270     | 10.000      |
| hydroxyzine            | 4.656 | 2044124 | 205.5         | 828225.5  | 9052033    | 10.000      |
| Imipramine             | 4.572 | 2566436 | 1482.9        | 1289.0    | 2552751    | 10.000      |
| Ketamine               | 3.391 | 1532822 | 3861.0        | 599.4     | 5994433    | 10.000      |
| Lamotrigine            | 3.574 | 120122  | 66815.7       | 3077.6    | 3991151    | 10.000      |
| Levamisole             | 2.887 | 1286084 | 845936.1      | 511.8     | 12391593   | 10.000      |
| Levetiracetam          | 2.601 | 292781  | 774.3         | 359.3     | 1487527    | 10.000      |
| Lorazepam              | 4.598 | 18917   | 4990287694520 | ∞         | 649102     | 10.000      |
|                        |       |         | 68.0          |           |            |             |
| Maprotiline            | 4.603 | 511756  | 535.2         | 169.8     | 2552751    | 10.000      |
| MDA                    | 3.042 | 1280370 | 816.8         | 354.8     | 14319543   | 10.000      |
| MDEA                   | 3.286 | 2415980 | 2946.8        | 6885.1    | 14319543   | 10.000      |
| MDMA                   | 3.118 | 2205154 | 974.9         | 434.0     | 14319543   | 10.000      |
| Meperidine             | 3.651 | 1032932 | 2777.8        | 59595.2   | 137270     | 10.000      |
| Meprobamate            | 3.716 | 194863  | 368.0         | 207.6     | 2809042    | 10.000      |
| Methadone              | 4.538 | 3638198 | 786260.7      | 7106.4    | 8707231    | 10.000      |
| Methamphetamine        | 3.028 | 1682415 | ∞             | ∞         | 14319543   | 10.000      |
| Methocarbamol          | 3.667 | 93944   | 2302.1        | 17.1      | 2809042    | 10.000      |
| Methylphenidate        | 3.576 | 4871937 | 1340.8        | 8275.6    | 6994986    | 10.000      |
| Metoprolol             | 3.495 | 543710  | 5345.7        | 509836.1  | 3991151    | 10.000      |
| Midazolam              | 4.475 | 281744  | 4617.5        | 38139.5   | 1487527    | 10.000      |
| Mirtazapine            | 3.712 | 1402647 | 1752.8        | 1565.3    | 1291070    | 10.000      |
| Mitragynine            | 4.225 | 228591  | 92535.0       | 331594.0  | 8269941    | 10.000      |
| Morphine               | 2.156 | 174310  | 1794.8        | 213.1     | 137270     | 10.000      |
| Norbuprenorphine       | 3.902 | 37934   | 18279.2       | 21661.4   | 1291070    | 10.000      |
| Nordiazepam            | 4.896 | 186055  | 74230.5       | ∞         | 3651250    | 10.000      |
| Norfentanyl            | 3.376 | 2722803 | 3912.0        | 410.8     | 14140534   | 10.000      |
| Norhydrocodone         | 2.943 | 121266  | 318.1         | 16726.6   | 3630909    | 10.000      |
| norketamine            | 3.377 | 180127  | 173.2         | 1436.4    | 5994433    | 10.000      |
| Normeperidine          | 3.684 | 1423642 | 694.2         | 266.6     | 137270     | 10.000      |
| Noroxycodone           | 2.911 | 551621  | ∞             | 46.3      | 3630909    | 10.000      |
| Nortriptyline          | 4.621 | 596933  | 141001.5      | 127089.0  | 869726     | 10.000      |
| O-desmethyl-tramadol   | 2.915 | 3937654 | 987.7         | 1197.0    | 4627864    | 10.000      |
| O-Desmethylvenlafaxine | 3.296 | 1156384 | 1648.4        | 10184.7   | 4627864    | 10.000      |
| Olanzapine             | 3.215 | 1079603 | 662.9         | 200.5     | 869726     | 10.000      |
| Oxazepam               | 4.710 | 100845  | 166.6         | 44.7      | 649102     | 10.000      |
| Oxycodone              | 2.909 | 1305002 | 599.8         | 94211.1   | 5994433    | 10.000      |
| Oxymorphone            | 2.242 | 698742  | 183.6         | 3108.8    | 137270     | 10.000      |
| Paroxetine             | 4.535 | 144599  | 523.8         | 13398.2   | 869726     | 10.000      |
| Phenazepam             | 4.826 | 238862  | 82691.7       | 53281.6   | 3651250    | 10.000      |
| Phencyclidine          | 4.006 | 3003827 | 4599.9        | 393.4     | 4627864    | 10.000      |



# AM #25 Multi-Drug Screen Results

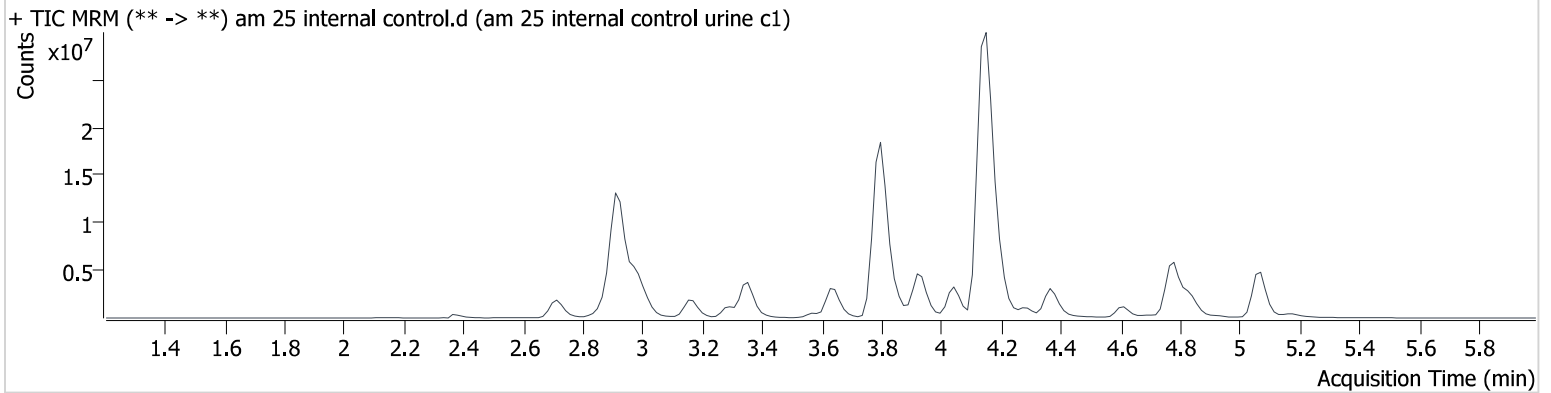
| Name            | RT    | Resp.    | S/N       | S/N      | ISTD Resp. | Calc. Conc. |
|-----------------|-------|----------|-----------|----------|------------|-------------|
| Phentermine     | 3.196 | 610534   | ∞         | 457.5    | 6994986    | 10.000      |
| Phenytoin       | 4.252 | 113567   | 14861.8   | 107.9    | 38808      | 10.000      |
| primidone       | 3.516 | 102123   | 64169.6   | 68.5     | 38808      | 10.000      |
| Promethazine    | 4.480 | 2248362  | 427556.9  | 270.7    | 2552751    | 10.000      |
| Pseudoephedrine | 2.722 | 44624991 | 3072.2    | 37187.4  | 6994986    | 10.000      |
| Quetiapine      | 4.410 | 2942913  | 3405.9    | 6565.1   | 6022282    | 10.000      |
| Risperidone     | 4.102 | 2826642  | 2273.7    | 437.2    | 6022282    | 10.000      |
| Sertraline      | 4.800 | 162844   | 1434.4    | 835.7    | 869726     | 10.000      |
| Sufentanil      | 4.333 | 84341    | 37603.8   | 142.2    | 8269941    | 10.000      |
| Tapentadol      | 3.500 | 2758214  | 875.5     | 1275.2   | 5994433    | 10.000      |
| Temazepam       | 4.862 | 818333   | 401.4     | 48.9     | 3651250    | 10.000      |
| Topiramate      | 3.905 | 5736     | 2044.4    | 1639.1   | 34375      | 10.000      |
| Tramadol        | 3.481 | 8744491  | ∞         | 113.0    | 1051678    | 10.000      |
| Trazodone       | 4.195 | 1908245  | 7010.4    | 2689.1   | 8707231    | 10.000      |
| Venlafaxine     | 3.910 | 4055151  | 49389.5   | 975.2    | 4627864    | 10.000      |
| Zaleplon        | 4.494 | 966054   | 298495.8  | 211738.9 | 649102     | 10.000      |
| Zolpidem        | 3.800 | 3639757  | 2076939.6 | 720.9    | 18204405   | 10.000      |
| Zopiclone       | 3.888 | 177201   | 84853.1   | ∞        | 851211     | 10.000      |

