Department of Forensic Science

TOXICOLOGY TRAINING MANUAL

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1. INTRODUCTION

1.1 Purpose and Scope

1.1.1 The purpose of this manual is to define the training program for forensic lab specialists, forensic scientists, and toxicologists working in the Toxicology section as employees of the Commonwealth of Virginia Department of Forensic Science. This work is intended to be used in a formal training program that will establish a certain minimum standard of professional competency throughout the Toxicology section statewide.

1.1.2 The manual is organized in modules and each module outlines the objectives, methods of instruction, modes of evaluation, and study questions.

1.1.3 The training program covers theory and methodology of instrumentation, analytical techniques, interpretation of analytical results, report writing, data and case review, and handling of evidence.

1.1.4 The training program provides exposure to courtroom testimony and legal aspects throughout the training and assists in developing the skills necessary to be an effective expert witness.

1.1.5 The program evaluates the progress and performance of the trainee with each module. Each module includes laboratory exercises, competency tests, and study questions. Upon completion of each module, the trainee will give an oral presentation on the module material which will be followed by a question/answer session to ensure the trainee understands the module material.

1.1.6 The sequence in which the modules are presented should not necessarily be considered as a mandatory order of instruction.

1.1.7 The trainee will complete a mini-technical examination after the first 6 modules and a second mini-technical final on the remaining modules.

1.1.8 It is recognized that some roles within the Toxicology section may only perform certain analyses or functions therefore, the following is a suggested list of modules to be completed by each role. This may be modified by the training coordinator (TC), Section Supervisor, and Program Manager (PM) as necessary to meet the goals of the fully trained position.

Module	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17
Forensic Laboratory Specialist	X	X	X	X	X	X	X	X*				X**				
Forensic Scientist	X	X	X	X	X	X	X	X	X	X	X	X**	X	X		
Forensic Toxicologist	X	X	X	X	X	X	X	X	X	X	X	X**	X	X	X	X

*Module 9 may be completed for Forensic Laboratory Specialists dependent upon their role.

**See 1.1.9 below for Alcoholic Beverage Analysis training.

1.1.9 Any member of the toxicology section who performs examinations of alcoholic beverages will be required to complete Module 13 (Alcoholic Beverage Analysis).

1.1.10 For Modules 9, 10, and 11, the laboratory exercises and competencies may be combined into one assignment that fulfills all requirements for each exercise/competency in those modules.

1.1.11 The program culminates in the final competency exercise which includes a practical test, an analytical technical oral examination, and a mock trial. For forensic toxicologists, the final competency also includes a pharmacology/toxicology oral examination.

1.2 Coordination of the Program

1.2.1 The TC is usually a supervisor in each laboratory however, this role may be delegated to a competent, experienced Forensic Scientist or Forensic Toxicologist.

1.2.2 The coordinator will be responsible for the overall training but may delegate certain duties and blocks of instruction to other individuals.

1.3 Training Period

1.3.1 The length of the training period is a highly variable matter and will be left to the determination of the TC and the PM. Some individual trainees may require less time than others depending on experience, education, and learning ability. The training period is usually completed within 12 months for forensic scientists and 18 month for forensic toxicologists.

1.4 Location of Training

1.4.1 Whenever practical, the bulk of an individual's training will occur in the laboratory to which they will be assigned.

1.5 Training Goals

The training should culminate such that the trainee has the following:

1.5.1 The knowledge of analytical chemistry.

1.5.2 The knowledge of the principles and practices of forensic toxicology related to the analysis of drugs and poisons within biological samples.

1.5.3 The knowledge of the theory and application of a variety of instruments used for the identification and quantitation of drugs.

1.5.4 The ability to perform accurate forensic toxicology analyses independently and proficiently.

1.5.5 The ability to skillfully present and defend analytical findings in courts of law.

1.6 Instructions to the Trainee

1.6.1 The trainee is expected to document all their training activity and to provide a weekly progress report to the training coordinator. The progress report should also include upcoming training goals.

1.6.2 Throughout the training period, the trainee will observe data reviews and may perform the data reviews of their own batches. The trainee may also conduct a practice batch review on casework batches however these reviews are not documented on the casework samples. This practice will expose the trainee to a wider variety of scenarios than they may be exposed to solely with practice, exercises, and competency samples.

1.6.3 Once the trainee has demonstrated their competence to perform a particular analysis through the completion of specific training module(s), the trainee may be authorized by the PM on recommendation by the Section Supervisor to perform those analyses on case work. This authorization will be documented via MFR. For Evidence Receiving and Handling, the trainee will be added to the Delegation of Authority letter. Batch data run by trainees must be reviewed by a qualified examiner and this review will documented on the batch summary worksheet. Trainees may not act as batch reviewers.

1.7 Instructions to Training Coordinators

1.7.1 As previously stated, the intent of the training manual is to define a program that will ensure each and every trainee receives certain basic principles and fundamentals necessary to the complete education

of lab specialists, forensic scientists, or toxicologists within the Toxicology section. All of the listed topics must be incorporated into the program for forensic scientists and toxicologists (refer to QM).

1.7.2 It is recognized that some of the forensic laboratory specialists may only perform certain analysis. Therefore, they are only required to complete the modules associated with the type of work they perform, not necessarily all of the modules throughout the training program (refer to 1.1.8).

1.7.3 The trainee is not permitted to perform supervised casework. "Supervised casework" is defined as the trainee performing active casework analysis under the direct supervision of a qualified examiner prior to successfully completing a competency exam.

