

Toxicology Laboratory – General Lab Meeting

November 8, 2016

Present – AB, AC, BEO, BP, CJ, CM, DN, DS, EW, FC, JK, LK, LN, MB, NN, RF

Old Business

Centrifuge repair company is moving and it may be another week before they can come to do repairs. Compost buckets are up by the lab exits and restrooms for paper towel waste. They can be emptied in the lunch room compost.

Safety - RF

Audit on 11/14, most likely no one in the lab will have to do anything for this.

QA/QC -AB

- Pipettes – New pipette schedule is out and should help with all the maintenance. Please be willing to share the loaner set of pipettes while out for calibration. PES Lite pipettes are designed to be more ergonomic (as the Biohit), and internal parts may be more susceptible to wear – please notify QA department if you suspect there are any issues with your pipette(s).
- Proficiency Testing – Just finished last set for year. Break until March. In general, try to get these complete as soon as you can and check for errors. When submitting proficiency data/results forms, please review completely, as you would for a case file and test report.
- Methods Update – Moving forward on synthetic cannabinoids method on the two SCIEX instruments. New amines method moving forward on the two Agilent instruments. Thanks for being flexible with instrument use.
- SIM Data – If a value or two fails, you do not need to line through anything. Make a note about the values being out and if it will be a retest or a remake.
- BAC Batch Data (Multiple Data Folders) – You do not have to write the batch number on every sheet of data if you run stops and you create a new data path. Just make a note on the worklist.
(Additional note from AB) - Since the data file folder (batch ID) prints on each page, that identifies the report to its batch. A copy of the worklist gets included with every report (either in the batch folder with the calibrator/control/blank or in case file folders with case reports), so this is the best place to note that there are multiple batch IDs.

Brianna

- Proficiency Tests – These are sent with discovery requests. Please do these properly.
- 1) Snohomish county – Judges are not always allowing one analyst to talk about the whole case. Work with the prosecutor and see if you can prioritize drugs to reduce number of analysts testifying or if you can talk about positive only results.

- 2) Subpoenas – We need to honor subpoenas, even if they have the wrong name or a typo on them. If it is our case we need to accommodate. It is also possible that our office has lost the subpoena in the 100s that come in every day. We are still responsible for these cases. CM asked about AK and last minute phone calls. BP -You can talk with supervisor if things seem unreasonable.
- 3) Analytical methods – synthetic cannabinoids is currently a “novel test method” and positives should go to AXIS. We will screen cases first. If they did not call Spice and our panel did not find anything, you can judge if a notation is made on the final report indicating testing was performed. If Spice was requested, and the panel is negative, you must put the test on the report.
- 4) SSRI testing - the volume and frequency of testing may be affecting the stability of sertraline and nortriptyline. These compounds currently will be report as positive only.
- 5) NSC meeting – National Safety Council issues minimum testing recommendations for laboratories. They have Tier 1 drugs, which all labs should look for and Tier 2 drugs that are seen locally or infrequently. PCP and Barbs have been moved from Tier 1 to Tier 2. Fentanyl / Nor , Bupropion / Nor, Tramadol and desmethyltramadol have been upgraded to Tier 1. Our lab is doing well in meeting these standards.
- 6) Administrative changes – Full PEC staff. Welcome Melvin. Please review cases and reports carefully as many people are learning or doing unfamiliar tasks. Especially note if tubes from the Medical Examiners match in regard to peripheral vs central blood. Also, we will be using the term vacutainer lavender (per manufacturer’s naming) instead of purple.

Lisa:

- 1) SIM solutions – our production schedule will remain the same. Try to make your batches in the first two weeks.
There are changes in the distribution of solution. Roanoke will be taking them from us and distributing them to all other agencies.
A change in Breath Test policy is requiring two 0.08 solutions from different lot numbers be used. We will be making 0.08 solutions more often. See Lisa for your monthly assignment.

Round Table

BP- If you have a standing desk you are not using, see her. There are those that would like one.

MB – excited to be working here. Please let him know if something is not being done correctly so he can fix it right away.

JK – SOFT was awesome. Already passed around some information. Try to get to the next one if you can. Has a standing desk available and will talk to BP.

CM- Went to STATs conference. It was a “hurt your brain” kind of meeting, but really good. She has notes if you are interested.

RF – Trying to fix QQQ1. Internal Standard response dropping as the run goes on. Troubleshooting requires at least 10 injections and the process may take a while. Thanks for your patience.