# AM #25 Multi-Drug Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\mds.batch.bin  
**Calibration Last Update** 8/22/2023 2:43:42 PM

|                         |                      |                  |                                 |
|-------------------------|----------------------|------------------|---------------------------------|
| <b>Instrument</b>       | 69679                | <b>Data File</b> | am 25 internal control.d        |
| <b>Type</b>             | Sample               | <b>Sample</b>    | am 25 internal control urine c1 |
| <b>Acq. Method</b>      | mds713.m             | <b>Operator</b>  | Anne Nord                       |
| <b>Sample Position</b>  | P2-C1                | <b>Comment</b>   |                                 |
| <b>Injection Volume</b> | 2.5                  |                  |                                 |
| <b>Acq. Date-Time</b>   | 8/17/2023 4:57:05 PM |                  |                                 |
| <b>Sample Info.</b>     |                      |                  |                                 |

## Sample Chromatogram



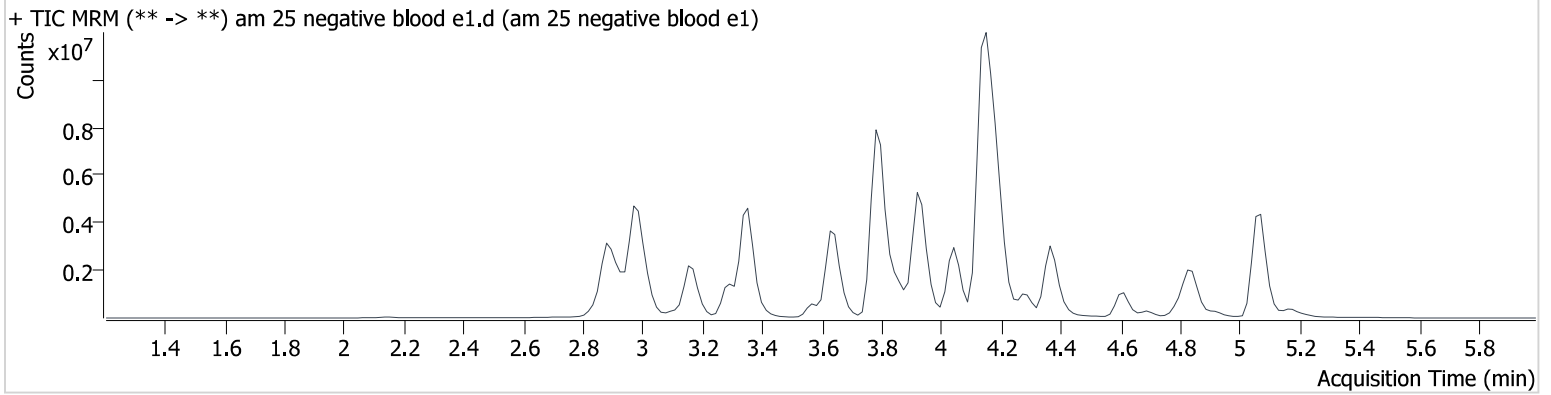
| Name            | RT    | Resp.    | S/N        | S/N           | ISTD Resp. | Calc. Conc. |
|-----------------|-------|----------|------------|---------------|------------|-------------|
| Alprazolam      | 4.769 | 9233581  | 1267.1     | 2023.3        | 3200631    | 82.124      |
| Amphetamine     | 2.922 | 10506853 | 1145.8     | 28321.5       | 3512445    | 97.403      |
| Codeine         | 2.713 | 2045151  | 2124.6     | 2146643.<br>8 | 3400504    | 118.834     |
| Diphenhydramine | 4.159 | 50462058 | 1395.0     | 5311.0        | 33723721   | 79.712      |
| Zolpidem        | 3.800 | 34221910 | 11970368.8 | 3585.6        | 14485124   | 118.164     |

# AM #25 Multi-Drug Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\mds.batch.bin  
**Calibration Last Update** 8/22/2023 2:43:42 PM

|                         |                      |                  |                           |
|-------------------------|----------------------|------------------|---------------------------|
| <b>Instrument</b>       | 69679                | <b>Data File</b> | am 25 negative blood e1.d |
| <b>Type</b>             | Sample               | <b>Sample</b>    | am 25 negative blood e1   |
| <b>Acq. Method</b>      | mds713.m             | <b>Operator</b>  | Anne Nord                 |
| <b>Sample Position</b>  | P2-E1                | <b>Comment</b>   |                           |
| <b>Injection Volume</b> | 2.5                  |                  |                           |
| <b>Acq. Date-Time</b>   | 8/17/2023 5:03:48 PM |                  |                           |
| <b>Sample Info.</b>     |                      |                  |                           |

## Sample Chromatogram



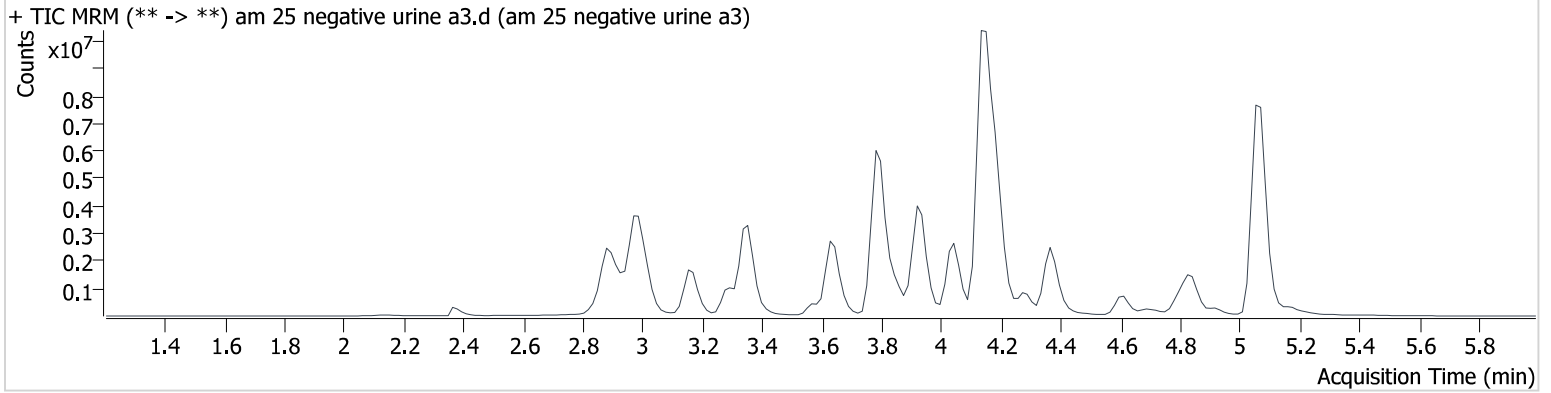


# AM #25 Multi-Drug Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\mds.batch.bin  
**Calibration Last Update** 8/22/2023 2:43:42 PM

|                         |                      |                  |                           |
|-------------------------|----------------------|------------------|---------------------------|
| <b>Instrument</b>       | 69679                | <b>Data File</b> | am 25 negative urine a3.d |
| <b>Type</b>             | Sample               | <b>Sample</b>    | am 25 negative urine a3   |
| <b>Acq. Method</b>      | mds713.m             | <b>Operator</b>  | Anne Nord                 |
| <b>Sample Position</b>  | P2-A3                | <b>Comment</b>   |                           |
| <b>Injection Volume</b> | 2.5                  |                  |                           |
| <b>Acq. Date-Time</b>   | 8/17/2023 6:24:12 PM |                  |                           |
| <b>Sample Info.</b>     |                      |                  |                           |