1.7.4 Samples used for laboratory exercises may consist of blind controls, spiked samples, simulated case samples, or de-identified aliquots of appropriate samples retained for quality assurance purposes. Samples for competency exams shall, to the best of the TC's ability, consist of de-identified aliquots or spiked authentic matrix.

1.7.5 The training coordinator is responsible for maintaining the Department's training program documentation during the training period. Each section of the training log must be dated and initialed upon the start and completion of the module. If any task is not completed, for any reason, this must be explained in the training file and approved by the PM.

1.7.5.1 Some required readings may not be available or are not published at this time therefore the TC may substitute readings/information from appropriate sources. Appropriate sources include textbooks, peer-reviewed articles, informational videos, and websites.

1.7.6 Once the trainee has satisfactorily completed all of the requirements of the program, the PM shall forward a written recommendation for certification to the Department Director.

1.7.7 If the trainee cannot meet the criteria expected of them during the training period, steps must be taken to determine and enact appropriate action.

1.7.8 The performance of the trainee will be evaluated during the course of the program. The evaluation will include the monthly training reports addressed in the QM. The TC must submit the monthly evaluations through the Qualtrax workflow (if available). This workflow should facilitate a discussion of this evaluation between the TC and the trainee prior to forwarding it. This discussion should include both preivous accomplishments, deficiencies, and future objectives. This report will be maintained within Qualtrax.

1.8 Mock Trials

1.8.1 The training coordinator is responsible for ensuring that the trainee is thoroughly prepared for legal questioning. This can be done by a combination of mock trials, prearranged as well as impromptu question and answer sessions, and observation of courtroom testimony given by experienced examiners.

1.8.2 The scheduling of practice mock trials is to be done by the TC. These are to be conducted throughout the training period.

1.9 Guidelines for Technical Examinations, Practical Test, and Final Mock Trial

1.9.1 Technical Examination

1.9.1.1 Prior to the final mock trial, a technical oral examination of the trainee will be conducted to ascertain the analytical knowledge of the individual. This will be limited to 3 hours.

1.9.1.2 After the examination, the evaluating members of the audience (minimally, the PM and TC) will discuss the trainee's performance.

1.9.1.3 The outcome of the examination will be satisfactory or not satisfactory.

1.9.1.4 If the panel determines that the trainee's performance was not satisfactory, steps must be taken to determine and enact an appropriate action.

1.9.2 Pharmacology Technical Examination (Toxicologists only)

1.9.2.1 Prior to the final mock trial, a pharmacology technical oral examination of the toxicologist trainee will be conducted to ascertain their knowledge of pharmacology, toxicology, and interpretation of results. This will be limited to 3 hours.

1.9.2.2 After the examination, supervision/management will discuss the trainee's performance.

1.9.2.3 The outcome of the examination will be satisfactory or not satisfactory.

1.9.2.4 If the panel determines that the trainee's performance was not satisfactory, steps must be taken to determine and enact an appropriate action.

1.9.3 Practical Test

1.9.3.1 Following successful completion of all training modules, the trainee will be given a practical test to work as though it were a real case.

1.9.3.2 The practical test will be a typical case involving at least 3 analytical procedures (e.g., alcohol screen, immunoassay screen, and confirmation/quantitation).

1.9.3.3 Acceptable performance is ±20% of the expected values of drug analyses. Acceptable performance for the alcohol analyses is ±0.004% w/v or 6%, whichever is greater, of the expected value.

1.9.3.4 The trainee will generate an associated case file and Certificate of Analysis for the practical test.

1.9.4 Mock Trial

1.9.4.1 A video-recorded final mock trial will be conducted regarding the analysis (and interpretation for Forensic Toxicologists) of the practical test.

1.9.4.2 The PM must agree with the selection of all participants.

1.9.4.3 The atmosphere will be formal, that is, it will be conducted in the same manner as a real courtroom situation. This includes dress, conduct, protocol, and all other aspects. Answers and explanations are to be directed as to a lay jury or judge.

1.9.4.4 The mock trial will not exceed 2 hours.

1.9.4.5 The role of the prosecutor will be assumed by the TC or designee.

1.9.4.6 The mock trial may be stopped at any time upon request of any of the involved parties.

1.9.4.7 After the court, supervision/management will assess the trainee's performance.

1.9.4.8 The outcome of the mock trial will be satisfactory or not satisfactory.

1.9.4.9 If the panel determines that the trainee's performance was not satisfactory, steps must be taken to determine and enact an appropriate action.

1.9.4.10 This evaluation will be immediately followed by a short performance critique.

1.9.4.11 The training coordinator will review the recording of the trial with the trainee as soon as possible. Other participants/observers should provide comments to the TC prior to this review.

1.10 Transition from Trainee to Examiner

1.10.1 After the new examiner has successfully completed this training, there follows a period of adjustment. The job of the TC is to ensure that this transition from trainee to qualified examiner takes place as smoothly as possible.

1.10.2 Casework will be introduced stepwise under the close supervision of a senior examiner.

1.10.3 The supervisor, TC, or designee will accompany and monitor the newly qualified examiner to court for the first few cases.

1.11 Continuing Education

1.11.1 All forensic lab specialists, forensic scientists, and toxicologists should participate in continuing education to maintain their skills and up-to-date knowledge in the field of forensic toxicology.

