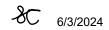
REVIEWED

By Celena Shrum at 1:28 pm, Jun 03, 2024



Worklist: 6829

LAB CASE ITEM ITEM TYPE DESCRIPTION

P2024-1367 2 BCK AM 28 Blood Multi-Drug Quant Panel 2 by LC-QQQ



8C

AM# 28: Multi-Drug Quantitation by LC-MS/MS

Extraction Date: 05/29/2024 Analyst: Sarah Collins

Plate lot#: 201207 Plate Retest Date: 06/07/2021 - external control ran Mobile phase A: 5mM Amm Form + 0.01% FA Mobile phase B: 0.01% Formic Acid in MeOH

Blank Blood Lot: <u>23A52595</u> Blank Urine Lot: <u>N/A</u>
Column: Agilent 120 EC-C18 (2.1x 100-2.7um) LCMS-QQQ ID: 069901

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.
- Urine Hydrolysis: In blank well, add 250μL urine, 40μL BG Turbo, and 100μL Instant Buffer I. Place on plate shaker for 5 minutes.
- Using a calibrated pipette, pipette 250μL blood or 250μL hydrolyzed urine in wells of analytical (standards) plate. Pipette ID: #42
- ☑ 4. Place on shaking incubator at ambient temp., 900rpm for 15 minutes.
- Σ 5. Pipette 250μL 0.5M ammonium hydroxide in wells of analytical plate.

- 8. 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)
- \boxtimes 9. Wait 5 minutes.
- \boxtimes 10. Add 900uL ethyl acetate.
- \boxtimes 11. Wait 5 minutes.
- □ 12. Apply positive pressure for approx. 15 seconds. (10-15 PSI- Selector to the left).
- □ 13. Add 900uL ethyl acetate.
- ⊠ 14. Wait 5 minutes.
- □ 15. Apply positive pressure for approx. 15 seconds. (10-15 PSI- Selector to the left).
- □ 16. Remove plate containing eluate. Place on SPE Dry and evaporate to dryness at approx. 35°C.
- 17. Add 50μL 1% HCl in MeOH to wells and place plate cover on plate before drying. This step is required for urine samples, but optional for blood samples.
- ⊠ 18. Reconstitute in 100μL 20% LC MeOH and heat seal plate with foil. Place in autosampler and run worklist.

Post-Analytic

- \boxtimes 3. Integration linear and R² values \ge 0.98 for each analyte.
- ☑ 5. Did all QCs pass for each analyte? Yes, see comments Add Control data to QC tracking spreadsheet.
- ⊠ 6. Central File Packet to include: LIMS Worklist, Method Checklist, Calibration and Control Reports.

COMMENTS:



Compounds Evaluated and Curve Range Limits: Amitriptyline Doxepin 5-500 – calibrator 8 dropped due to peak shape Flurazepam 5-250 – calibrators 7 and 8 dropped due to accuracy

	1	2	3	4	5	6	7	8	9	10	11	12
Α	IS + Cal. 1	IS + QC_1		IS + Cal. 1	IS + QC_1				IS + Cal. 8			IS + Cal. 8
В	IS + Cal. 2	IS + QC_2		IS + Cal. 2	IS + QC_2				IS + Cal. 7			IS + Cal. 7
С	IS + Cal. 3	IS + QC_3		IS + Cal. 3	IS + QC_3				IS + Cal. 6			IS + Cal. 6
D	IS + Cal. 4	IS + QC_4		IS + Cal. 4	IS + QC_4				IS + Cal. 5			IS + Cal. 5
Е	IS + Cal. 5			IS + Cal. 5	negative blood			IS + QC_4	IS + Cal. 4		IS + QC_4	IS + Cal. 4
F	IS + Cal. 6			IS + Cal. 6	external blood			IS + QC_3	IS + Cal. 3		IS + QC_3	IS + Cal. 3
G	IS + Cal. 7			IS + Cal. 7	p2024-1367-2			IS + QC_2	IS + Cal. 2		IS + QC_2	IS + Cal. 2
Н	IS + Cal. 8			IS + Cal. 8				IS + QC_1	IS + Cal. 1		IS + QC_1	IS + Cal. 1