## Sample Chromatogram





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

Extraction Date: 8/17/23 Analyst: Anne Nord

Plate lot#: 230627 Plate retest date: 12/27/2023

**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:** 23C57106 **Urine Blank:** 8423

**Column:** Agilent Phenyl Hexyl (4.6x50mm: 2.7 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: c2023-1904-1 and C2023-1725-1 dried out and did not inject. The were reconstituted and injected the next morning 8/18/23.  
that injection was evaluated.

|   | 1                           | 2                         | 3              | 4      | 5 | 6 |
|---|-----------------------------|---------------------------|----------------|--------|---|---|
| a | cal 1                       | Internal control<br>urine | 1821-1         | 1906.1 |   |   |
| b | cal 2                       | negative blood            | 1832-1         | 1735-1 |   |   |
| c | cal 3                       | 1739-1                    | 1874-1         | 1815-3 |   |   |
| d | cal 4                       | 1741-3                    | 1779-1         | 1890-1 |   |   |
| e | cal 5                       | 1750-1                    | 1875-1         | 1748.1 |   |   |
| f | cal 6                       | 1766-1                    | 1904-1         |        |   |   |
| g | cal 7                       | 1767-1                    | negative urine |        |   |   |
| h | Internal control<br>(blood) | 1816-1                    | 1725-1         |        |   |   |

Plate position 3

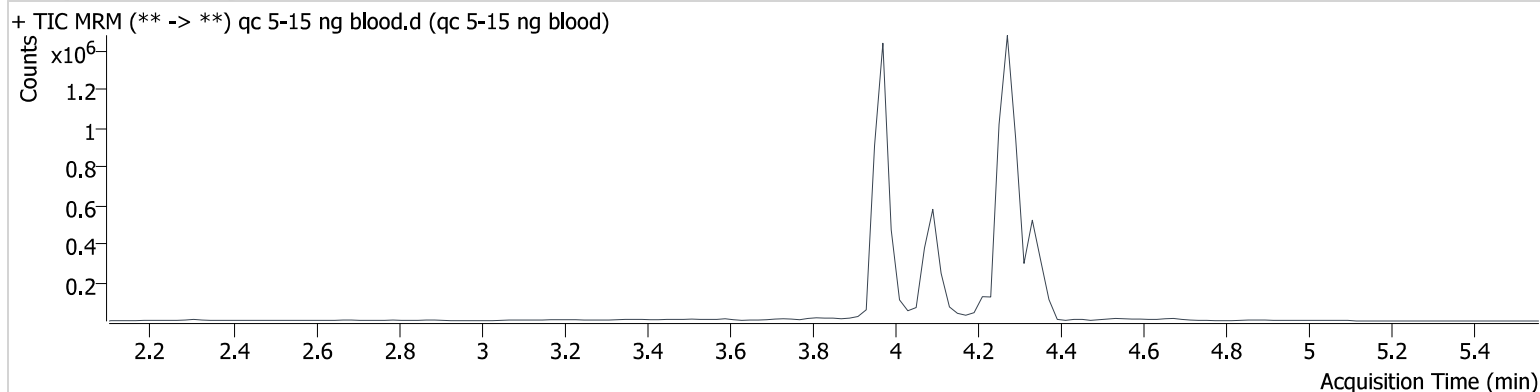
c2023-\_\_\_\_-\_\_

# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |                    |
|-------------------------|-------------------------|------------------|--------------------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | qc 5-15 ng blood.d |
| <b>Type</b>             | QC                      | <b>Sample</b>    | qc 5-15 ng blood   |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord          |
| <b>Sample Position</b>  | P3-H1                   | <b>Comment</b>   |                    |
| <b>Injection Volume</b> | 5                       |                  |                    |
| <b>Acq. Date-Time</b>   | 8/17/2023 9:04:07 PM    |                  |                    |
| <b>Sample Info.</b>     |                         |                  |                    |

## Sample Chromatogram



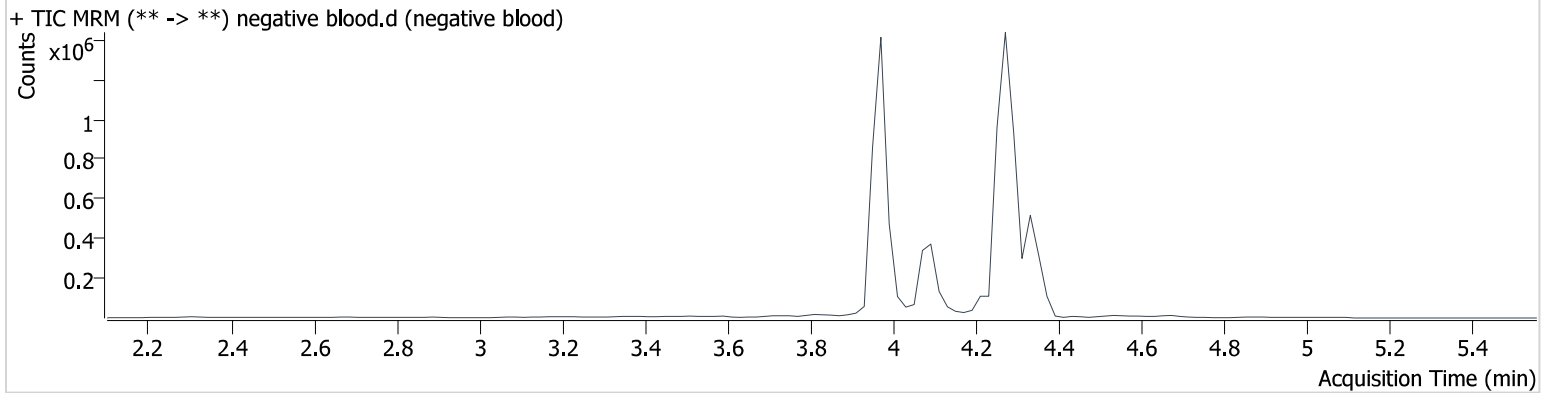
| Name     | RT    | Resp.  | ISTD Resp. | Final Conc.  |
|----------|-------|--------|------------|--------------|
| THC      | 4.345 | 19739  | 684804     | 3.867 ng/ml  |
| THC-COOH | 4.093 | 195858 | 889136     | 15.222 ng/ml |
| THC-OH   | 3.979 | 23736  | 3353179    | 4.606 ng/ml  |

# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |                  |
|-------------------------|-------------------------|------------------|------------------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | negative blood.d |
| <b>Type</b>             | Sample                  | <b>Sample</b>    | negative blood   |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord        |
| <b>Sample Position</b>  | P3-B2                   | <b>Comment</b>   |                  |
| <b>Injection Volume</b> | 5                       |                  |                  |
| <b>Acq. Date-Time</b>   | 8/17/2023 9:10:35 PM    |                  |                  |
| <b>Sample Info.</b>     |                         |                  |                  |