1.11.2 Examples of continuing education include:

1.11.2.1 Attendance at meetings, workshops, or seminars.

1.11.2.2 Participation in study groups or scientific working groups.

1.11.2.3 Review of current literature.

1.11.2.4 Publication or presentation of research or case reports.

1.11.2.5 Education/training/teaching in the field of forensic toxicology.

1.11.2.6 Participation in specialized courses.

2 ORIENTATION

2.1 Minimum Requirements for Orientation

2.1.1 Introduction to local operating facilities and personnel.

2.1.2 Coverage of the following:

2.1.2.1 Quality Manual

2.1.2.2 Administrative Policies

2.1.2.3 Regional Operating Procedures

2.1.2.4 Toxicology Procedures Manual

2.1.2.5 DFS Safety Manual

2.1.2.6 Organization of the Department of Forensic Science

2.1.3 Introduction to the technical capabilities of all regional laboratories.

2.1.4 Explanation of the purpose of the training program including an insight into the course of events and what the trainee is expected to accomplish.

2.1.5 Explanation of the operations of local, state, and federal law enforcement agencies and court systems.

2.1.6 Clarification of the duties of forensic laboratory specialists, forensic scientists, and toxicologists within the Section.

2.1.7 Introduction to the LIMS system.

3 EVIDENCE RECEIVING AND HANDLING

3.1 Objectives

3.1.1 Understand physical evidence handling procedures used by DFS as detailed in the Quality Manual.

3.1.2 Understand physical evidence handling procedures pertinent to the Toxicology section.

3.1.3 Receive and process evidence for the Office of the Chief Medical Examiner (OCME), driving under the influence (DUI/DUID), and Toxicology-Other (police or TO) cases.

3.2 Methods of Instruction

3.2.1 Lectures and/or Self-Directed Study

3.2.1.1 Receiving and processing evidence.

3.2.1.2 Evidence security.

3.2.1.3 Chain of custody.

3.2.1.4 LIMS system.

3.2.2 Required Reading

3.2.2.1 DFS Quality Manual.

3.2.2.2 Toxicology Procedures Manual.

3.2.2.3 Code of Virginia §§18.2-266, 18.2-268.1 – 18.2-268.7, 18.2-269.

3.2.3 Demonstration

3.2.3.1 Evidence receiving and processing will be observed from beginning to end and notes will be taken by the Trainee.

3.2.4 Initial Competency

3.2.4.1 The trainee will receive and process 5 simulated cases (a mixture of DUI/DUID, OCME, TO).

3.2.4.2 Succesful completion of this task will be recorded on the "Toxicology Training Module Documentation Form" within the Evidence Handling Comment Grid with the TC's initials and date of completion (e.g., "Initial competency completed 5/21/2020").

3.2.4.3 Upon successful completion of the initial competency, the trainee will be approved by the TC to complete the Laboratory Exercises with casework samples.

3.2.5 Laboratory Exercises

3.2.5.1 The trainee will receive and process evidence for at least 20 OCME or Toxicology-Other samples and 20 DUI/DUID samples.

3.2.5.2 The trainee will seal at least 10 OCME or TO cases.

3.2.5.3 The trainee will maintain a list of processed samples for the training file.

3.3 Evaluation

3.3.1 Completion of written study questions.

3.3.2 Oral presentation followed by technical question/answer session.

3.4 Study Questions

3.4.1 List all procedural steps involving evidence receiving to final disposition for each of the following: DUI/DUID, OCME, TO cases.

3.4.2 Define the following terms: chain of custody, lock box, evidence seal, convenience packaging, RFLE, FS Lab#, LIMS.

3.4.3 Define a proper seal.

3.4.4 Who has access to the main evidence storage room? Toxicology storage refrigerators?

3.4.5 Who has access to your work area?

3.4.6 What actions are taken to ensure the proper preservation of evidence?

3.4.6.1 What are the acceptable temperatures for refrigerators? Freezers?

3.4.6.2 How often must the temperature be checked?

3.4.7 When is evidence returned to the originating agency?

3.4.8 List commonly encountered problems associated with the receipt of evidence and subsequent actions taken.

3.4.9 Specify the official chain-of-custody record for the following:

3.4.9.1 Submission of a DUI/D case with an RFLE.

3.4.9.2 Submission of a DUI/D case without an RFLE.

3.4.9.3 Submission of an OCME case.

3.4.9.4 Placement of DUI/D samples into section storage.

3.4.9.5 Removal of an item from section storage for analysis.

3.4.9.6 Return of item to section storage after analysis.

3.4.10 List the prioritization of samples for an OCME case.

3.4.11 Describe what actions to take if you receive a low volume sample.

4 BASIC LABORATORY SKILLS

4.1 Objectives

4.1.1 Understand general chemistry principles required for preparation of laboratory solutions.

4.1.2 Understand proper use, maintenance, and quality assurance requirements for basic laboratory equipment (i.e., thermometers, heat blocks, turbovaps, pipettes, balances, glassware).

4.1.3 Demonstrate proficiency for using basic laboratory equipment.

4.1.4 Understand preparation of sample homogenates.

4.1.5 Demonstrate proficiency for preparation of samples homogenates (where applicable, to include homogenization of clotted blood specimens and/or alternative matrices).

4.1.6 Understand the uncertainty of measurement including how it is calculated and explained in court (see Uncertainty of Measurement).

4.2 Methods of Instruction

4.2.1 Lectures and/or Self-Directed Study

4.2.1.1 Basic Laboratory Equipment Use and Quality Assurance.

4.2.1.2 Reagent Preparation.

4.2.1.3 Drug Standards Preparation.

4.2.1.4 Sample Homogenates and Dilutions.

4.2.2 Required Reading

4.2.2.1 Toxicology Procedures Manual.

4.2.3 Demonstration

4.2.3.1 Use of the pH meter.

4.2.3.2 Pipette use, to include (where applicable) positive displacement pipettes, air displacement pipettes, and/or serological pipettes.

