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# 1.0 Training Overview

## 1.1 Training Purpose and Description

### 1.1.1 Purpose and Goals

The purpose of this training program is to provide a uniform training process for analysts in the Toxicology Section at the Vermont Forensic Laboratory (VFL). This program is designed to ensure and document that those individuals who will be working as analysts are knowledgeable and competent to perform their technical, analytical, and legal duties.

### 1.1.2 Scope

This training manual should be completed in conjunction with TOX_P300_Intro to Toxicology and TOX_P306_Toxicology Screening Training Manuals. Training will concentrate on confirmation methods for drugs in whole blood currently in use at the VFL. A final evaluation, including competency testing, written examination, and mock trial will take place following the completion of this manual and TOX_P306. This program is designed for new employees or current employees without prior toxicology experience. A trainee with previous experience in forensic or other toxicology analysis may not require all modules or steps; it is the responsibility of the Toxicology Section Supervisor to determine the duration and scope of the training program for a trainee with previous experience. Similarly, the module content may be tailored as applicable to anticipated job responsibilities.

### 1.1.3 Documentation

The trainee will compile all documentation associated with training work completed. These files may include, but are not limited to, worksheets, reports, and review sheets. The trainer will review these materials and document completion of required training components. Documentation of training will be maintained at the laboratory.

## 1.2 Trainee Responsibilities

### 1.2.1 Instructions for Trainee

The length of time needed to complete the training program will vary and is left to the discretion of the trainer and supervisor. The trainee will be provided access to any required or suggested readings and will be exposed to samples and situations expected to be encountered during routine work in the Toxicology Section. The trainee will keep records, where appropriate, of how training tasks were accomplished (e.g. who did the trainee observe testify in court, what additional papers

not listed in Appendix I did the trainee reference, etc.). At the conclusion of training, the trainee will evaluate the effectiveness of the training program and suggest any improvements to the section supervisor.

### 1.2.2 Required Training Modules

The trainee, trainer, and section supervisor shall discuss which portions of the training manual are to be completed by the trainee based on the trainee's anticipated job responsibilities and prior experience. This section may also be used to outline re-training requirements for current employees if needed. The requirements for the trainee are outlined below:

| | Required? | | Completed? | |
|---|---|---|---|---|
| | Yes | No | Date | Trainer |
| 1. Training Overview | | | | |
| 2. Laboratory Introduction | | | | |
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| 9.1 Competency Tests | | | | |
| 9.2 Mock Trial | | | | |
| 10. Authorization | | | | |

### 1.3 Trainer Responsibilities

The trainer is responsible for instructing the trainee in the operations of the laboratory and the processes and procedures that will ultimately comprise the trainee's job duties. The trainer will ensure that the trainee is exposed to all relevant topics within the training program. The trainer will provide sample sets for the trainee to analyze and will meet with the trainee periodically to monitor progress, review work, and provide feedback. The trainer will assist the trainee in preparing for any assessments, which include a competency test, written examination, and a mock trial. At the conclusion of training, the trainer will evaluate the effectiveness of the training program and suggest any improvements to the section supervisor.

### 1.4 Acknowledgement of Training Plan

The signatures of the trainee, trainer(s), and section supervisor below indicate that the expected responsibilities and required training modules have been discussed and agreed upon.

Trainee: _____ Date: _____

Trainer(s): _____ Date: _____

Trainer(s): _____ Date: _____

Section Supervisor: _____ Date: _____

## 2.0 Laboratory Introduction

Refer to TOX_P300_Intro to Toxicology Training Manual.

## 3.0 Sample and Evidence Control

Refer to TOX_P300_Intro to Toxicology Training Manual.

## 4.0 Fundamental Scientific Knowledge

Refer to TOX_P300_Intro to Toxicology Training Manual.

## 5.0 Applied Scientific Knowledge

This training module will ensure that the trainee has received appropriate education and training to apply principles of sample extraction techniques, liquid chromatography, and mass spectrometry to the analysis of forensic toxicology samples.