All wells to contain 60 μl of Trapping Solution

Samples were moved on the SLE plate; column 4 to column 11 and column 5 to column 12





D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin **Batch results** Calibration Last Update 6/3/2024 9:52:47 AM

Instrument Type Acq. Method **Sample Position Injection Volume** Falco (069901) Sample

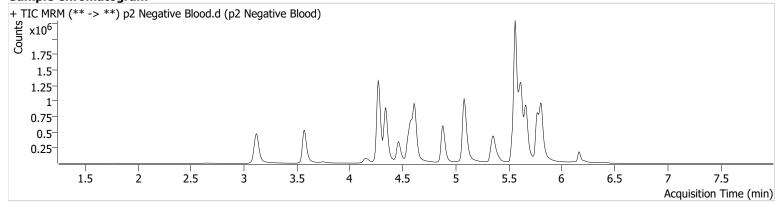
AM 28 P2-Updated Dec 2023.m P2-E12

5 5/29/2024 5:19:06 PM

Acq. Date-Time Sample Info.

Data File Sample Operator Comment p2 Negative Blood.d p2 Negative Blood Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.





Idaho State Police Forensic Services

AM #28 Blood/Urine Multi-Drug Confirmatory Analysis by LCMS-QQQ---Panel 2

Methanol External Control Solution (Lot: 052824)

100 ul each 1 mg/mL stock solution in 9800 ul MeOH

Component	Source	Source Lot Number	Expiration Date	
Methanol (LCMS)	Fisher	222621	N/A	
Amitriptyline	Cerilliant	FN02202004	03/31/2025	
Flurazepam	Cerilliant	FE08231902	11/30/2024	
Prepared:	05/28/2024			
Prepared By:	Tamara Salazar			
Expires:	11/30/2024			

Blood External Control Solution (Lot: WS052824)

100 ul of methanol external control solution was added to 9900ul of blood.

Approximately 100ng/mL of each compound.

Component	Source	Source Lot Number
Negative Blood	Lampire	23A52595
Methanol External Control Solution	-	052824
Prepared:	05/28/2024	
Prepared by:	Tamara Salazar	
Expires:	11/30/2024	





Batch results
D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin
Calibration Last Update
6/3/2024 9:52:47 AM

Instrument
Type
Acq. Method
Sample Position
Injection Volume

Falco (069901) Sample

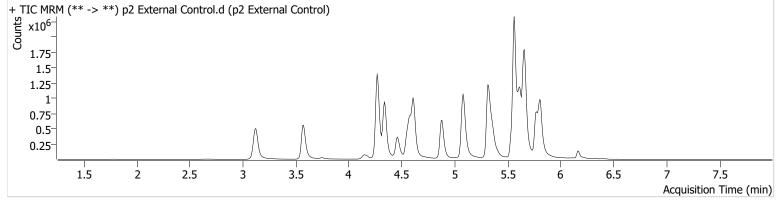
AM 28 P2-Updated Dec 2023.m P2-F12

5

Acq. Date-Time 5/29/2024 5:40:37 PM **Sample Info.**

Data File Sample Operator Comment p2 External Control.d p2 External Control Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	528220	1682.38	127.0	8774.27	389733	73.6409 ng/ml
Flurazepam	5.312	2766693	1172.28	13.6	2506.96	914128	60.8140 ng/ml





D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin **Batch results** Calibration Last Update 6/3/2024 9:52:47 AM

Instrument **Type** Acq. Method **Sample Position** Falco (069901) QC

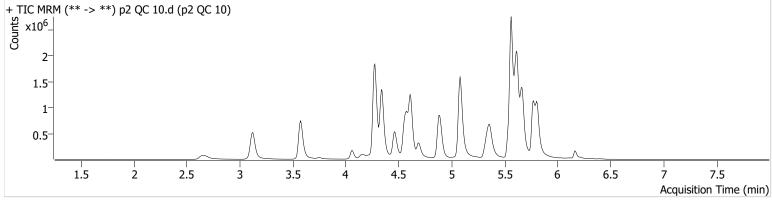
AM 28 P2-Updated Dec 2023.m P2-A12

5 5/29/2024 4:14:36 PM

Injection Volume Acq. Date-Time Sample Info.