AC- noticed Dilutor #1 is starting to stick. CM suggested unplugging and replugging in the handle to the side. The whole handle can be replaced when it fails, just order a new one.

Made new IS for BDS that is ready for use.

SOFT conference covered some case studies with extreme differences between ante and post mortem THC samples. Interpretation can be tricky.

LK – Sent around slides already from SOFT. Poster went well.

NN – CAT conference was nice and small with good interactions. BP has electronic files and will distribute.

JK – Does the training in December supersede court? BP- no firm answer. The training is not recorded, so you may be able to tell prosecutors ahead of time that you are “unavailable”

See page 2 of meeting minutes
Archived 5/18/17
Responsible for qualitative only reporting

CONFIRMATION OF SELECTIVE SEROTONIN REUPTAKE INHIBITOR COMPOUNDS BY GAS CHROMATOGRAPHY - MASS SPECTROMETRY

33.1 POLICY

This test method may be used to confirm the presence of fluoxetine (FLX) and norfluoxetine (NFX), using fluoxetine-d₆ (FLX-d₆) internal standard, and sertraline (SER) and desmethylsertraline (norsertaline, NSR), using protriptyline (PRO) internal standard, in biological samples. Quantitative results obtained through the use of this method will only be reported within the validated dynamic range. Reporting of results following the application of this method will be contingent upon a thorough review and acceptance of quality control data and the qualification of individual results under the criteria for acceptance.

Any adjustments or deviations from the procedures below must be approved by a member of TLD Management, and appropriately documented in the batch file.

33.2 PURPOSE

The purpose of this standard operating procedure (SOP) is to provide technical direction for the identification and quantitation of selective serotonin reuptake inhibitor (SSRI) compounds present in biological specimens. This procedure will serve as the laboratory document describing sample preparation, instrumental analysis, data analysis, criteria for acceptance and reporting of the specified compounds.

33.3 PRINCIPLE

The targeted compounds and internal standards are isolated from whole blood, serum, plasma, urine or other submitted biological samples by the use of solid phase extraction (SPE). Following SPE, the specimens, now termed extracts, are subjected to chemical derivatization with heptafluorobutyric acid anhydride (HFBA or HFAA).

Measured volumes of the extracts are injected into a GC where they are separated between a gaseous mobile and liquid stationary phase. Each compound exits the GC at a reproducible time which is termed its retention time.

The GC is coupled to a mass spectrometer (MS) detector equipped with an electron ionization source. As each compound is ionized in the source, selected-ion-monitoring is used to measure the mass-to-charge ratios of each compound and its related fragments. Multiple-point, internal standard calibration is used to generate a calibration curve. The concentration of any target compound identified in a sample is determined from its calibration curve.

33.4 SPECIMENS

33.4.1 The specimen volume is 1 mL.

- 33.4.2 Specimens include whole blood, serum, plasma, urine, and tissue homogenate.
- 33.4.3 Dilutions of specimens may be analyzed at the Forensic Scientist's discretion; however, this should be done in addition to testing the standard specimen volume, unless sample quantity dictates otherwise.
- 33.4.4 Analysis of larger specimen volumes must be approved and documented.

33.5 REAGENTS, MATERIALS AND EQUIPMENT

33.5.1 REAGENTS

33.5.1.1 Acetic acid (glacial)

33.5.1.2 0.1M acetic acid

To 800mL of DI H₂O, add 5.72 mL glacial acetic acid. Dilute to 1 L with DI H₂O. Store this in glass bottle at room temperature for up to 6 months. Adjustments to final volume are permitted as long as proportions are maintained.

33.5.1.3 Acetonitrile (ACN)

33.5.1.4 Ammonium hydroxide (NH₄OH), concentrated

33.5.1.5 Certified blank blood

33.5.1.6 Deionized water (DI H₂O)

33.5.1.7 Elution solvent

To 20 mL isopropanol, add 2 mL concentrated ammonium hydroxide and mix. Add 78 mL methylene chloride and mix. Store in glass flask/bottle at room temperature and use on date of preparation only. Adjustments to final volume are permitted as long as the proportions of the elution solvent are maintained.

33.5.1.8 Ethyl acetate

33.5.1.9 Heptafluorobutyric acid anhydride (HFBA, HFAA)

33.5.1.10 Isopropanol (IPA)

33.5.1.11 Methanol (MeOH)

33.5.1.12 Methylene chloride (dichloromethane, CH₂Cl₂)

33.5.1.13 0.1M Phosphate buffer (pH 6):

Dissolve 1.7 g Na₂HPO₄ and 12.14 g NaH₂PO₄ in 800mL DI H₂O. Dilute to 1L with DI H₂O and mix. Check the pH and, if necessary, adjust to 6 +0.5. Store the buffer in glass bottle at room temperature

for up to one year. Adjustments to final volume are permitted as long as proportions are maintained.