### 5.1 General Forensic Toxicology

The trainee will develop and demonstrate a working knowledge of drug pharmacology and physiology as it applies to DUID, including the pharmacokinetics and pharmacodynamics, and impairing effects of various classes of drugs. This section may be completed concurrently with TOX_P306.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| I have read and understand the required readings outlined for this section in Appendix I. | | | |
| I have answered the questions outlined for this section in Appendix II and received feedback on my answers. | | | |

## 5.2 Drug Confirmation Analysis

### 5.2.1 Sample Preparation and Extraction Theory

The trainee will develop and demonstrate a working knowledge of the extraction methods and techniques in use at the VFL.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| I have read and understand the required readings outlined for this section in Appendix I. | | | |
| I have answered the questions outlined for this section in Appendix II and received feedback on my answers. | | | |

### 5.2.2 Liquid Chromatography Theory

The trainee will develop and demonstrate a working knowledge of techniques in use at the VFL to analyze blood samples for drugs, including the use of liquid chromatography.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| I have read and understand the required readings outlined for this section in Appendix I. | | | |
| I have answered the questions outlined for this section in Appendix II and received feedback on my answers. | | | |

### 5.2.3 Mass Spectrometry Theory

The trainee will develop and demonstrate a working knowledge of techniques in use at the VFL to analyze blood samples for drugs, including the use of mass spectrometry.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| I have read and understand the required readings outlined for this section in Appendix I. | | | |
| I have answered the questions outlined for this section in Appendix II and received feedback on my answers. | | | |

### 5.3 Statistics and Data Analysis

The trainee will develop and demonstrate a working knowledge of the quantitative reporting, statistical calculations, and measurement uncertainty factors applied to blood drug confirmation testing in use at the VFL.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| I have read and understand the required readings outlined for this section in Appendix I. | | | |
| I have reviewed the measurement uncertainty budgets for blood drug confirmation analysis. | | | |
| I have answered the questions outlined for this section in Appendix II and received feedback on my answers. | | | |

## 6.0 Laboratory Analysis

The trainee will demonstrate the ability to apply knowledge of the currently validated methods and technologies to the analysis of training samples representing the range and type of samples routinely encountered in casework analysis.

### 6.1 Confirmation Analysis

The trainee will develop and demonstrate knowledge of the instrumentation and procedures used for quantitative drug confirmation, including reagent preparation, sample preparation and extraction, instrument set up and operation, quality control and maintenance of the LC-MS/MS and peripheral instrumentation. The trainee and trainer will document this by both initialing the boxes below.

| Task/Procedure | Initials & Date Observed | Initials & Date Performed |
|---|---|---|
| Mobile phase preparation | | |
| Extraction reagent preparation | | |
| Calibrators/QC stock preparation | | |
| Sample preparation and extraction procedures including use of the positive pressure manifold | | |
| General use of the LC-MS/MS including: software, calibration and QC, startup/shutdown procedures | | |
| Instrument maintenance and record keeping | | |
| I have read and understand the required readings outlined for this section in Appendix I. | | |
| I have answered the questions outlined for this section in Appendix II and received feedback on my answers. | | |
| I have been authorized to use the LC-MS/MS. | | |

### 6.2 Training Sample Processing

The trainee will apply knowledge of the protocols and procedures in use at the VFL to the analysis of training samples before being authorized to analyze casework samples. The trainer will provide a set of relevant, previously analyzed samples for the trainee to process. The trainee will document each analysis performed and provide the trainer with results from the analyses. The trainee will demonstrate an understanding of the limitations of selectivity and sensitivity for each analytical method, be able to select appropriate testing methods for each sample, and apply the appropriate quality control parameters. The trainee and trainer will document this by both initialing the boxes below.

The number and type of samples processed by the trainee will be sufficient to demonstrate the trainee's ability to competently conduct drug confirmation analyses and produce reliable results. If the section supervisor determines that the sample number or type requirements for the trainee differ from those listed below, documentation of this shall be kept with the training materials.