## Sample Chromatogram

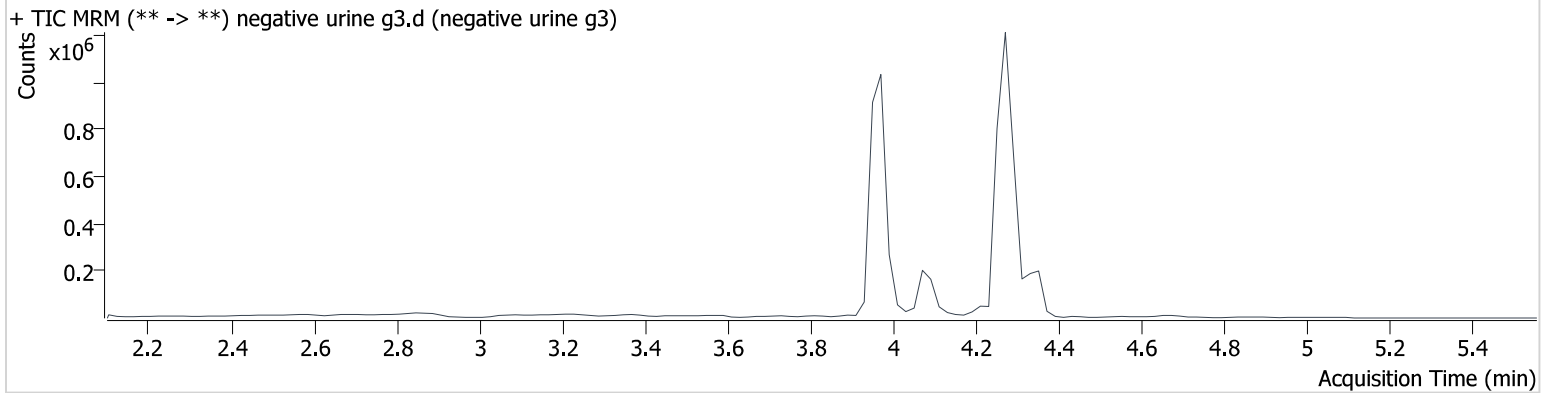


# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |                     |
|-------------------------|-------------------------|------------------|---------------------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | negative urine g3.d |
| <b>Type</b>             | Sample                  | <b>Sample</b>    | negative urine g3   |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord           |
| <b>Sample Position</b>  | P3-G3                   | <b>Comment</b>   |                     |
| <b>Injection Volume</b> | 5                       |                  |                     |
| <b>Acq. Date-Time</b>   | 8/17/2023 10:34:40 PM   |                  |                     |
| <b>Sample Info.</b>     |                         |                  |                     |

## Sample Chromatogram

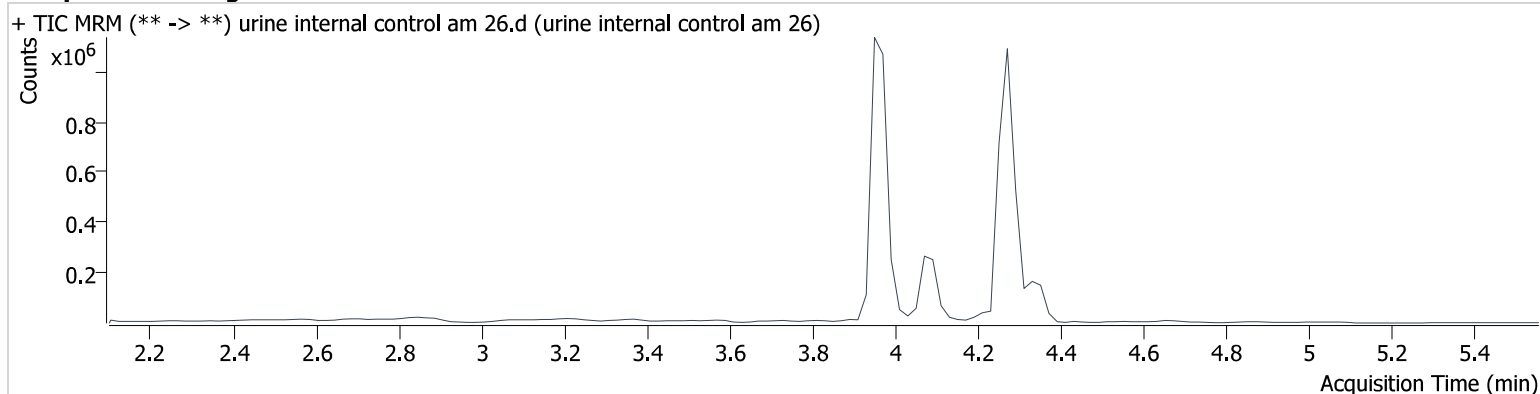


# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |                                |
|-------------------------|-------------------------|------------------|--------------------------------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | urine internal control am 26.d |
| <b>Type</b>             | Sample                  | <b>Sample</b>    | urine internal control am 26   |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord                      |
| <b>Sample Position</b>  | P3-A2                   | <b>Comment</b>   |                                |
| <b>Injection Volume</b> | 5                       |                  |                                |
| <b>Acq. Date-Time</b>   | 8/17/2023 11:19:56 PM   |                  |                                |
| <b>Sample Info.</b>     |                         |                  |                                |

## Sample Chromatogram



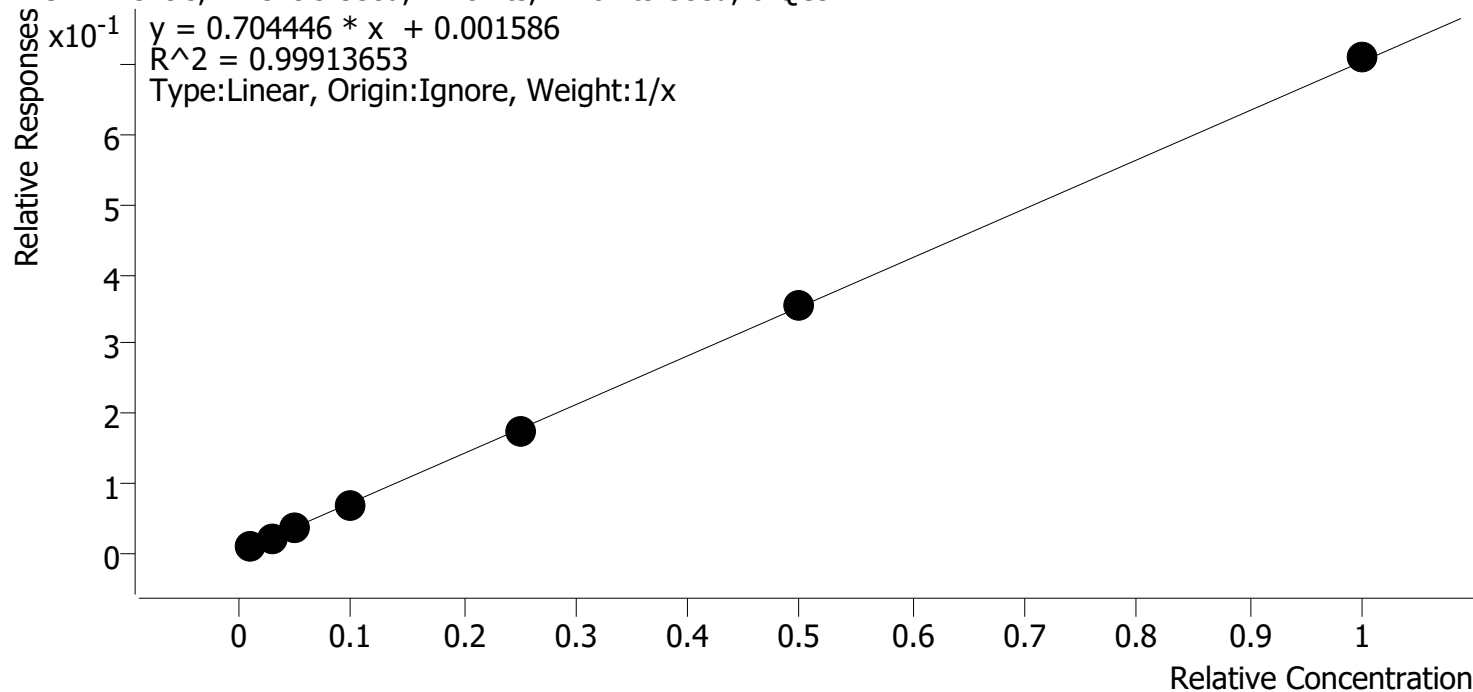
| Name     | RT    | Resp. | ISTD Resp. | Final Conc.  |
|----------|-------|-------|------------|--------------|
| THC      | 4.345 | 8092  | 246358     | 4.438 ng/ml  |
| THC-COOH | 4.093 | 95579 | 477439     | 13.845 ng/ml |
| THC-OH   | 3.979 | 19431 | 2944843    | 4.292 ng/ml  |