Data File Sample Operator Comment p2 QC 10.d p2 QC 10 Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	93328	679.63	141.7	1410.72	541760	9.1799 ng/ml
Doxepin	5.3 4 3	226060	428.68	43.4	390.70	831564	10.3276 ng/ml
Flurazepam	5.312	417850	311.86	12.7	142.19	905508	9.6722 ng/ml





D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin **Batch results** Calibration Last Update 6/3/2024 9:52:47 AM

Instrument **Type** Acq. Method **Sample Position** Falco (069901) QC

AM 28 P2-Updated Dec 2023.m

P2-C12 5

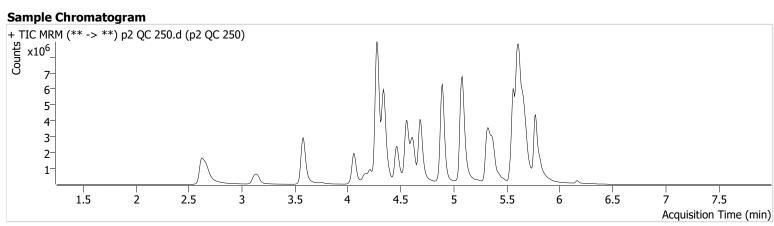
Injection Volume Acq. Date-Time 5/29/2024 4:36:06 PM

Sample Info.

Data File Sample Operator Comment

p2 QC 250.d p2 QC 250 Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	1605092	2535.15	131.2	289.64	384292	227.3686 ng/ml
Doxepin	5.343	3613708	5937.91	49.0	11984.03	581022	234.5793 ng/ml
Flurazepam	5.312	6943982	61713.82	14.2	3355.15	483572	286.7665 ng/ml





Batch results D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin
Calibration Last Update 6/3/2024 9:52:47 AM

Instrument
Type
Acq. Method
Sample Position
Injection Volume

Falco (069901) QC

AM 28 P2-Updated Dec 2023.m P2-D12

5

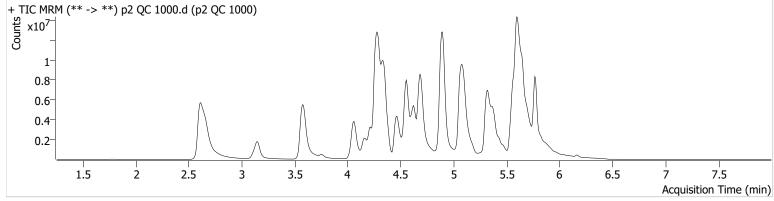
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Acq. Date-Time Sample Info.

Data File Sample Operator Comment

p2 QC 1000.d p2 QC 1000 Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.646	4027346	16659.53	125.3	123.78	239703	915.2366 ng/ml
Doxepin	5.337	9360755	20110.46	50.2	39984.32	367359	960.8172 ng/ml
Flurazepam	5.305	17048916	992682.09	13.6	2906.31	234584	1449.4588 ng/ml





Batch results
D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin
6/3/2024 9:52:47 AM

Instrument
Type
Acq. Method
Sample Position

Falco (069901) QC

AM 28 P2-Updated Dec 2023.m P2-B12

Injection Volume 5

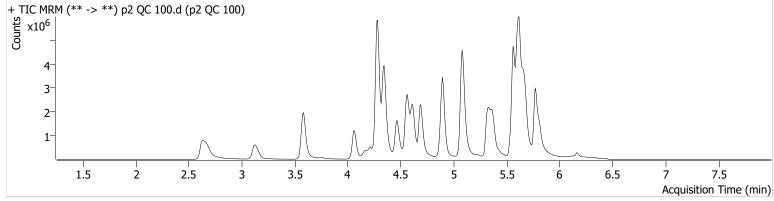
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Acq. Date-Time Sample Info.