Prior to processing training samples, the trainee will observe a qualified analyst(s) perform the method. At minimum, the samples processed for training will include the following:

1. Under the general supervision of an experienced analyst, analyze 2 batches, containing at least 3 samples each, for each method, from the pool of previously analyzed samples.
2. Independently analyze 2 batches, containing at least 3 samples each, for each method, from the pool of previously analyzed samples.
3. Demonstrate competency by analyzing at least three previously analyzed samples and preparing batch and case specific files for each which will undergo technical review and be approved by the Toxicology Section Supervisor. The acceptance criteria for these samples will be outlined by the section supervisor prior to analysis. Successful completion will be documented with a written authorization specifying that the trainee can now act as a technician for a qualified analyst.
4. Under the supervision of a qualified analyst, analyze 50 casework samples using the Randox Evidence Investigator including confirmation testing on at least 25 of those samples. Each confirmation method should be performed at least once. (The trainee should initial case documents for work that they performed under supervision. The reporting analyst will initial all pages of the examination documentation).

Where appropriate, measurement uncertainty will be applied to the results of these analyses.

| Task/Procedure | Initials & Date Observed | Initials & Date Performed Under Supervision | Initials & Date Performed Independently |
|---|---|---|---|
| THC and Metabolites | | | |
| Opioids and Stimulants | | | |
| Benzodiazepines | | | |
| Other: | | | |
| I have demonstrated technical competency and am authorized to act as a technician. | | | |
| As a technician, I have successfully analyzed at least 50 casework samples. | | | |

## 7.0 Reports and Notifications

The trainee will be familiar with the creation of casework files and reports following the requirements of the Toxicology section.

## 7.1 Report Writing

The trainee will develop and demonstrate knowledge of procedures and documentation requirements for reporting analytical results in accordance with laboratory policy. Training will include, but is not limited to, report format, language used, and the use of the LIMS. The trainee and trainer will document this by both initialing the boxes below.

| Task/Procedure | Initials & Date Observed | Initials & Date Performed Under Supervision | Initials & Date Performed Independently |
|---|---|---|---|
| Generated a blood drug confirmation batch file | | | |
| Generated a blood drug confirmation case file | | | |
| I have answered the questions outlined for this section in Appendix II and received feedback on my answers. | | | |

## 7.2 Review

The analyst trainee will become familiar with the policies, procedures, and forms for technical, administrative, and director review of case files. If the trainee has observed the director review of other toxicology case files, that observation may be used to sign off on the requirement for observing a director review of blood drug confirmation case files.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| I am familiar with the technical review requirements as outlined in the QA and Blood Drug Confirmation Manuals | | | |
| I have observed the technical review of blood drug casework by an experienced analyst. | | | |
| I have performed “pre-review” technical review(s) of blood drug casework. | | | |
| I have observed the administrative review of blood drug case files. | | | |
| I have observed the director review Toxicology case files. | | | |

### 7.3 Mock Cases

The trainer will prepare at least three mock cases for the trainee using previously analyzed samples. The acceptance criteria for these samples will be outlined by the section supervisor prior to analysis. Samples will be screened following the training outlined in TOX_P306 and confirmed for the appropriate method based on those results. Samples may be run in the same batch(es), but unique cases will be created in FA. The trainee will analyze these samples, create case files, perform appropriate calculations, and write reports. The trainer(s) will perform reviews of these cases and the trainee will revise the case files as needed to meet review criteria. Mock cases for confirmation analysis will be completed in conjunction with TOX_P306.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| I have analyzed samples, prepared case files, and had those files reviewed for these mock cases.<br><br>Case Numbers: | | | |
| | | | |
| | | | |
| | | | |