# Compound Calibration Report



**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Last Cal. Update** 8/21/2023 10:41 AM  
**Analyst Name** ISP\datastor  
**Analyte** THC **Internal Standard** THC-d3

THC - 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                    | 1.2                 | 121.9    |
| cal 2  | 2     | ✓       | 3.0                    | 2.7                 | 90.1     |
| cal 3  | 3     | ✓       | 5.0                    | 4.6                 | 92.2     |
| cal 4  | 4     | ✓       | 10.0                   | 9.5                 | 95.0     |
| cal 5  | 5     | ✓       | 25.0                   | 24.8                | 99.2     |
| cal-6  | 6     | ✓       | 50.0                   | 50.3                | 100.7    |
| cal-7  | 7     | ✓       | 100.0                  | 100.8               | 100.8    |

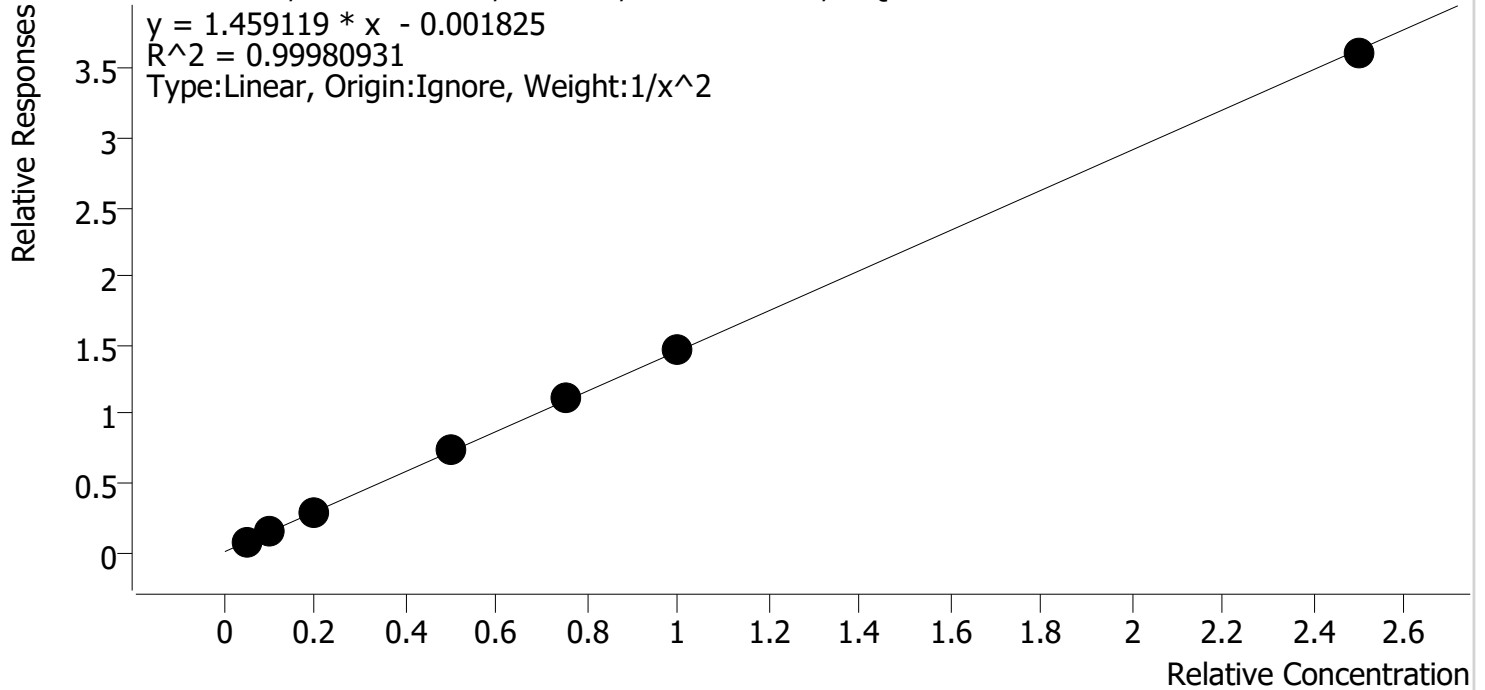


# Compound Calibration Report



**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Last Cal. Update** 8/21/2023 10:41 AM  
**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



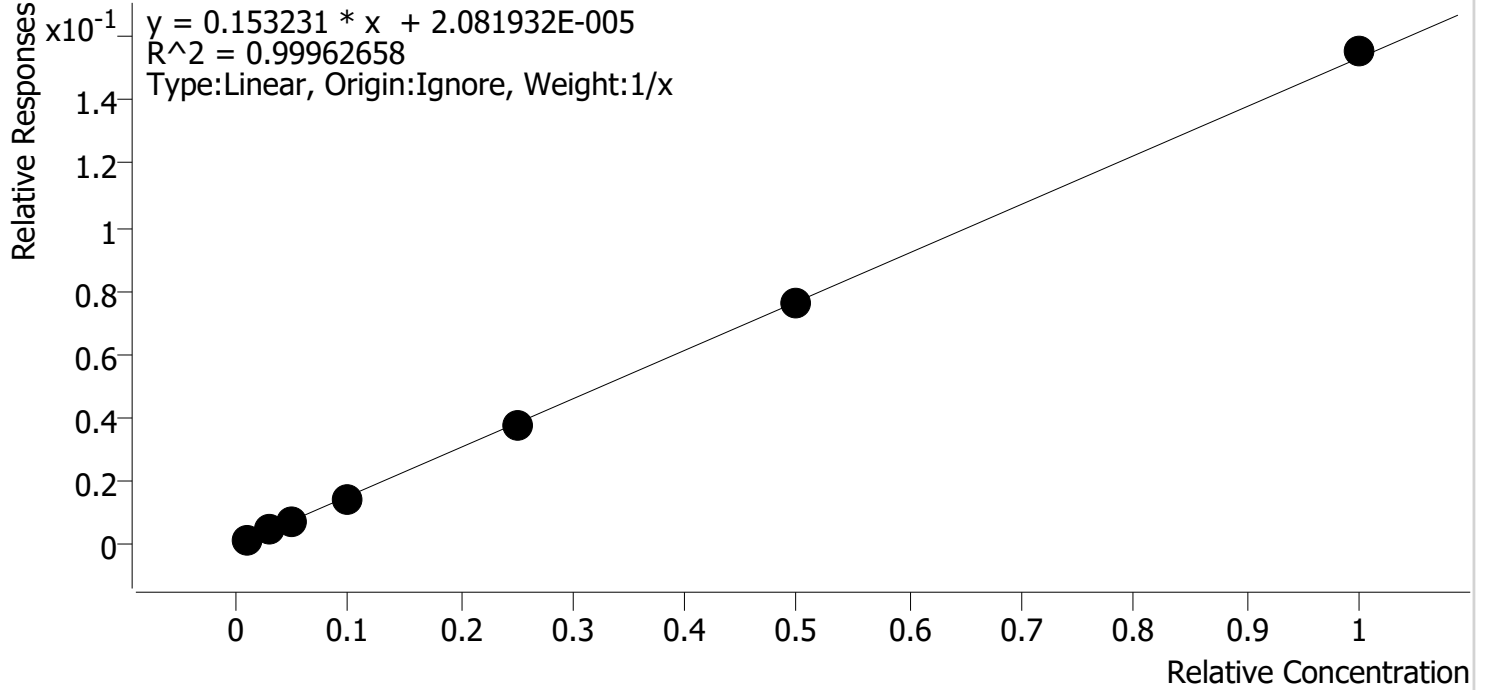
| Sample | Level | Enabled | Expected Concentration | Final Concentration | Accuracy |
|--------|-------|---------|------------------------|---------------------|----------|
| cal 1  | 1     | ✓       | 5.0                    | 5.0                 | 100.9    |
| cal 2  | 2     | ✓       | 10.0                   | 9.8                 | 98.2     |
| cal 3  | 3     | ✓       | 20.0                   | 19.9                | 99.4     |
| cal 4  | 4     | ✓       | 50.0                   | 50.1                | 100.1    |
| cal 5  | 5     | ✓       | 75.0                   | 75.8                | 101.1    |
| cal-6  | 6     | ✓       | 100.0                  | 101.1               | 101.1    |
| cal-7  | 7     | ✓       | 250.0                  | 247.9               | 99.2     |