4.2.3.3 Required documentation of reagent and drug standards preparation and quality assurance logs for basic laboratory equipment.

4.2.3.4 Reagent preparation.

4.2.3.5 Drug standards preparation (should include single component and multi-component preparations of calibrator, QC, or internal standard).

4.2.3.6 Sample homogenization and dilution.

4.2.4 Laboratory Exercises

4.2.4.1 Perform an intermediate (i.e. gravimetric) check on at least 3 calibrated pipettes of varying volume capacities (e.g., 10–100 µL, 100–1000 µL, 1–5 µL).

4.2.4.2 Perform a monthly balance check.

4.2.4.3 Calibrate the pH meter for use.

4.2.4.4 Create a mock reagent log sheet and properly document preparation for a specified reagent of the TC or designee's choice. Training Coordinator or designee will review the preparation log,

and any additional supporting documentation, for accuracy and completeness and document their review on the log.

4.2.4.5 Prepare a single or multi-component drug standard using appropriate equipment and documentation. Training Coordinator or designee will review the preparation log, and any additional supporting documentation, for accuracy and completeness and document their review.

4.2.4.6 Prepare appropriate documentation for a sample homogenate. Where or if possible, perform a sample homogenization.

4.2.4.7 Dilute a blood or urine sample at ¼ and 1/10. Dilute a blood specimen for a diphenhydramine quantitation at 1/50 and 1/100. Blank blood may be used if authentic samples are not available.

4.3 Evaluation

4.3.1 Completion of written study questions.

4.3.2 Laboratory Competency Testing

4.3.2.1 Prepare two reagents, at least 1 requiring the use of the pH meter. The reagents must be considered acceptable for use upon verification by a qualified analyst. Training Coordinator or designee will review the preparation log, and any additional supporting documentation, for accuracy and completeness and document their review.

4.3.2.2 Prepare a calibrator, QC, and internal standard. Each must be considered acceptable for use upon verification by a qualified analyst. Training Coordinator or designee will review the preparation log, and any additional supporting documentation, for accuracy and completeness and document their review.

4.3.3 Oral presentation followed by technical question/answer session.

4.4 Study Questions

4.4.1 What is NIST? Why is it important?

4.4.2 Solution Preparation

4.4.2.1 How much conc. HCl is required to make 500 mL of 1 M HCl? Describe the preparation procedure in detail.

4.4.2.2 How much NaOH is required to make 1 L of a 1 M solution? Describe the preparation procedure in detail.

4.4.3 What is the difference between “to deliver” (TD) and “to contain” (TC)?

4.4.4 Describe the DFS Toxicology quality assurance requirements and intervals for each of the following laboratory equipment:

4.4.4.1 Pipettes

4.4.4.2 Balances

4.4.4.3 Thermometers

4.4.4.4 Heat blocks

4.4.4.5 Evaporators

4.4.4.6 Refrigerators

4.4.4.7 Freezers

4.4.4.8 Diluters

4.4.5 Describe the preparation of sample homogenates to include clotted blood samples, gastric content, and a tissue.

4.4.6 Describe the preparation requirements for each of the following:

4.4.6.1 Reagent preparation

4.4.6.2 Calibrator preparation

4.4.6.3 QC preparation

4.4.6.4 Internal standard preparation

4.4.7 What are the general recommendations for dilutions in the Toxicology section? What is the maximum dilution for a blood sample for methamphetamine quantitation? THC quantitation? Where are the maximum allowable dilutions defined?

4.4.8 What are the conventions for notating dilutions? What is the difference between 1:4 dilution and ¼ dilution?

4.4.9 Explain the ethanol interconversion between mg/L, mg/dL, µg/mL, and gm%. Present 5 examples of each.

5 UNCERTAINTY OF MEASUREMENT

5.1 Objectives

5.1.1 To familiarize the trainee with traceability and its associated concepts.

5.1.2 To familiarize the trainee with concepts of uncertainty of measurement.

5.2 Methods of Instruction

5.2.1 Lectures and/or Self-Directed Study

5.2.1.1 Presentations prepared by Research Section Supervisor and DFS, available in Qualtrax.

5.2.2 Required Reading

5.2.2.1 ASCLD/LAB Policy on Measurement Uncertainty (AL-PD-3060)

5.2.2.2 ASCLD/LAB Policy on Measurement Traceability (AL-PD-3057)

5.2.2.3 ASCLD/LAB Guidance on Measurement Traceability (AL-PD-3058)

5.2.2.4 ASCLD/LAB Guidance on Estimation of Measurement Uncertainty – Overview (AL-PD-3061)

5.2.2.5 ASCLD/LAB Guidance on Estimation of Measurement Uncertainty – ANNEX A: Details on the NIST 8 Step Process (AL-PD-3062)

5.2.2.6 ASCLD/LAB Guidance on Measurement Traceability – Measurement Assurance (AL-PD-3059)

5.2.2.7 ASCLD/LAB Guidance on the Estimation of Measurement Uncertainty – ANNEX D: Toxicology Testing Discipline Example – Concentration of Ethanol in Antemortem Blood Specimen.

5.2.2.8 Toxicology Procedures Manual

5.2.2.9 Optional Additional Resources

5.2.2.9.1 Measurement Uncertainty in Forensic Science – A Practical Guide. Bell, Suzanne. CRC Press – Taylor and Francis Group. Copyright 2017.