## 8.0 Legal Knowledge

### 8.1 Expert Testimony

The trainee will develop and demonstrate knowledge of the responsibilities of expert witnesses and strategies for effective expert testimony.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| I have read and understand the readings outlined in Appendix I. | | | |
| I have answered the questions outlined for this section in Appendix II and received feedback on my answers. | | | |
| I have observed an experienced analyst testify in court as an expert witness regarding blood drug confirmation testing, including the principles of sample preparation and | | | |

| | | | |
|---|---|---|---|
| extraction techniques, liquid chromatography, and mass spectrometry. Transcripts may be reviewed in lieu of live testimony. | | | |
| I have practiced direct and cross examination pertaining to blood drug physiology, pharmacology, and impairment with more than one qualified analyst and received feedback. | | | |
| I have practiced direct and cross examination pertaining to blood drug confirmation testing with more than one qualified analyst and received feedback. | | | |
| I have practiced presenting a report as an exhibit. | | | |

### 8.2 Document Preparation

The trainee will practice preparing documents that would be requested from an analyst preparing to appear in court.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| I am familiar with what is included in a blood drug discovery packet. | | | |
| I have reviewed at least one blood drug case discovery packet prepared by a qualified analyst or designee. | | | |
| I have prepared a practice blood drug case discovery packet and had it reviewed by the Toxicology Section Supervisor. | | | |

## 9.0 Final Evaluation

At the completion of this training program, the trainee's ability to perform the duties of an analyst will be assessed. The nature of final assessment and evaluation may differ based on the trainee's experience and anticipated job responsibilities. The section supervisor is responsible for determining what assessment and evaluation is necessary for the trainee and documenting this.

Final evaluation, including the competency test, written examination, and mock court, will be completed in conjunction with TOX_P306.

### 9.1 Competency Tests

The trainee will pass all applicable competency tests as determined and documented by the section supervisor prior to beginning work as an analyst. Competency testing includes a written exam as

well as written reports for mock cases that will undergo technical, administrative and director reviews. Satisfactory completion of a competency test is required for all analysts regardless of previous experience. The number and type of samples required for the competency test should be sufficient to cover the anticipated spectrum of assigned duties and to evaluate the individual's ability to perform proper testing methods.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| I have completed all modules of the Toxicology Screening Training Manual (TOX_P306) | | | |
| I have successfully completed all competency tests pertaining to blood drug analysis. | | | |
| I have successfully completed a written examination pertaining to blood drug analysis and received feedback on my results. | | | |
| I have successfully completed a written examination pertaining to drug physiology, pharmacology, and impairment and received feedback on my results. | | | |

### 9.2 Mock Trial

The trainee will understand that each case or sample set examined may potentially require them to testify as an expert witness. As such, the trainee will demonstrate their knowledge of drug testing methods and ability to testify as an expert witness by participating in a mock trial, including both direct and cross-examination. The trainee will be evaluated for aspects of performance to include testimony content, response to cross-examination, demeanor, and attire. The mock trial will take place prior to the trainee completing independent casework as a qualified analyst.

If the trainee has previous testimony experience, this requirement may be modified at the discretion of the section supervisor. This modification will be documented. If a mock trial is not required, an alternate form of oral examination must be selected by the section supervisor.

The trainee will testify to a competency test case, a fabricated case, or a case which has previously been completed by a qualified analyst. The case selected will be agreed upon by the trainee and trainer and approved by the section supervisor.

The mock trial will include, but is not limited to, questions on qualifications, chain of custody, evidence handling, drug screening and confirmation analysis, measurement uncertainty, and technical aspects of the case at hand. Questioning by both the prosecutor and defense attorneys should be relevant and realistic. The atmosphere of the trial will be formal and will be conducted in the same manner as a real courtroom, including conduct and protocol. The trainee should present themselves accordingly. The outcome of the mock trial evaluation will be satisfactory or unsatisfactory. If it is determined that the trainee's performance was not satisfactory, the section supervisor will determine what corrective action must be taken. The trainee will need to complete a mock trial with satisfactory performance before beginning work as a qualified analyst. Participants in the mock trial shall provide feedback for the trainee regardless of whether performance was satisfactory or unsatisfactory.

| Task | Trainee | Trainer | Date Completed |
|---|---|---|---|
| A case has been chosen for mock court.<br>Case Number: | | | |
| I have offered testimony in mock court, including both direct and cross-examination regarding the analysis of blood samples for drugs, including immunoassay screening, sample preparation and extraction, liquid chromatography and mass spectrometry. | | | |
| I have offered testimony in mock court, including both direct and cross-examination regarding drug physiology, pharmacology, and impairment. | | | |

## 10.0 Authorization

Completion of appropriate training modules to support this determination will be documented in section 1.2.2. At the completion of training, the section supervisor shall provide written documentation authorizing the trainee to conduct independent casework as a qualified analyst.