# Compound Calibration Report



**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Last Cal. Update** 8/21/2023 10:41 AM  
**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



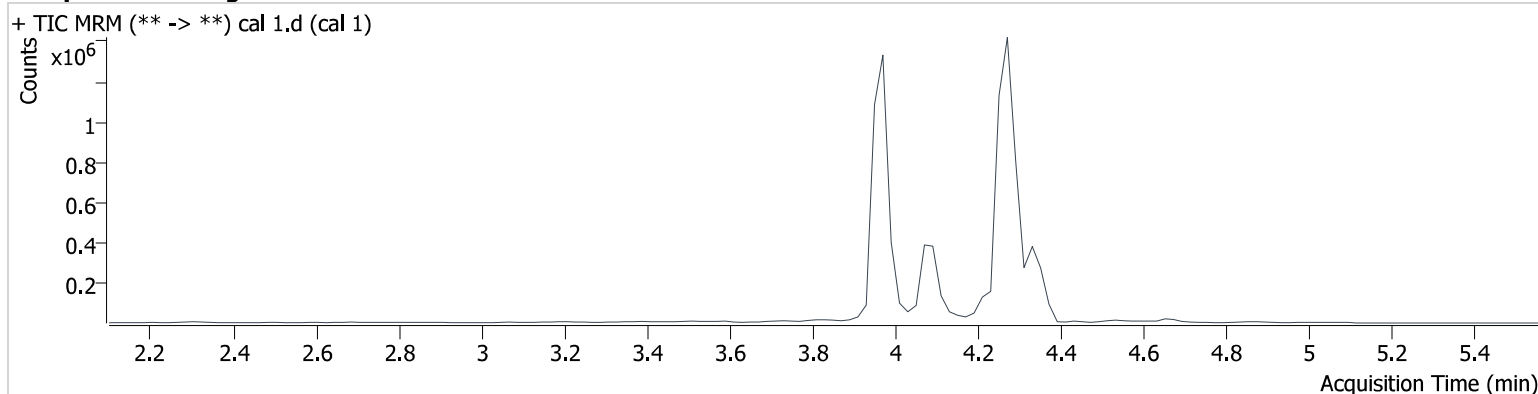
| Sample | Level | Enabled | Expected Concentration | Final Concentration | Accuracy |
|--------|-------|---------|------------------------|---------------------|----------|
| cal 1  | 1     | ✓       | 1.0                    | 1.1                 | 107.9    |
| cal 2  | 2     | ✓       | 3.0                    | 3.0                 | 101.1    |
| cal 3  | 3     | ✓       | 5.0                    | 4.8                 | 96.9     |
| cal 4  | 4     | ✓       | 10.0                   | 9.5                 | 95.2     |
| cal 5  | 5     | ✓       | 25.0                   | 24.6                | 98.2     |
| cal-6  | 6     | ✓       | 50.0                   | 49.7                | 99.3     |
| cal-7  | 7     | ✓       | 100.0                  | 101.3               | 101.3    |

# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |           |
|-------------------------|-------------------------|------------------|-----------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | cal 1.d   |
| <b>Type</b>             | Cal                     | <b>Sample</b>    | cal 1     |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord |
| <b>Sample Position</b>  | P3-A1                   | <b>Comment</b>   |           |
| <b>Injection Volume</b> | 5                       |                  |           |
| <b>Acq. Date-Time</b>   | 8/17/2023 8:18:40 PM    |                  |           |
| <b>Sample Info.</b>     |                         |                  |           |

## Sample Chromatogram



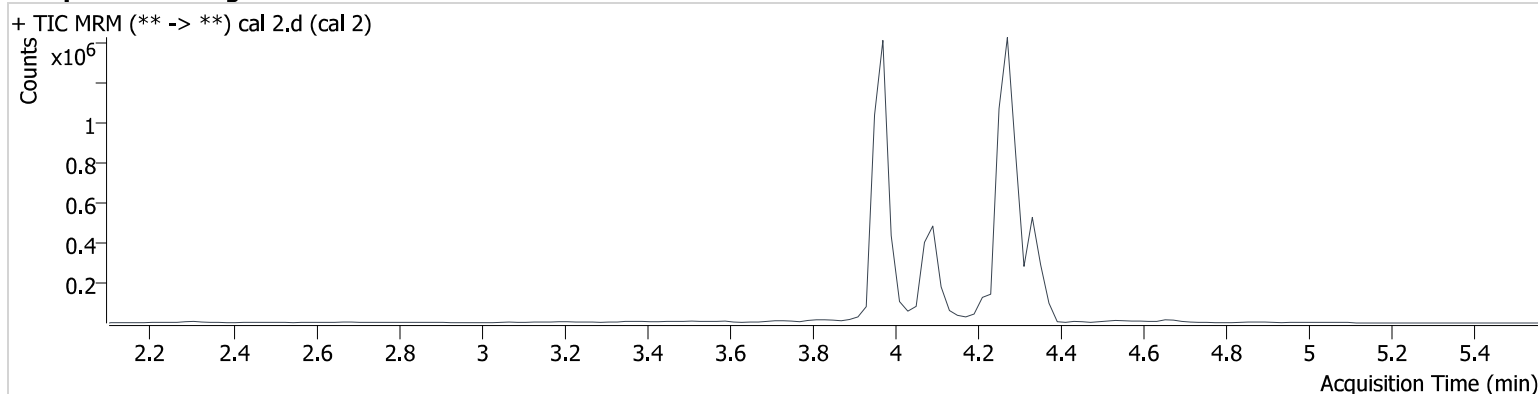
| Name     | RT    | Resp. | ISTD Resp. | Final Conc.            |
|----------|-------|-------|------------|------------------------|
| THC      | 4.345 | 5284  | 519365     | 1.219 ng/ml <b>Low</b> |
| THC-COOH | 4.093 | 63130 | 879017     | 5.047 ng/ml <b>Low</b> |
| THC-OH   | 3.979 | 5934  | 3543848    | 1.079 ng/ml <b>Low</b> |

# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |           |
|-------------------------|-------------------------|------------------|-----------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | cal 2.d   |
| <b>Type</b>             | Cal                     | <b>Sample</b>    | cal 2     |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord |
| <b>Sample Position</b>  | P3-B1                   | <b>Comment</b>   |           |
| <b>Injection Volume</b> | 5                       |                  |           |
| <b>Acq. Date-Time</b>   | 8/17/2023 8:25:18 PM    |                  |           |
| <b>Sample Info.</b>     |                         |                  |           |