5.2.2.9.1.1.1 Chapters listed by title (due to potential changes between editions).

5.2.2.9.2 Forensic Measurements, Metrology, and Uncertainty

5.2.2.9.2.1 Sources of Uncertainty

5.2.2.9.2.2 Foundational Concepts

5.2.2.9.2.3 Processes and Procedures

5.2.2.9.3 Evaluation of measurement data – Guide to the expression of uncertainty in measurement. BIPM. First Edition, 2008. (Website, Alternate Website (accessed October, 7, 2020)).

5.2.2.9.4 A Beginner's Guide to Uncertainty of Measurement. Measurement Good Practice Guide No. 11 (Issue 2), Bell, S., ISSN 1368-6550.

5.3 Evaluation

5.3.1 Completion of written study questions.

5.3.2 Oral presentation followed by technical question/answer session.

5.4 Study Questions

5.4.1 Define the following terms (use drawings/images if necessary):

5.4.1.1 ANAB

5.4.1.2 Mean

5.4.1.3 Median

5.4.1.4 Mode

5.4.1.5 Range

5.4.1.6 Accuracy

5.4.1.7 Precision

5.4.1.8 Gaussian Distribution

5.4.1.9 Confidence Interval

5.4.1.10 Coverage Factor

5.4.1.11 Measurement

5.4.1.12 Type A Evaluation

5.4.1.13 Type B Evaluation

5.4.2 Draw and explain what a Gaussian distribution is and how it relates to measurement uncertainty. Demonstrate two Gaussian distributions where one has high variability and one has low variability.

5.4.3 Obtain an uncertainty budget used in the Toxicology section. Define the elements and from where the information is obtained.

5.4.4 Within the Toxicology section, find a calibration standard that is traceable to NIST. Write a brief description of the traceability of that item.

5.4.5 Explain the day-of uncertainty of measurement. When is this used? How is it calculated?

5.4.6 The Combined Standard Uncertainty for alprazolam is 10.80197208, calculate the expanded uncertainty at k=2 and k=3. Calculate the uncertainty of the measurement of alprazolam at 0.082 mg/L at both k=2 and k=3. Demonstrate how this measurement of alprazolam would be reported with its associated uncertainty at the 95.45% level of confidence.

6 BLOOD ALCOHOL ANALYSIS

6.1 Objectives

6.1.1 Understand the theory and application of headspace gas chromatography (HS-GC).

6.1.2 Comprehend the function and the specifics of operation of HS-GC.

6.1.3 Prepare specimens for analysis by HS-GC.

6.1.4 Operate the HS-GC.

6.1.5 Calibrate the instrument and quantitate ethanol, methanol, acetone, and 2-propanol.

6.1.6 Interpret results by thoroughly examining and explaining the chromatograms.

6.1.7 Understand the use of internal standards.

6.1.8 Demonstrate proficiency by analyzing two runs (20 samples each) of blood alcohol cases.

6.1.9 Process results and record results of medical examiner, DUI/DUID, and TO casework.

6.1.10 FLS (Optional)/FS (Required): Understand the theory and practical aspects of gas chromatography (GC).

6.2 Methods of Instruction

6.2.1 Lectures and/or Self-Directed Study

6.2.1.1 Principles of HS-GC.

6.2.1.2 Operation of the HS-GC.

6.2.1.3 Specimen preparation (dilution, internal standard).

6.2.1.4 Calibration and QC (Pre-run).

6.2.1.5 Result interpretation.

6.2.2 Required Reading

6.2.2.1 Garriot, J.C., Medicolegal Aspects of Alcohol. 4th Ed. 2003, Lawyers and Judges Pub. Co., Inc.

6.2.2.1.1 Chapters 5, 6, 9.3 D – edition specific.

6.2.2.2 Levine, B. Principles of Forensic Toxicology. 2003.

6.2.2.2.1 Chapter 13 – Alcohol (or the appropriately titled chapter in a different edition).

6.2.2.3 Toxicology Procedures Manual.

6.2.2.4 Code of Virginia §§18.2-266 – 18.2-269.

6.2.2.5 Moffat, A.C., editor. Clarke's Analysis of Drugs and Poisons, 3rd Ed. London: The Pharmaceutical Press, 2004, pp. 53-67.

6.2.3 Demonstration

6.2.3.1 Blood alcohol analysis and operation of the HS-GC will be observed from beginning to end and notes will be taken by the Trainee.

6.2.3.2 Paperwork processing in medical examiner, DUI/DUID, and TO cases.

6.2.4 Laboratory Exercises

6.2.4.1 Analyze one batch of 20 OCME biological specimens for ethanol. At least 5 of the specimens will be positive for ethanol and at least one specimen will be negative. {Note: Where feasible, provide at least one alternative matrix to the trainee for analysis (e.g., liver or gastric content or other tissue is preferred however urine or vitreous are acceptable).

6.2.4.2 Analyze one batch of 20 DUI/DUID blood specimens for ethanol. At least 10 of the specimens will be positive for ethanol and at least one specimen will be negative.

6.3 Evaluation

6.3.1 Completion of written study questions.

6.3.2 Laboratory Competency Testing.

6.3.2.1 A series of at least 20 previously-analyzed OCME biological specimens will be presented to the Trainee for routine blood alcohol analysis. Trainee's results must fall within 0.004% w/v or 6%, whichever is greater, of the expected value.

6.3.2.2 A series of at least 20 previously-analyzed DUI/DUID blood specimens will be presented to the Trainee for routine blood alcohol analysis. Trainee's results must fall within 0.004% w/v or 6%, whichever is greater, of the expected value.