33.5.1.14 Sodium phosphate, dibasic anhydrous (Na_2HPO_4)

33.5.1.15 Sodium phosphate, monobasic monohydrate ($\text{NaH}_2\text{PO}_4 \cdot \text{H}_2\text{O}$)

33.5.2 MATERIALS

33.5.2.1 Autosampler vials, inserts and caps

33.5.2.2 Disposable 16 x 100mm tubes with closures

33.5.2.3 Disposable screw-cap tubes or centrifuge tubes with closures

33.5.2.4 Disposable pipette tips

33.5.2.5 Extraction column: United Chemical Technologies' Clean Screen SPE cartridge (CSDAU206 200mg/6mL), or equivalent

33.5.2.6 GC column (Agilent HP-5MS; 30 m x 0.250 mm i.d. x 0.250 μm film thickness, or equivalent)

33.5.2.7 Laboratory glassware (graduated cylinders, flasks)

33.5.2.8 Volumetric glassware (flasks)

33.5.3 EQUIPMENT

33.5.3.1 Agilent GC (6890 or equivalent)

33.5.3.2 Agilent MS (5973, or equivalent)

33.5.3.3 Calibrated, adjustable piston pipettes

33.5.3.4 Centrifuge

33.5.3.5 Evaporator (Caliper LS, formerly Zymark, TurboVap)

33.5.3.6 Oven, dry-bath, or wet-bath

33.5.3.7 pH Meter and/or indicating pH paper

33.5.3.8 Rotary mixer

33.5.3.9 Vacuum manifold

33.5.3.10 Vortex mixer

33.6 STANDARDS, CALIBRATORS AND CONTROLS

33.6.1 STANDARDS

33.6.1.1 Reference materials (referred to interchangeably in this method as stock standards) are used for the preparation of working standards which in turn are used to produce calibrators, positive controls and the working internal standard.

33.6.1.2 Stock standards and stock internal standards are purchased from an approved reference material supplier and include the following:

- a. Fluoxetine: 1.0 mg/mL
- b. Fluoxetine-d₆: 0.1 mg/mL
- c. Norfluoxetine: 1.0 mg/mL
- d. Protriptyline: 1.0 mg/mL
- e. Sertraline: 1.0 mg/mL
- f. Desmethylsertraline (norsertaline): 0.1 mg/mL

33.6.1.3 Working standard (10 ng/μL)

- a. Using a calibrated pipette, measure 250 μL each of FLX, NFX and SER, and 2.5 mL of NSR stock standards into a 25 mL class-A volumetric flask.
- b. Add MeOH to the flask to the designated volume.
- c. The final concentration of the working standard is 10 ng/μL. The working standard is stored in the freezer in an amber bottle and expires one year from the date of preparation (if a CRM expires prior to one year from date of preparation, the prepared solution expiration date becomes the first day of the month in which the CRM with the earliest expiration date expires). Volumes may be adjusted, provided that proportions remain constant.

33.6.1.4 Working internal standard (10 ng/μL)

- a. Using a calibrated pipette, measure 2.5 mL FLX-d₆ and 250 μL PPO stock standards into a 25 mL class-A volumetric flask.
- b. Add MeOH to the flask to the designated volume.
- c. The final concentration of the working internal standard is 10 ng/μL. The working internal standard is stored in the freezer in an amber bottle and expires one year from the date of preparation (if a CRM expires prior to one year from date of preparation, the prepared solution expiration date becomes the first day of the month in which the CRM with the earliest expiration date expires). Volumes may be adjusted, provided that proportions remain constant.

33.6.2 CALIBRATORS

33.6.2.1 Calibrators are prepared in certified blank blood at the time of analysis using the working standard. The preparation of the calibrators is detailed in 33.7 SAMPLE PREPARATION. If necessary, calibrators may be prepared in alternate matrices provided that the matrix has been previously determined to not contain any of the compounds tested for by this procedure.

33.6.3 CONTROLS

33.6.3.1 Negative Control

- a. At least one negative whole blood control is tested with every batch. The negative control is prepared using certified blank blood.
- b. When testing different sample types, wherever possible, include a negative control prepared from that matrix. For example, when analyzing whole blood and urine samples the batch shall include at least one negative whole blood control and at least one negative urine control.