Data File Sample Operator Comment

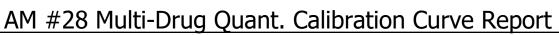
p2 QC 100.d p2 QC 100 Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	840393	775.74	132.6	1165.12	503634	90.7125 ng/ml
Doxepin	5.343	1975 44 3	16037.10	4 7.9	937.18	756812	98.4929 ng/ml
Flurazepam	5.312	3969894	351.92	13.6	1022.77	724465	109.7235 ng/ml

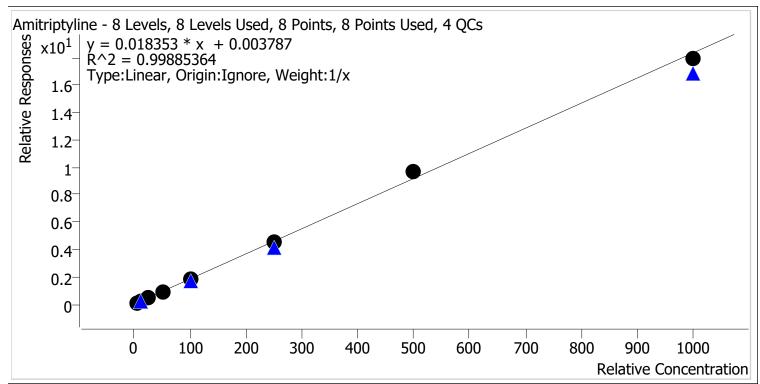




Batch results D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin

Last Cal. Update 6/3/2024 9:52 AM
Analyst Name ISP\Datastor

Analyte Amitriptyline Internal Standard Amitriptyline-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
p2 Cal 1-5ng	1	~	5.0	4.8	95.4
p2 Cal 2-10ng	2	~	10.0	10.5	105.1
p2 Cal 3 -25ng	3	~	25.0	24.7	98.9
p2 Cal 4-50ng	4	~	50.0	49.6	99.3
p2 Cal 5-100ng	5	~	100.0	99.0	99.0
p2 Cal 6-250ng	6	~	250.0	248.2	99.3
p2 Cal 7-500ng	7	~	500.0	526.6	105.3
n2 Cal 8-1000ng	8	V	1000.0	976.6	97 7

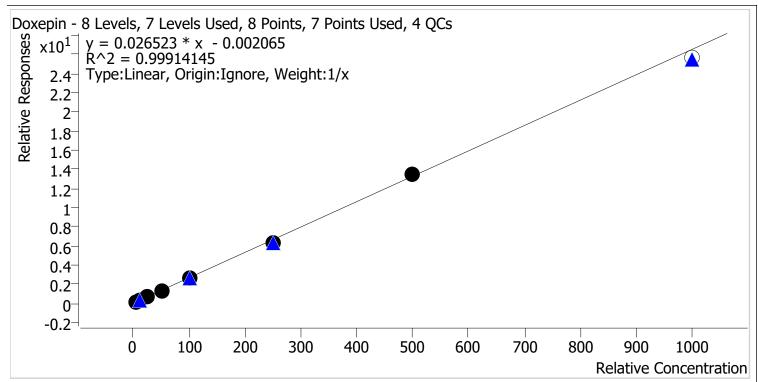




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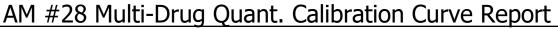
Last Cal. Update6/3/2024 9:52 AMAnalyst NameISP\Datastor

Analyte Doxepin Internal Standard Doxepin-D3



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
p2 Cal 1-5ng	1	V	5.0	5.0	99.2
p2 Cal 2-10ng	2	V	10.0	10.3	103.0
p2 Cal 3 -25ng	3	V	25.0	25.4	101.5
p2 Cal 4-50ng	4	V	50.0	50.1	100.2
p2 Cal 5-100ng	5	V	100.0	97.9	97.9
p2 Cal 6-250ng	6	V	250.0	240.2	96.1
p2 Cal 7-500ng	7	V	500.0	511.2	102.2
p2 Cal 8-1000ng	8	х	1000.0	966.8	96.7

