1/5/2023

REVIEWED
By Britany Wylie at 8:51 pm, Jan 05, 2023

Worklist: 6210

LAB_CASE	ITEM	ITEM_TYPE	DESCRIPTION
C2022-2701	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2701	2	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2708	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2710	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2729	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2773	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2776	4	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2780	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2791	1	UCK	AM 25/AM 26 Urine MultiDrug/THC Screen by LC-QQQ
C2022-2792	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2814	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2816	2	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2817	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2818	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2842	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2847	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2900	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ
C2022-2903	1	UCK	AM 25/AM 26 Urine MultiDrug/THC Screen by LC-QQQ
C2022-2908	1	BCK	AM 25/AM 26 Blood MultiDrug/THC Screen by LC-QQQ

	1	2	3	4	5	6	7	8	9	10	11	12
A			negative blood	2780-1	2900-1							
B	cal 1		2701-1	2792-1	2908-1							2791-1
C			2701-2	2814-1								2903-1
D			2708-1	2816-2								
E			2710-1	2817-1								
F			2729-1	2818-1								
G			2773-1	2842-1							postive control urine	
H			2776-4	2847-1							negative urine	

C2022-____-_

plate position 2

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

Extraction Date: 1/3/23 Analyst: Anne Nord
Plate lot#: 220805 Plate retest date: 02/05/23

Mobile phase A: 10mM Ammonium Formate **Mobile phase B:** 0.1% Formic Acid in MeOH
0.5M Ammonium Hydroxide Ethyl Acetate LC 20% Methanol
Blank Blood Lot: 22B52016-1 **Blank Urine lot:** 12522 **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 worklist was started the autosampler rammed the hotel after the injection of the first sample. In the process of trying to reset the autosampler the instrument computer shut down and would not restart. Due to the anticipated timeframe to repair or replace the computer this extraction will not be injected or evaluated*

On 1/17/23 after the computer and instrument were repaired these samples were injected to test the function of the instrument. They were not evaluated due to the time between extraction and injection being outside the approved window.

	1	2	3	4	5	6
a	cal 1	Internal control urine	2776-4	2847-1		
b	cal 2	negative blood	2780-1	2900-1		
c	cal 3	2701-1	2792-1	2908-1		
d	cal 4	2701-2	2814-1			
e	Cal 5	2708-1	2816-2	2791-1		
f	cal 6	2710-1	2817-1	2903-1		
g	cal 7	2729-1	2818-1	negative urine		
h	Internal control (blood)	2773-1	2842-1			

Plate position 3

c2022-____-_

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

Extraction Date: 1/3/23 Analyst: Anne Nord

Plate lot#: 220802 Plate retest date: 2/02/23

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: 22B52016-1 **Urine Blank:** 12522
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: I41142J**
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 8**00 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: *When the worklist was started the autosampler rammed the hotel after the injection of the first sample. In the process of trying to reset the autosampler the instrument computer shut down and would not restart. Due to the anticipated timeframe to repair or replace the computer this extraction will not be injected or evaluated.*

On 1/17/23 after the computer and instrument were repaired these samples were injected to test the function of the instrument. They were not evaluated due to the time between extraction and injection being outside the approved window.