6.3.3 Oral presentation followed by technical question/answer session.

6.4 Study Questions

6.4.1 Explain the principle and operation of HS-GC including a schematic and explain the parts (including the detector).

6.4.2 Explain when calibration or recalibration of the HS-GC is necessary. How is recalibration accomplished?

6.4.3 Discuss the relationship between the concentration of alcohol in blood with that in urine, serum, liver, and vitreous humor.

6.4.4 Explain what causes the blood alcohol concentration in a specimen to either decrease or increase. What measures can be taken to prevent this?

6.4.5 What is the purpose of running a mixed volatile control during the pre-run?

6.4.6 Manually calculate BAC based on the response of ethanol in a sample, internal standard, and calibrators.

6.4.7 What are the properties of a good internal standard?

7 IMMUNOASSAY

7.1 Objectives

7.1.1 Understand and explain immunoassay.

7.1.2 Understand the theory of commonly used immunoassay testing methods.

7.1.3 Understand the theory and practice of Immunalysis ELISA system.

7.1.4 Perform Immunalysis ELISA screening.

7.1.5 Interpret results by thoroughly explaining the calculations and instrument printouts.

7.1.6 Understand the quality control aspects of ELISA screening.

7.2 Methods of Instruction

7.2.1 Lectures and/or Self-Directed Study

7.2.1.1 Principles of immunoassay.

7.2.1.2 Types of immunoassasys.

7.2.1.3 Components and operations of ELISA.

7.2.1.4 Specimen preparation.

7.2.1.5 Specimen analysis.

7.2.1.6 Result interpretation.

7.2.2 Required Reading

7.2.2.1 Levine, B. Principles of Forensic Toxicology. 2003. pp. 117-137 (edition specific).

7.2.2.2 Moffat, A.C., editor. Clarke's Analysis of Drugs and Poisons, 3rd Ed. London: The Pharmaceutical Press, 2004, pp. 301-312 (edition specific).

7.2.2.3 Operator's Guide for the current ELISA system.

7.2.2.4 Toxicology Procedures Manual.

7.2.3 Demonstration

7.2.3.1 ELISA analyses will be observed from beginning to end and notes will be taken by the Trainee.

7.2.4 Laboratory Exercises

7.2.4.1 Analyze one batch of 10 biological specimens by ELISA screening for at least 10 different classes of drugs. At least 5 specimens will be above the cutoff concentration and at least one specimen below the cutoff.

7.3 Evaluation

7.3.1 Completion of written study questions.

7.3.2 Laboratory Competency Testing.

7.3.2.1 A series of at least 10 previously analyzed blood specimens will be presented to the Trainee for a routine DUID panel according to the Toxicology Procedures Manual. Qualitative results obtained by the Trainee must agree with previous results.

7.3.3 Oral presentation followed by technical question/answer session.

7.4 Study Questions

7.4.1 Explain the advantages and disadvantages of screening for the presence of drugs.

7.4.2 Explain the following terms as they apply to ELISA

7.4.2.1 Antigen

7.4.2.2 Antibody

7.4.2.3 Monoclonal/polyclonal antibody

7.4.2.4 Microplate

7.4.2.5 Substrate

7.4.2.6 Horseradish peroxidase

7.4.2.7 Cross-reactivity

7.4.2.8 Cutoff limit of detection

7.4.2.9 True-positive

7.4.2.10 False-positive

7.4.2.11 Sensitivity

7.4.2.12 False-negative

7.4.2.13 Specificity

7.4.3 Distinguish between homogeneous (e.g., enzyme multiplied immunoassay technique (EMIT)) and heterogeneous immunoassays (ELISA).

7.4.4 Explain cross-reactivity, stating advantages and disadvantages. Include the significance of immunoassay specificity for a specific drug vs. the specificity for a drug class.

7.4.5 Name the chemical compounds that is the primary target of the antibody in each of the DFS ELISA assays, and the respective cut-off level (PC) concentration.

7.4.6 For at least 5 of the immunoassay kits DFS uses, provide examples of drugs commonly encountered in casework that cross-react well, and those that do not cross-react well.

7.4.7 Explain the relationship between absorbance and the concentration of the drug being determined.

7.4.8 Explain B/B_0. How is it calculated?

7.4.9 Explain the role of the negative control (NC), ½ cutoff (LPC), cutoff (PC), and high positive control (HPC).

7.4.10 Describe the components of the ELISA kits and explain the purpose each.

8 SPECTROPHOTOMETRY

8.1 Objectives

8.1.1 Understand and explain the principles of ultraviolet-visible (UV/VIS) spectrophotometric measurements.

8.1.2 Understand the practice of UV/VIS spectrophotometry and the specifics of operation of the spectrophotometers at DFS. Understand the practice of carboxyhemoglobin confirmation with palladium chloride.

8.1.3 Perform instrumental analysis of carboxyhemoglobin using a UV/VIS spectrophotometer.

8.1.4 Interpret the results by thoroughly examining and explaining the instrument printout.

8.1.5 Understand the quality control aspects of spectrophotometric testing.

8.2 Methods of Instruction

8.2.1 Lectures and/or Self-Directed Study

8.2.1.1 Principles of spectrophotometry.

8.2.1.2 Components and operation of the UV/VIS spectrophotometer.

8.2.1.3 Specimen preparation and analysis.

8.2.1.4 Results interpretation.

8.2.1.5 Palladium chloride diffusion confirmation test.

8.2.2 Required reading.

8.2.2.1 Levine, B. Principles of Forensic Toxicology, 2003, pp 79-88 (edition specific).

8.2.2.2 Moffatt, A.C., editor. Clarke's Analysis of Drugs and Poisons, 3rd Ed. London: The Pharmaceutical Press, 2004, pp 313-327 (edition specific).