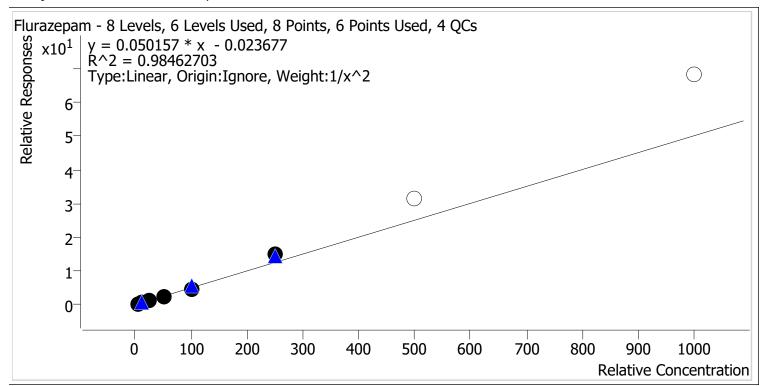




Batch results D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin

Last Cal. Update 6/3/2024 9:52 AM
Analyst Name ISP\Datastor

Analyte Flurazepam Internal Standard Midazolam-D4



Sample	Level	Enabled	Expected Concentration	Final Concentration	Accuracy
p2 Cal 1-5ng	1	V	5.0	5.2	104.1
p2 Cal 2-10ng	2	V	10.0	9.3	93.3
p2 Cal 3 -25ng	3	V	25.0	24.6	98.5
p2 Cal 4-50ng	4	V	50.0	48.9	97.7
p2 Cal 5-100ng	5	V	100.0	88.6	88.6
p2 Cal 6-250ng	6	V	250.0	294.6	117.8
p2 Cal 7-500ng	7	Х	500.0	627.0	125.4
p2 Cal 8-1000ng	8	×	1000.0	1362.5	136.3





D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin **Batch results** Calibration Last Update 6/3/2024 9:52:47 AM

Instrument Type Acq. Method **Sample Position** Falco (069901) Cal

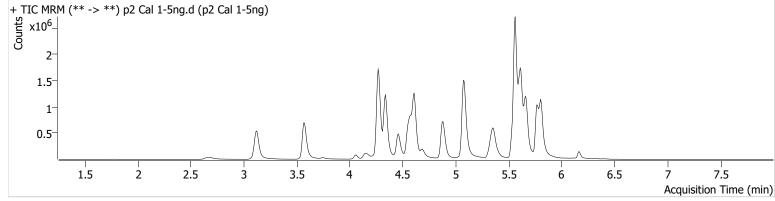
AM 28 P2-Updated Dec 2023.m P2-A11

5

Injection Volume Acq. Date-Time 5/29/2024 2:16:09 PM Sample Info.

Data File p2 Cal 1-5ng.d **Sample** p2 Cal 1-5ng Operator Sarah Collins Comment

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	46484	332.29	141.4	8772.21	508847	4.7711 ng/ml
Doxepin	5.336	107991	499.91	44.3	174.27	834367	4.9578 ng/ml
Flurazepam	5.312	234854	1142.49	12.6	1737.65	989260	5.2052 ng/ml





D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin **Batch results** Calibration Last Update 6/3/2024 9:52:47 AM

Instrument Type Acq. Method **Sample Position** Falco (069901) Cal

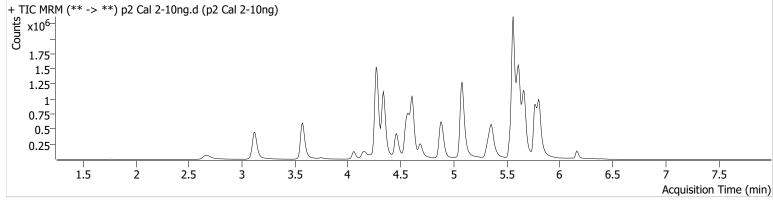
Injection Volume Acq. Date-Time Sample Info.