33.6.3.2 Positive Controls

- a. Two positive whole blood controls are tested with every batch. The positive controls are prepared using certified blank blood to which the designated volume of control working standard has been added.
- b. Control stock standards are obtained from an approved reference material supplier.
- c. The control stock standards must be either a different lot number or from a different supplier to those used in producing the working standard. If the same lot must be used, the working control standard must be prepared by someone other than the person that prepared the working standard.
- d. The control working standard (10 ng/μL) is prepared as described in 33.6.1.3.
- e. The preparation of the positive whole blood controls is detailed in 33.7 SAMPLE PREPARATION. Alternatively, quality assurance personnel may provide in-house positive controls.
- f. When testing different sample types, wherever possible, include at least one positive control prepared from that matrix.

33.7 SAMPLE PREPARATION

NOTE 1: Stability evaluation in method validation indicates that prepared extracts should be run on the instrument no more than 24 hours after extraction.

NOTE 2: If nortriptyline, citalopram or codeine has been identified in a case sample, an alternative test method must be used for confirmation/quantitation of nortriptyline. If clonidine or trazodone has been identified, an alternative test method must be used for confirmation/quantitation of norfluoxetine.

- 33.7.1 Label a clean 16 x 100mm tube for each member of the test batch. (i.e. Calibrator, control, case sample).
- 33.7.2 Add 4 mL of 0.1M phosphate buffer (pH6) into each tube.
- 33.7.3 Add 1 mL certified blank whole blood into each of the five calibrator tubes, the two positive control tubes and the negative control tube(s).

- 33.7.4 Prepare a 1:10 dilution of the working standard. (1 ng/μL)
- Using a calibrated pipette, combine 0.1 mL of the working standard with 0.9 mL of ACN or MeOH in a labeled tube.
 - Cap and vortex mix. This dilution shall be disposed of after calibrator preparation.
- 33.7.5 Using the working standard and the prepared dilution, spike the calibrators according to the following table.

Calibrator Description	Volume (μL) Added	Working Standard
Calibrator 1 – 25 ng/mL	25	1 ng/μL
Calibrator 2 – 50 ng/mL	50	1 ng/μL
Calibrator 3 - 100 ng/mL	100	1 ng/μL
Calibrator 4 - 500 ng/mL	50	10 ng/μL
Calibrator 5 - 1000 ng/mL	100	10 ng/μL

- 33.7.6 Prepare a 1:10 dilution of the control working standard. (1 ng/μL)
- Using a calibrated pipette, combine 0.1 mL of the working standard with 0.9 mL of ACN or MeOH in a labeled tube.
 - Cap and vortex mix. This dilution shall be disposed of after control preparation.
- 33.7.7 Using the control working standard, spike the positive controls according to the following table.

Control Description	Volume (μL) Added	Control Working Standard
Control 1 - 75 ng/mL	75	1 ng/μL
Control 2 - 800 ng/mL	80	10 ng/μL

- 33.7.8 If in-house positive controls are being used, transfer 1 mL of each into their labeled tubes.
- 33.7.9 Sample 1 mL of each case sample into its respective tube.
- 33.7.10 Add 50 μL of the working internal standard solution to each tube. Final concentration of the internal standard is 500 ng/mL.
- 33.7.11 Cap the tubes and briefly vortex mix. Centrifuge the tubes for 5 minutes between 3000-3500 rpm.
- 33.7.12 Place new, labeled SPE columns into the vacuum manifold.
- 33.7.13 Condition the SPE columns by passing each of the following solvents completely through under force of gravity.

- a. 3 mL MeOH
- b. 3 mL DI H₂O
- c. 2 mL 0.1 M phosphate buffer (pH6)

Do not let columns dry out between each conditioning step.