8.2.2.3 Toxicology Procedures Manual.

8.2.3 Demonstration

8.2.3.1 The use of the UV/VIS spectrophotometer for the semi-quantitative analysis of carbon monoxide will be observed from beginning to end and notes will be taken by the trainee.

8.2.4 Laboratory Exercises

8.2.4.1 Analyze low, medium, and high controls for the presence of carbon monoxide (CO).

8.2.4.2 Screen one batch of 5 blood specimens for the presence of CO. At least 2 of the specimens will be positive and at least one specimen will be negative. In the absence of appropriate case samples, blind CO controls may be used for the laboratory exercises. Determine the approximate % saturation of each specimen.

8.2.4.3 Confirm the presence of CO using the palladium chloride diffusion test.

8.3 Evaluation

8.3.1.1 Completion of written study questions.

8.3.1.2 Laboratory Competency Testing.

8.3.1.2.1 A series of at least 5 previously analyzed blood specimens will be presented to the Trainee for the CO analysis. The results obtained by the Trainee must agree within 20% of the expected value. If previously analyzed casework specimens are not available, blind controls may be substituted at the TC's discretion.

8.3.1.3 Oral presentation followed by a technical question/answer session.

8.4 Study Questions

8.4.1 What are the wavelength ranges for visible and ultraviolet electromagnetic radiation?

8.4.2 Explain what effects a change in solvent might have on the spectrum of a solute.

8.4.3 Discuss why a change in the pH of a solution can be important when using UV/VIS for analysis.

8.4.4 List and discuss some common sources of error in spectrophotometric measurements.

8.4.5 Define the following terms:

8.4.5.1 Wavelength

8.4.5.2 Absorbance

8.4.5.3 Transmittance

8.4.5.4 Excitation

8.4.5.5 Emission

8.4.5.6 Bandwidth

8.4.5.7 Beer-Lambert Law

8.4.6 In the semi-quantitative carboxyhemoglobin analysis, explain deoxyhemoglobin, oxyhemoglobin, methemoglobin, and carboxyhemoglobin.

8.4.7 How are the results reported for CO on a certificate of analysis?

8.4.8 Explain the principle of the palladium chloride confirmation.

8.4.9 When should the analyst use lead acetate in the palladium chloride confirmation?

9 EXTRACTIONS

9.1 Objectives

9.1.1 Understand the theoretical and practical aspects of extractions.

9.1.2 Extract representative compounds (basic, acidic, and neutral) from various matrices.

9.1.3 Understand the use of internal standards and quality controls as applied to qualitative and quantitative analyses.

9.2 Methods of Instruction

9.2.1 Lectures and/or Self-Directed Study

9.2.1.1 Principles of extraction.

9.2.1.2 Henderson-Hasselbach equation, acid-base equilibrium.

9.2.1.3 Buffers and ionization.

9.2.1.4 Extraction.

9.2.1.5 Liquid-liquid extraction (LLE).

9.2.1.6 Solid phase extraction (SPE).

9.2.1.7 Automated liquid handling system (ALH).

9.2.1.8 Specimen preparation (dilution, internal standard, derivatization).

9.2.2 Required Reading

9.2.2.1 Toxicology Procedures Manual.

9.2.2.2 Moffatt, A.C., editor. Clarke's Analysis of Drugs and Poisons. 3rd Ed. London: The Pharmaceutical Press. 2004 pp 80-108, 379-391, 425-499 (edition specific).

9.2.2.3 Levine, B. Principles of Forensic Toxicology. 2003. Pp 67-78, 89-116, 139-153 (edition specific).

9.2.2.4 Juhascik, M. and Jenkins, A. Comparison of Liquid/Liquid and Solid Phase Extraction for Alkaline Drugs. Journal of Chromatographic Science. Vol. 47, August 2009, pp 553-557.

9.2.2.5 Hamilton MicroLab STAR User's Guide.

9.2.2.6 Additional References:

9.2.2.6.1 Hamilton MicroLab STAR Guide (powerpoint available on intranet).

9.2.2.6.2 Mills, T. and Robinson, JC. Instrumental Data for Drug Analysis. 2nd Ed. Vols. 1-7, New York, Elsevier, 1987.

9.2.3 Demonstration

9.2.3.1 The following extraction techniques will be observed from beginning to end and notes will be taken by the Trainee: LLE, SPE, ALH, qualitative extraction, quantitative extraction, derivatization.

9.2.4 Laboratory Exercises

9.2.4.1 Perform a LLE or SPE (at the discretion of the TC or designee) of base screen drug mixes and 5 previously analyzed biological specimens for analysis by GC/NPD and GC/MS.

9.2.4.2 Perform a LLE or SPE of acidic/neutral drug mixes and 3 previously analyzed biological specimens for quantitative analysis by GC/MS.

9.2.4.3 Perform an SPE quantitation of calibrators and controls.

9.2.4.4 Spike and extract a quetiapine, hydroxyzine, or barbiturate calibration curve and the required QC samples.

9.2.4.5 Perform an OpiCoc extraction using an ALH.

9.2.4.6 Spike and extract a calibration curve and quality control samples for any LCMSMS method (may be combined with other required laboratory exercises at the discretion of the TC or designee).

9.2.4.7 Note – FLS Trainee data work-up will be performed by an approved FS/Toxicologist. FS/Toxicologist Trainees will perform their own data work-up.