AM 28 P2-Updated Dec 2023.m P2-B11 5

5/29/2024 2:27:04 PM

Data File p2 Cal 2-10ng.d **Sample** p2 Cal 2-10ng Operator Sarah Collins Comment

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	72192	469.84	131.8	1137.33	366892	10.5148 ng/ml
Doxepin	5.336	16 4 976	465.57	41.9	4 95.66	608654	10.2975 ng/ml
Flurazepam	5.312	354918	1463.67	12.9	21418.96	798935	9.3290 ng/ml





D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin **Batch results** Calibration Last Update 6/3/2024 9:52:47 AM

Instrument **Type** Acq. Method **Sample Position Injection Volume** Falco (069901) Cal

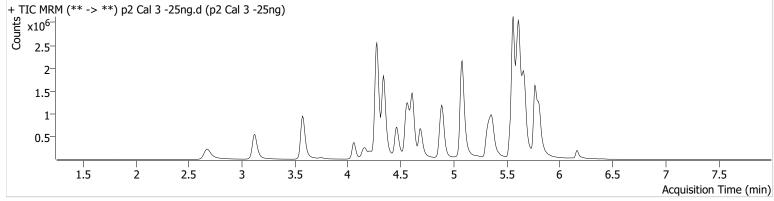
AM 28 P2-Updated Dec 2023.m P2-C11

5 5/29/2024 2:37:48 PM

Acq. Date-Time Sample Info.

Data File Sample Operator Comment p2 Cal 3 -25ng.d p2 Cal 3 -25ng Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	257193	1404.45	137.9	1002.76	562009	24.7282 ng/ml
Doxepin	5.336	559962	4095.63	44.4	395.65	834755	25.3699 ng/ml
Flurazepam	5.312	1075921	4453.72	13.3	139007.03	888516	24.6144 ng/ml





D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin **Batch results** Calibration Last Update 6/3/2024 9:52:47 AM

Instrument Type Acq. Method **Sample Position** Falco (069901) Cal

AM 28 P2-Updated Dec 2023.m

P2-D11 5

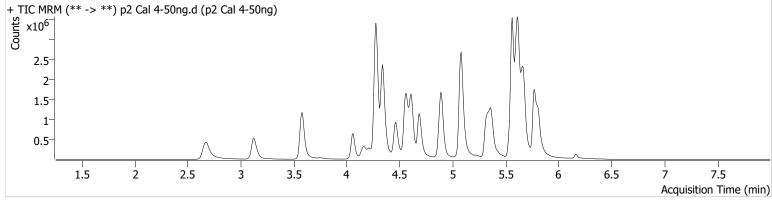
Injection Volume Acq. Date-Time 5/29/2024 2:48:33 PM

Sample Info.

Data File Sample Operator Comment

p2 Cal 4-50ng.d p2 Cal 4-50ng Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	378454	1633.57	135.1	1328.61	413739	49.6330 ng/ml
Doxepin	5.337	899035	1174.53	44.9	487.40	677581	50.1044 ng/ml
Flurazepam	5.312	1877003	8445.78	13.2	588.43	773309	48.8644 ng/ml





D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin **Batch results** Calibration Last Update 6/3/2024 9:52:47 AM

Instrument Type Acq. Method **Sample Position** Falco (069901) Cal

AM 28 P2-Updated Dec 2023.m

P2-E11 5

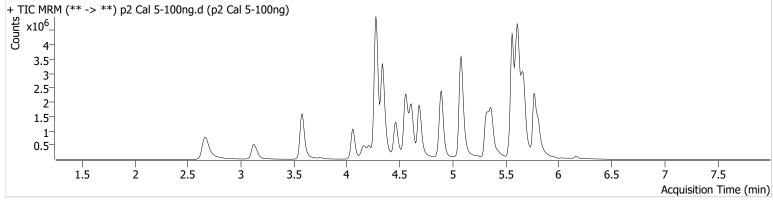
Injection Volume Acq. Date-Time 5/29/2024 2:59:19 PM

Sample Info.

Data File Sample Operator Comment

p2 Cal 5-100ng.d p2 Cal 5-100ng Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	596036	1797.58	136.1	7356.09	327375	98.9941 ng/ml
Doxepin	5.336	1393815	3740.18	46.6	2231.32	537460	97.8565 ng/ml
Flurazepam	5.312	3016470	117290.19	14.0	289.25	682531	88.5855 ng/ml





Batch results
D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin
Calibration Last Update
6/3/2024 9:52:47 AM