## Sample Chromatogram



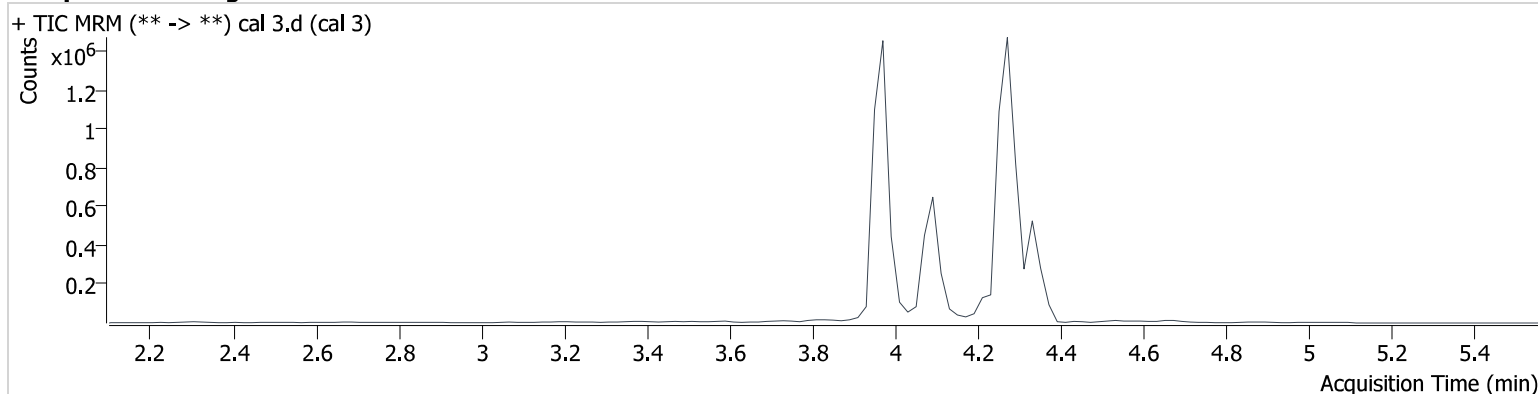
| Name     | RT    | Resp.  | ISTD Resp. | Final Conc.            |
|----------|-------|--------|------------|------------------------|
| THC      | 4.345 | 14282  | 692023     | 2.704 ng/ml <b>Low</b> |
| THC-COOH | 4.093 | 128697 | 909992     | 9.818 ng/ml <b>Low</b> |
| THC-OH   | 3.979 | 16534  | 3540143    | 3.034 ng/ml            |

# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |           |
|-------------------------|-------------------------|------------------|-----------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | cal 3.d   |
| <b>Type</b>             | Cal                     | <b>Sample</b>    | cal 3     |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord |
| <b>Sample Position</b>  | P3-C1                   | <b>Comment</b>   |           |
| <b>Injection Volume</b> | 5                       |                  |           |
| <b>Acq. Date-Time</b>   | 8/17/2023 8:31:46 PM    |                  |           |
| <b>Sample Info.</b>     |                         |                  |           |

## Sample Chromatogram



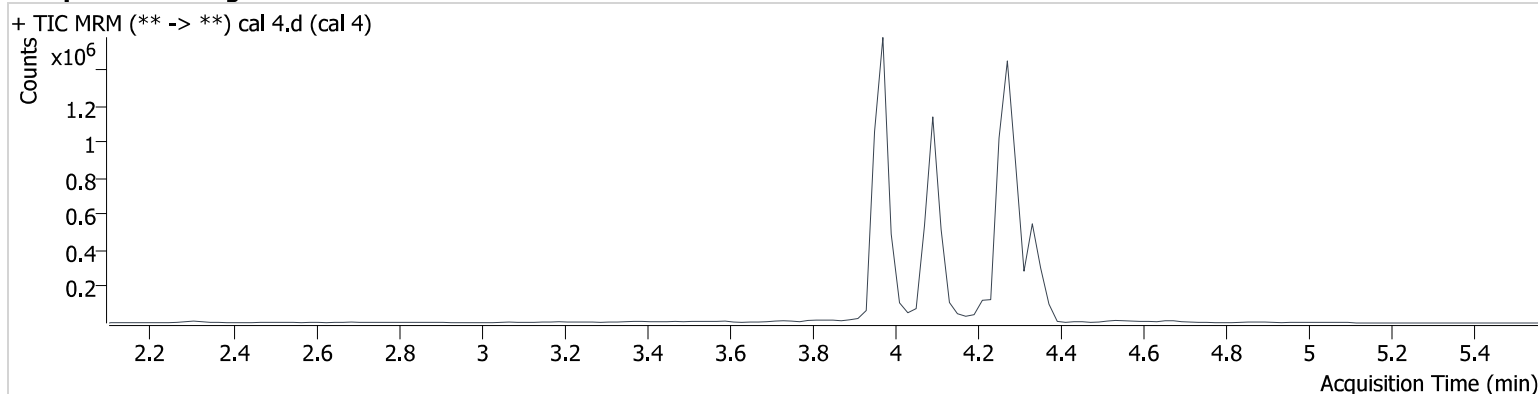
| Name     | RT    | Resp.  | ISTD Resp. | Final Conc.  |
|----------|-------|--------|------------|--------------|
| THC      | 4.345 | 22772  | 668627     | 4.609 ng/ml  |
| THC-COOH | 4.093 | 263399 | 913984     | 19.876 ng/ml |
| THC-OH   | 3.979 | 26520  | 3562891    | 4.844 ng/ml  |

# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |           |
|-------------------------|-------------------------|------------------|-----------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | cal 4.d   |
| <b>Type</b>             | Cal                     | <b>Sample</b>    | cal 4     |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord |
| <b>Sample Position</b>  | P3-D1                   | <b>Comment</b>   |           |
| <b>Injection Volume</b> | 5                       |                  |           |
| <b>Acq. Date-Time</b>   | 8/17/2023 8:38:14 PM    |                  |           |
| <b>Sample Info.</b>     |                         |                  |           |

## Sample Chromatogram



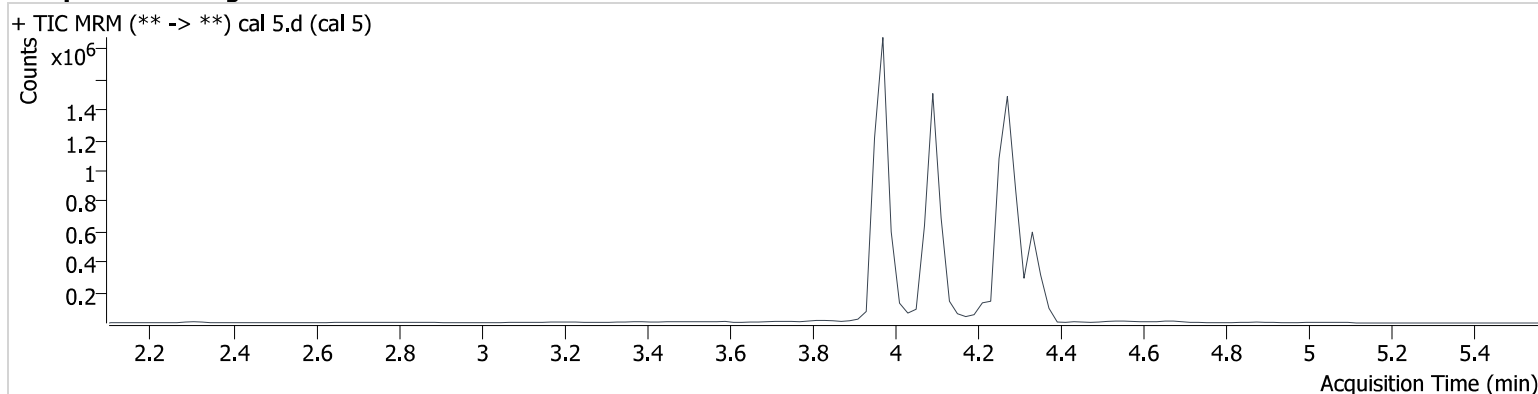
| Name     | RT    | Resp.  | ISTD Resp. | Final Conc.  |
|----------|-------|--------|------------|--------------|
| THC      | 4.345 | 47190  | 688738     | 9.501 ng/ml  |
| THC-COOH | 4.093 | 666179 | 914179     | 50.067 ng/ml |
| THC-OH   | 3.979 | 50418  | 3451220    | 9.520 ng/ml  |

# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |           |
|-------------------------|-------------------------|------------------|-----------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | cal 5.d   |
| <b>Type</b>             | Cal                     | <b>Sample</b>    | cal 5     |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord |
| <b>Sample Position</b>  | P3-E1                   | <b>Comment</b>   |           |
| <b>Injection Volume</b> | 5                       |                  |           |
| <b>Acq. Date-Time</b>   | 8/17/2023 8:44:42 PM    |                  |           |
| <b>Sample Info.</b>     |                         |                  |           |

## Sample Chromatogram



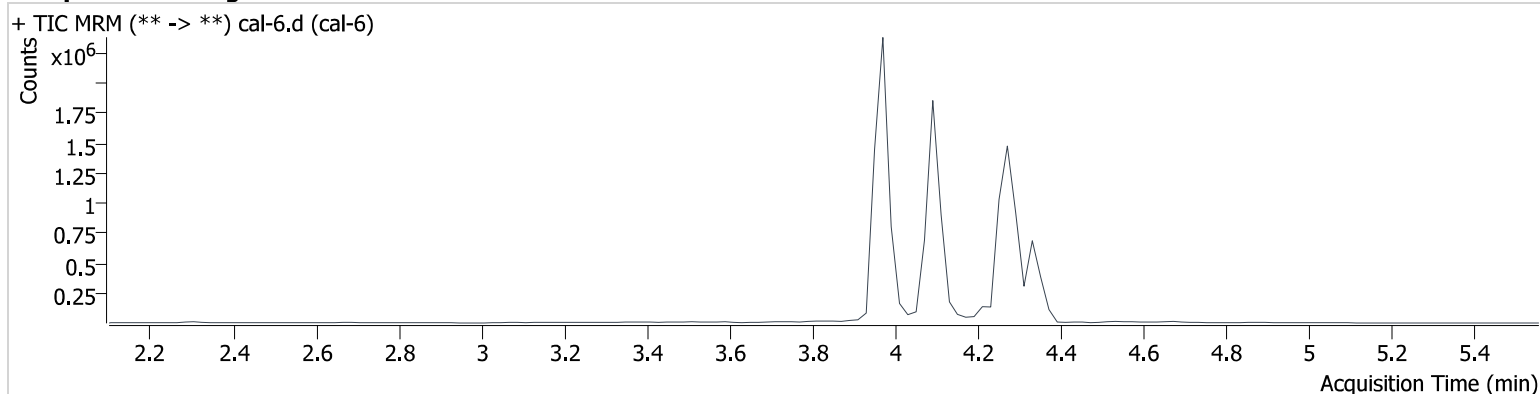
| Name     | RT    | Resp.  | ISTD Resp. | Final Conc.  |
|----------|-------|--------|------------|--------------|
| THC      | 4.345 | 118681 | 672894     | 24.812 ng/ml |
| THC-COOH | 4.093 | 991160 | 897556     | 75.807 ng/ml |
| THC-OH   | 3.979 | 129641 | 3442574    | 24.562 ng/ml |

# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |           |
|-------------------------|-------------------------|------------------|-----------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | cal-6.d   |
| <b>Type</b>             | Cal                     | <b>Sample</b>    | cal-6     |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord |
| <b>Sample Position</b>  | P3-F1                   | <b>Comment</b>   |           |
| <b>Injection Volume</b> | 5                       |                  |           |
| <b>Acq. Date-Time</b>   | 8/17/2023 8:51:11 PM    |                  |           |
| <b>Sample Info.</b>     |                         |                  |           |

## Sample Chromatogram



| Name     | RT    | Resp.   | ISTD Resp. | Final Conc.   |
|----------|-------|---------|------------|---------------|
| THC      | 4.345 | 248321  | 697031     | 50.347 ng/ml  |
| THC-COOH | 4.093 | 1285416 | 872332     | 101.113 ng/ml |
| THC-OH   | 3.979 | 259049  | 3403902    | 49.652 ng/ml  |

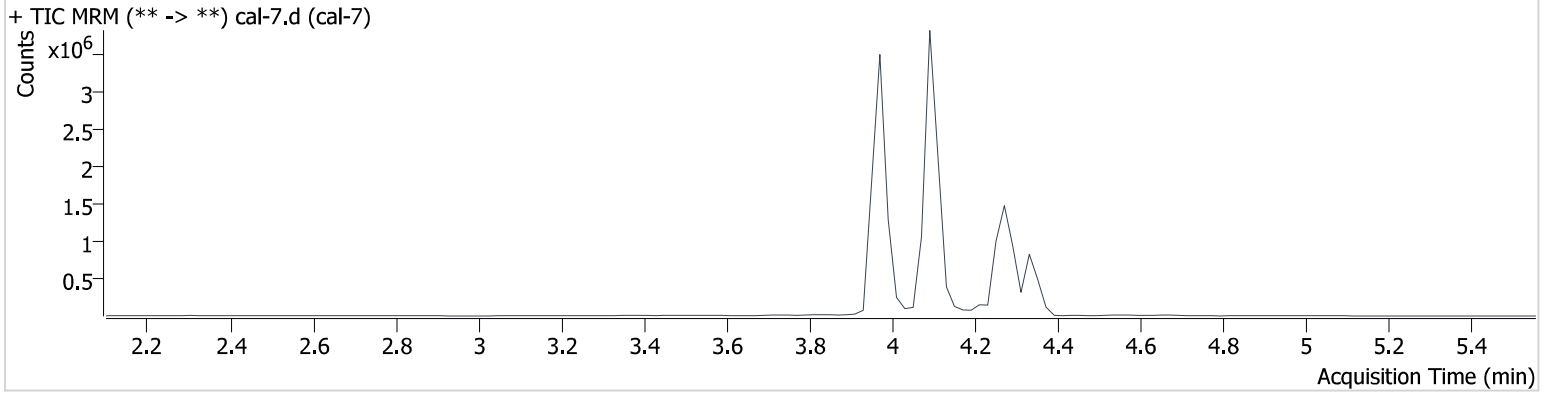


# AM #26 Cannabinoids Screen Results

**Batch results** D:\MassHunter\Data\2023\am 25-26\081723r\QuantResults\cann.batch.bin  
**Calibration Last Update** 8/21/2023 10:41:59 AM

|                         |                         |                  |           |
|-------------------------|-------------------------|------------------|-----------|
| <b>Instrument</b>       | 69679                   | <b>Data File</b> | cal-7.d   |
| <b>Type</b>             | Cal                     | <b>Sample</b>    | cal-7     |
| <b>Acq. Method</b>      | am 26 cann scr 5-5-20.m | <b>Operator</b>  | Anne Nord |
| <b>Sample Position</b>  | P3-G1                   | <b>Comment</b>   |           |
| <b>Injection Volume</b> | 5                       |                  |           |
| <b>Acq. Date-Time</b>   | 8/17/2023 8:57:39 PM    |                  |           |
| <b>Sample Info.</b>     |                         |                  |           |

## Sample Chromatogram



| Name     | RT    | Resp.   | ISTD Resp. | Final Conc.   |
|----------|-------|---------|------------|---------------|
| THC      | 4.345 | 486922  | 684154     | 100.807 ng/ml |
| THC-COOH | 4.093 | 2983229 | 825015     | 247.944 ng/ml |
| THC-OH   | 3.979 | 520621  | 3353311    | 101.308 ng/ml |