# Appendix I: Readings

## A: Required Readings

The resources listed below are provided to give the trainee the necessary information to perform their duties. Depending on the trainee's education and experience, they may not need to complete every reading in its entirety. The location of various training tools such as readings, videos, or websites are included for convenience. If file locations change or links break, alternative training materials may be used. Supplemental documents are available through the laboratory's shared drive under the training folder, but are not required unless designated as such by the section supervisor. The trainee should be familiar with the readings provided in the TOX_P300 manual in order to assist them in answering the questions in Appendix II.

## 5.0 Applied Scientific Knowledge

### 5.1. General Forensic Toxicology

- ☐ Baselt, Randall C. Disposition of Toxic Drugs and Chemicals in Man.
- ☐ Baselt, Randall C. Drug Effects on Psychomotor Performance.
- ☐ Advanced Roadside Impaired Driving Enforcement training
- ☐ Drug Recognition Expert 7-day Course manual. NHTSA
- ☐ Brunton, Laurence, Chabner, Bruce, and Knollman, Bjorn. Goodman and Gilman's The Pharmacological Basis of Therapeutics.
- ☐ Levine B., Principles of Forensic Toxicology.
- ☐ Negrusz, Adam and Cooper, Gail, ed. Clarke's Analytical Forensic Toxicology. London, UK: Pharmaceutical Press.
- ☐ Klaassen, Curtis D., ed. Casarett and Doull's Toxicology: The Basic Science of Poisons.
- ☐ Raymon, Lionel P. Clinical Pharmacology. Downers Grove IL: Becker Professional Education.
- ☐ Maurer, H. "How Can Analytical Diagnostics in Clinical Toxicology Be Successfully Performed Today?" The Drug Monit vol. 34 (5), October 2012.
- ☐ Maurer, H. "Mass spectrometric approaches in impaired driving toxicology". Anal Bioanal Chem 393, 2009.

### 5.2. Drug Confirmation Analysis

#### 5.2.1. Sample Preparation and Extraction Theory

- ☐ Beginner's Guide to SPE. Waters Corporation, 2012
- ☐ Sample Preparation techniques, Tox digital library; CHROMacademy.

#### 5.2.2. Liquid Chromatography Theory

- ☐ Fundamentals of HPLC. Waters Corporation, 2014.
- ☐ Beginner's guide to UPLC. Waters Corporation, 2009.
- ☐ Maurer, Hans. What is the Future of (Ultra) High Performance Liquid Chromatography Coupled to Low and High Resolution Mass Spectrometry for Toxicological Drug Screening? J Chrom A, 2012.
- ☐ The theory of HPLC, CHROMacademy
- ☐ Quantitative and Qualitative HPLC, CHROMacademy
- ☐ Acquity H-Class QSM Overview and Maintenance Guide.

- ☐ Acquity H-Class SM/FTN Overview and Maintenance Guide.
- ☐ Webcast on HPLC troubleshooting, CHROMacademy

#### 5.2.3. Mass Spectrometry Theory

- ☐ The Mass Spectrometry Primer. Waters Corporation, 2009.
- ☐ Xevo TQ-S Micro Overview and Maintenance Guide.
- ☐ Electrospray Ionization Theory under Fundamentals LC-MS, CHROMacademy
- ☐ Levine B., Principles of Forensic Toxicology: Mass Spectrometry