- 33.7.14 Transfer the contents of each tube to its respective SPE column and allow them to flow through under force of gravity. (Moderate, positive pressure or vacuum may be applied if the flow is insufficient.)
- 33.7.15 Wash the SPE columns by passing by passing each of the following completely through under force of gravity. (Moderate, positive pressure or vacuum may be applied if the flow is insufficient.)
 - a. 3 mL DI H₂O
 - b. 2 mL 0.1 M acetic acid
 - c. 3 mL methanol
- 33.7.16 Dry the columns for 10 minutes under vacuum.
- 33.7.17 Place clean, labeled centrifuge tubes in the collection rack underneath their corresponding SPE columns.
- 33.7.18 Pass 3 mL of elution solvent through each SPE column and collect the extracts.
- 33.7.19 Transfer the tubes to the evaporator and evaporate the extracts to dryness at 40°C. Extracts must be completely dry for efficient chemical derivatization.
- 33.7.20 In a fume hood, add 100 µL ethyl acetate and 50 µL HFBA to each tube and immediately cap and vortex.
- 33.7.21 Incubate the tubes for 30 minutes at 50°C.
- 33.7.22 Remove the tubes from the heat and cool to room temperature. Alternatively, transfer the tubes to a centrifuge and spin for 2 minutes at 2000 rpm.
- 33.7.23 Transfer the tubes to the evaporator and evaporate the extracts to dryness at 40°C. Make sure the extracts are evaporated to dryness before reconstitution.
- 33.7.24 Reconstitute the extracts by the addition of 50 µL ethyl acetate to each tube. Briefly vortex-mix the tubes. If necessary, centrifuge the tubes for 2 minutes at 2000 rpm to collect the extracts at the bottom of the tubes.
- 33.7.25 Transfer the extracts to labeled autosampler vials and cap.

33.8 INSTRUMENTAL PARAMETERS

The instrumental parameters can be found in Appendix A. Prepare a sequence table by first setting the data path in ChemStation to the date of the test. After entering all vial locations, sample descriptions, comments and/or lot numbers in the sequence table ensure that the method listing in the table is SSRIHF for each line.

33.9 DATA ANALYSIS

- 33.9.1 Analysis of the batch data is conducted using ChemStation quantitative instrumental data analysis software.
- 33.9.2 Quantitative calculations are generated by internal standard, multi-point, linear regression with a $1/a$ (inverse of concentration) weighting factor. The calibration curve is updated using the calibrator results for the batch; no historical calibration curves are permitted.
- 33.9.3 Printed reports for each vial in the batch are generated for review along with the updated calibration curves.
- 33.9.4 Technical review of the batch is conducted according to the criteria listed below.

33.10 CRITERIA FOR BATCH ACCEPTANCE

If the analysis of the batch meets the criteria listed below, the results for the specimens are accepted.

33.10.1 CALIBRATORS AND CALIBRATION CURVES

- 33.10.1.1 Chromatographic peaks for FLX, NFX, SER, NSR and internal standards shall appear symmetrical (i.e. no co-elution, split peaks, or shoulders).
- 33.10.1.2 Retention times shall be within $\pm 2\%$ and ion ratios shall be within $\pm 20\%$ of those in calibrator 4. These are inclusive ranges.
- 33.10.1.3 Quantitative results for FLX, NFX, SER and NSR in each calibrator shall be within $\pm 20\%$ of the target value with the exception of calibrator 1 which shall be within $\pm 25\%$ of the target. These are inclusive ranges. Result comparisons will use whole integer, truncated results in units of ng/mL.
- 33.10.1.4 The calibration curves for FLX, NFX, SER and NSR shall have a correlation coefficient ≥ 0.99 .
- 33.10.1.5 The failure to meet any of these criteria for one compound does not invalidate the acceptability of another compound.

33.10.2 CONTROLS

33.10.2.1 The negative control(s) shall not identify FLX, NFX, SER or NSR above limits of detection. Identification is based on a) acceptable retention time matching, b) distinct peaks present for all selected ions, and c) acceptable ion ratios.

33.10.2.2 Positive controls

- a. Chromatographic peaks for FLX, NFX, SER, NSR and internal standards shall appear symmetrical.
- b. Retention times shall be within $\pm 2\%$ and ion ratios shall be within $\pm 20\%$ of those in calibrator 4. These are inclusive ranges.
- c. Quantitative results for FLX, NFX, SER and NSR in each control shall be within $\pm 20\%$ of the target value. These are inclusive ranges. Result comparisons will use whole integer, truncated results in units of ng/mL.
- d. At least one positive control must meet these criteria the batch to be accepted.

33.11 CRITERIA FOR CASE SAMPLE ACCEPTANCE

If the criteria for batch acceptance have been satisfied, the results of individual case samples are deemed suitable for reporting if the following criteria are met.

- 33.11.1 Any chromatographic peak for FLX, NFX, SER or NSR and internal standards shall appear symmetrical.
- 33.11.2 The retention times for FLX, NFX, SER or NSR and internal standards are within $\pm 2\%$ and the ion ratios are within $\pm 20\%$ of those in calibrator 4. These are inclusive ranges.
- 33.11.3 The quantitative result for FLX, NFX, SER or NSR must be within the dynamic range of the test method.
- 33.11.4 When dilutions of case samples are tested, the quantitative result(s) before multiplication shall be within the dynamic range of the test method.
- 33.11.5 If any target compound in a given case sample is outside of the dynamic range it does not invalidate the result for other compounds.