Instrument
Type
Acq. Method
Sample Position
Injection Volume

Falco (069901) Cal

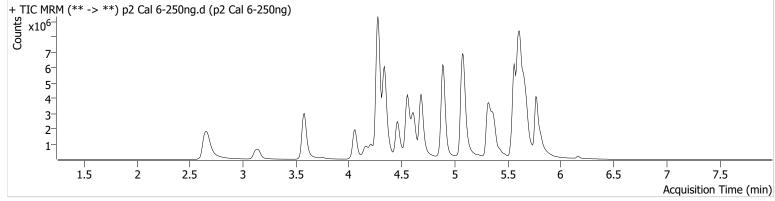
AM 28 P2-Updated Dec 2023.m P2-F11

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Acq. Date-Time 5/29/2024 3:10:05 PM **Sample Info.**

Data File Sample Operator Comment p2 Cal 6-250ng.d p2 Cal 6-250ng Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	1541678	1900.08	128.9	7613 4 .93	338157	248.1993 ng/ml
Doxepin	5.336	3354564	238.94	49.4	373.12	526718	240.2061 ng/ml
Flurazepam	5.312	7632792	230580.02	13.8	5383.33	517412	294.5836 ng/ml





D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin **Batch results** Calibration Last Update 6/3/2024 9:52:47 AM

Instrument Type Acq. Method **Sample Position** Falco (069901) Cal

5/29/2024 3:20:50 PM

AM 28 P2-Updated Dec 2023.m

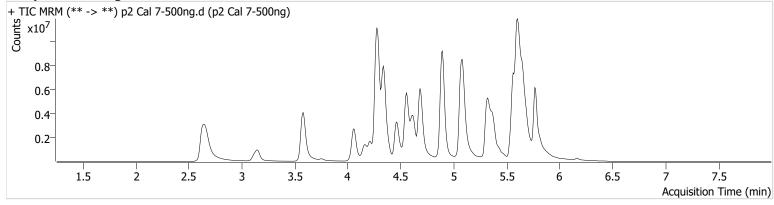
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Injection Volume Acq. Date-Time Sample Info.

P2-G11

Data File Sample Operator Comment p2 Cal 7-500ng.d p2 Cal 7-500ng Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	2777833	4805.18	126.3	329.38	287299	526.6089 ng/ml
Doxepin	5.336	5955036	33096.18	49.4	1963.60	439277	511.2077 ng/ml
Flurazepam	5.312	11272807	18299.25	13.6	575.78	358745	626.9588 ng/ml





D:\MassHunter\Data\2024\AM 27 28\052924 AM 28 P2 SC\QuantResults\AM 28 P2.batch.bin **Batch results** Calibration Last Update 6/3/2024 9:52:47 AM

Instrument Type Acq. Method **Sample Position** Falco (069901) Cal

AM 28 P2-Updated Dec 2023.m P2-H11

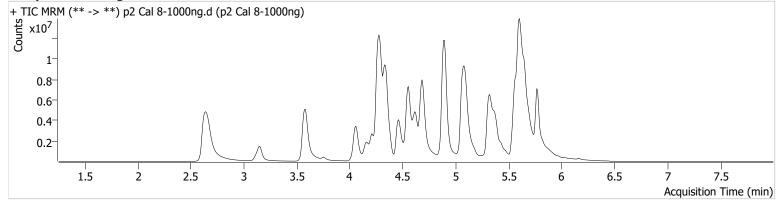
Injection Volume 5

Acq. Date-Time 5/29/2024 3:31:35 PM Sample Info.

Data File Sample Operator Comment

p2 Cal 8-1000ng.d p2 Cal 8-1000ng Sarah Collins

Only drugs and concentrations listed on the laboratory report itself are appropriate to be used for interpretation purposes. Any drugs or values included in the notes but not included on the report are used by laboratory personnel to make determinations/reach conclusions within the confines of the methods.



Name	RT	Resp.	S/N	Ratio	S/N	ISTD Resp.	Final Conc.
Amitriptyline	5.652	3686715	1876.59	125.7	1369.83	205655	976.5505 ng/ml
Doxepin	5.336	8208851	102520.91	49.6	22664.33	320169	966.7705 ng/ml
Flurazepam	5.305	15589740	147388.40	13.8	4965.36	228196	1362.5308 ng/ml