### 5.3. Statistics and Data Analysis

- ☐ Tox Data Summary spreadsheet
- ☐ Confirmation Testing Fishbone
    - THC
    - BNZ
    - OPS
- ☐ Current and historical MU budgets
    - THC
    - BNZ
    - OPS

## 6.0 Laboratory Analysis

### 6.1. Confirmation Analysis

- ☐ Toxicology Confirmation Manual TOX_P700
- ☐ Confirmation of THC and Metabolites TOX_P701
- ☐ Confirmation of Opiates, Opioids and Stimulants TOX_P702
- ☐ Confirmation of Benzodiazepines TOX_P703
- ☐ Confirmation Validations
    - THC
    - OPS
    - BNZ
- ☐ Blood drug reagent preparation log
- ☐ LC-MS/MS instrument maintenance log
- ☐ Instrument Shutdown procedures

## 8.0 Legal Knowledge

### 8.1. Expert Testimony

- ☐ Guidelines for Opinions and Testimony in Forensic Toxicology. ANSI/ASB Best Practice Recommendation 037, First Edition 2019.
- ☐ Logan, B et al. “Recommendations for Toxicological Investigation of Drug-Impaired Driving and Motor Vehicle Fatalities”. J Anal Tox (most recent version).
- ☐ Principles of Pharmacology: Pharmacokinetics and Pharmacodynamics of Drugs.
- ☐ Review various sample scripts of chemist questions for blood drug screening testimony

## B: VFL Toxicology Reference Library

Analysts are expected to be familiar with the contents of the VFL Toxicology Reference Library and be alert for articles and/or references that can be added. Updated references should be added to the VFL Toxicology Reference Library during the review period, when they become available, or when new methodologies or technologies are incorporated into the laboratory protocols.

# Appendix II: Training Questions

Questions listed below are intended to be answered by analyst trainees. Headings are numbered in accordance with the section numbering in the body of this manual.

## 5.0 Applied Scientific Knowledge

### 5.1. General Forensic Toxicology

1. What are the most common routes of drug administration?
2. What is bioavailability? Give an example of how bioavailability may change depending on the route of administration.
3. What processes play a role in drug absorption? What factors might affect the rate of absorption?
4. What is the difference between pharmacology, pharmacokinetics, and pharmacodynamics?
5. How are drugs distributed throughout the body? Why are drugs generally more lipid-soluble than water-soluble?
6. Explain the concept of Volume of Distribution (Vd).
7. What is first pass metabolism?
8. What processes contribute to the biotransformation of drug compounds? What is the difference between Phase I and Phase II biotransformation?
9. What is "half-life" ($t_{1/2}$)? How does $t_{1/2}$ affect the rate of biotransformation of drugs?
10. Describe the basic pharmacokinetic models (zero-order, first-order, and Michaelis-Menton).
11. What are the routes of drug elimination? What factors might affect the rate of elimination?
12. What is the relationship between drug dose and response?
13. What is a therapeutic index (TI) and what does it indicate about a particular drug? Give an example of a drug with a low TI and one with a high TI.
14. Discuss the difference between potency, efficacy, and affinity. By what mechanisms might the potency, efficacy, and affinity of a drug be modulated?

### 5.2. Drug Confirmation Analysis

#### 5.2.1. Sample Preparation and Extraction Theory

1. What is the purpose of performing an extraction on a whole blood sample?
2. What are functional groups? Explain their role in sample extraction.
3. What is pKa?
4. pH should be how many units above or below the pKa of the compound of interest? Why?
5. How is pH adjusted in whole blood samples?
6. What are the most common types of SPE phases? Which one(s) would you use to extract acidic and basic drugs?
7. How rapidly should samples be eluted from a column when using a positive pressure manifold?
8. In what solvent(s) are samples typically reconstituted after elution or evaporation? Why?
9. How are samples prepared at the VFL?