33.12 REPORTING

- 33.12.1 Results for FLX, NFX, SER and NSR are reported in units of milligrams per liter (mg/L).
- 33.12.2 The whole integer, truncated results are converted from ng/mL to mg/L.
- 33.12.3 Converted results are truncated to two significant figures for reporting.
 - a. Example 1: FLX is measured as 642.73 ng/mL.
 - b. The unit conversion step truncates the result to 642 ng/mL and then represents the result as 0.642 mg/L.

- c. The result is truncated to 0.64 mg/L (two significant figures) and reported.
- 33.12.4 Quantitative results for samples that contain one or more target SSRI compounds above the ULOQ will be reported from the sample with the lowest dilution factor that brings all compounds within the dynamic range.
- a. For example, a case specimen has estimated fluoxetine concentration near 4000 ng/mL and norfluoxetine concentration near 2000 ng/mL. The specimen is tested at a 1:4 and 1:10 dilution. The 1:4 dilution result for norfluoxetine is within the dynamic range, but the fluoxetine result is still greater than the ULOQ. The 1:10 dilution results for both fluoxetine and norfluoxetine are within the dynamic range and values for both compounds are reported from the 1:10 dilution sample.

33.13 METHOD PERFORMANCE

- 33.13.1 Limit of detection: 5 ng/mL
- 33.13.2 Lower limit of quantification: 25 ng/mL
- 33.13.3 Dynamic range: 25 – 1000 ng/mL
- 33.13.4 Upper limit of quantitation: 1000 ng/mL
- 33.13.5 Upper limit of linearity: 3000 ng/mL (SER 2500 ng/mL)

33.14 TRACEABILITY

Traceability of the reference materials to SI units is provided through the certificate of analysis provided by the approved reference material supplier.

33.15 REFERENCES

- 33.15.1 Dawn Sklebov, in-house method development.
- 33.15.2 I. Papadatsis, A. Khraiwesh, P. Nikolaou, C. Pistos, C. Spiliopoulou, S. Athanasielis, *A fully validated method for the simultaneous determination of 11 antidepressant drugs in whole blood by gas chromatography – mass spectrometry*, J Pharm. Biomed. Anal. 70 (2012) 557-562.

APPENDIX A
 INSTRUMENTAL PARAMETERS

GAS CHROMATOGRAPH

Split/Splitless Inlet	
Mode	Splitless
Inlet Liner	4mm splitless w/ glass wool plug
Temperature	250° C
Purge Flow	10 mL/min
Purge Time	0.7 min
Gas Type	Helium
Autosampler	
Injection Volume	1.0 µL
Solvent Wash A	3 (Ethyl acetate)
Solvent Wash B	3 (Ethyl acetate)
Sample Pumps	2

Oven / Column	
Carrier Gas Mode	Constant Flow
Carrier Gas Flow	1.5 mL/min
Initial Temperature	150° C
Initial Time	1.00 min
Ramp Rate 1	20° C/min
Final Temp 1	300° C
Final Time	5.0 min
Transfer Line Temp	280 ° C

MASS SPECTROMETER

Solvent Delay	4.00 min	MS Quad Temperature	150° C
EM Offset	Set in tune	MS Source Temperature	230° C
Resolution	Low	Dwell Time	50 msec
Signals	Ions	Ion ratios	
Norfluoxetine	330,117, 226	117/330, 226/330	
Fluoxetine-d6	350,123	123/350	
Fluoxetine	344,117	117/344	
Desmethylsertraline	487,274,239	274/487, 239/487	
Protriptyline	459,191	459/191	
Sertraline	501,274,238	274/501, 238/501	

Archived 5/16/17



LIST OF CHANGES

Revision Date	Description	Page Number
7/13/15	Method approved by Washington State Toxicologist. See DRA dated 7/9/15. Method released for use in evidentiary testing as of 7/13/15.	All
2/4/16	Added note regarding CRM expiration dates to 33.6.1.3 and 33.6.1.4. Edited 33.12.3 to reflect that only two significant figures are used for reporting. Changed wording in 33.12.4 to describe that quantitative results for samples that contain one or more SSRI compounds >ULOQ will be reported from the lowest dilution factor that brings all compounds within the dynamic range, and included an example in 33.12.4.a.	9-10

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