### 5.2.2. Liquid Chromatography Theory

1. Why is confirmation of a presumptively positive sample necessary for forensic drug testing?
2. Describe the principles of liquid chromatography and the general operation of an LC system.
3. How do liquid chromatography, HPLC, and UHPLC/UPLC differ?
4. What is normal phase liquid chromatography? Reverse phase?
5. Discuss the difference between isocratic and gradient liquid chromatography.
6. What factors can affect resolution in a liquid chromatographic system?

### 5.2.3. Mass Spectrometry Theory

1. Describe the path a target compound molecule takes through the tandem MS instrument.
2. Why is ionization important in mass spectrometry? List some common techniques used.
3. How does a quadrupole affect the flight path of an ionized molecule?
4. What is the function of the collision cell? What gas is used for this purpose?
5. What is MRM? How does it differ from SIM and Full Scan modes?
6. What is fragmentation? What molecules are being observed during analysis?
7. What is “unit mass” resolution? How does it differ from “high” resolution?
8. Describe how one might differentiate configurative isomers, e.g. 4-methoxyamphetamine and ephedrine. What are some other examples of compounds that might be difficult to differentiate?
9. Is MS able to differentiate stereoisomers? Why or why not?

## 5.3. Statistics and Data Analysis

**Statistical Analysis**

1. How many calibration points are required for a calibration curve based on the ASB guidelines?
2. How are drug results rounded and truncated?
3. How are results reported?

**Excel**

1. How does Excel source the raw data file into the Tox Data Summary?

**Measurement Uncertainty**

1. How is the measurement uncertainty of the blood drug methods estimated?
2. How often is the budget updated?
3. What is the timeframe of the data used in the budget?
4. What confidence interval is applied for confirmation testing?
5. What contributors are evaluated?
6. Which uncertainty value do we input for the pipettes? From Cerilliant/Lipomed COAs?

# 6.0 Laboratory Analysis

## 6.1. Confirmation Analysis

1. Describe the preparation, quality control, and documentation procedures as described in the confirmation manuals for each of the following:
   a. Mobile phases
   b. Extraction reagents

c. Purchased reagents
d. Calibration and QC stocks

2. Describe which reagents are required in each confirmation panel and their preparation for each of the following processes:
   a. Sample pre-treatment
   b. Solid phase extraction
   c. Liquid chromatography
3. Describe the quality control procedures that are used to ensure the LC-MS/MS system is acceptable for use in casework.
4. Why does the LC-MS/MS system get calibrated for each analyte? Describe the calibration procedure.
5. What is an internal standard? Why are internal standards used in the quantitative analysis of drug compounds?
6. What are the maintenance requirements of the LC-MS/MS system and the frequency with which they must be performed?
7. Describe the procedure for cleaning the ESI source.

## 7.0 Reports and Notifications

### 7.1. Report Writing

1. What documents are included in blood drug batch and case files?
2. How does the VFL communicate what testing was performed to our customers?
3. What is the language used for samples above or below the reporting range?
4. What information must be included on a blood drug report?
5. Give some examples of how the VFL communicates to our customer when we analyze a sample but we are not reporting results.

## 8.0 Legal Knowledge

### 8.1. Expert Testimony

1. Define the following using language that would be appropriate for a jury:
   a. Drug
   b. First-pass metabolism
   c. Blood-brain barrier
   d. Glucuronidation
   e. Non-competitive antagonist
   f. Synergism
   g. Therapeutic index
2. Can a correlation be drawn between drug levels in blood and level of impairment? Why or why not?
3. Define the following using language that would be appropriate for a jury:
   a. Solid phase extraction
   b. Liquid chromatography
   c. Electrospray Ionization
   d. Tandem quadrupole mass spectrometry
   e. Product ion

f. Peak area

4. If a sample screens positive but confirms negative for an analyte, what might that indicate about the sample? What about the opposite?

| DOCUMENT HISTORY | | | |
|---|---|---|---|
| **DATE** | **VERSION** | **APPROVED BY** | **ACTIVITY OR REVISION** |
| 7/18/2024 | 1 | Lab Director | New document; content previously part of TOX_P301_Toxicology Training Manual |
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