9.3 Evaluation

9.3.1 Completion of written study questions.

9.3.2 Laboratory Competency Testing.

9.3.2.1 LLE or SPE – a series of 5 previously analyzed biological specimens will be presented to the Trainee for extraction (base or acidic/neutral screen, as specified by TC or designee). Extracts will be run on the GC-NPD and the GC/MS. Qualitative findings must agree with previously established results.

9.3.2.2 Perform a LCMSMS quantitation of 5-10 samples. The quantitation performed shall be specified by the TC or designee. Quantitative results must agree within ±20% of the analyte's established target value (reported or reanalysis) or of the target analyte's uncertainty of measurement for the current year.

9.3.3 Oral presentation followed by technical question/answer session.

9.4 Study Questions

9.4.1 Describe LLE and SPE stating the advantages and disadvantages of each type.

9.4.2 List and describe chemical forces which drive the movement of solute between aqueous and organic phases in LLE.

9.4.3 Explain the effects of pH on extractions.

9.4.4 List at least three different types of SPE sorbents and how they interact with the analytes being extracted.

9.4.5 List and explain the typical steps in an SPE procedure.

9.4.6 Describe the operation of the automated liquid handling system.

9.4.7 Define the following terms

9.4.7.1 Matrix

9.4.7.2 Functional group

9.4.7.3 Polarity

9.4.7.4 Solvents

9.4.7.5 pH

9.4.7.6 pKa

9.4.7.7 Henderson-Hasselbach equation

9.4.7.8 Basic

9.4.7.9 Acidic

9.4.7.10 Neutral

9.4.7.11 Amphoteric molecules

9.4.7.12 Conjugate acid

9.4.7.13 Conjugate base

9.4.7.14 Internal standard

9.4.7.15 External standard

9.4.8 Describe when it is appropriate to open the 2nd vial in a DUID case and the subsequent actions taken.

9.4.9 What action is performed if a sample is consumed in analysis? What are the volume thresholds for analysis for screening v. confirmation (e.g., for an extraction requiring 1 mL of sample, what do you do if there is only 0.9 mL of samples? Or only 0.4 mL of sample?)?

9.4.10 What are potential problems that may arise when building your batch from the worklist (or similarly named list of needed analyses)? What subsequent actions should be taken?

10 DRUG SCREENING, QUANTITATION, AND CONFIRMATION

10.1 Objectives

10.1.1 Understand the requirements for screening and confirmation in forensic toxicology at DFS.

10.1.2 Perform qualitative and quantitative drug identification of biological specimen extractions using full scan or selected ion monitoring (SIM) gas chromatography mass spectrometry (GC/MS), gas chromatography nitrogen phosphorus detector (GC/NPD), and liquid chromatography tandem mass spectrometry (LCMSMS).

10.1.3 Become proficient in the use of GC, GC/MS, and LCMSMS for qualitative and quantitative analyses in Toxicology.

10.1.4 Examine and interpret gas chromatographic data.

10.1.5 Generate and evaluate chromatographic and mass spectral information to identify, confirm, and quantitate the drugs being analyzed.

10.1.6 Examine and interpret full scan GC/MS results by explaining and comparing the mass spectra to libraries and databases.

10.1.7 Generate accurate and precise quantitative results.

10.1.8 Perform derivatized drug quantitations. Understand the role of derivatization.

10.1.9 Construct and apply calibration curves using GC, GC/MS, and LCMSMS software.

10.1.10 Understand and explain the criteria for acceptance of qualitative and quantitative data.

10.1.11 Demonstrate a working knowledge of reporting qualitative and quantitative results in the manner used in the Toxicology section.

10.2 Methods of Instruction

10.2.1 Lectures and/or Self-Directed Study

10.2.1.1 Requirements for screening and confirmation.

10.2.1.2 Acquiring and evaluating mass spectra.

10.2.1.3 Use of libraries and databases.

10.2.1.4 Preparation of a calibration curve.

10.2.1.5 Operation of GC/MS in full scan mode.

10.2.1.6 Selected Ion Mode (SIM) of operation.

10.2.1.7 Spectral interpretation.

10.2.1.8 Tandem Mass Spectrometry modes of operation including MS1/MS2 Scan, SRM, MRM, Dynamic MRM, Product Ion Scan, Precursor Ion Scan

10.2.1.9 Use of instrument software (e.g., OpenLab, Chemstation, and MassHunter) to generate calibration curves for quantitative data and qualitative data.

10.2.1.10 Derivatized extractions.

10.2.1.11 Acquisition of data.

10.2.1.12 Qualitative results.

10.2.1.13 Quantitative analysis including overview, data interpretation, batch report generation.

10.2.2 Required reading

10.2.2.1 Toxicology Procedures Manual

10.2.2.2 Moffatt, A.C. editor. Clarke's Analysis of Drugs and Poisons. 3rd Ed. London: The Pharmaceutical Press. 2004, pp 80-108, 379-391, 425-499 (edition specific).

10.2.2.3 Willard, H.H., Merritt, L.L. Jr., Dean, J., Settle, F.A., Instrumental Methods of Analysis, 7th Ed., 1988, Wadsworth Pub. Co. pp 540-578 (edition specific).

10.2.2.4 Levine, B. Principles of Forensic Toxicology. 2003. Pp 67-78, 89-116, 139-153 (edition specific).

10.2.2.5 McLafferty, F.W., Interpretation of Mass Spectra, 3rd Ed. Chapter 1.

10.2.2.6 Additional References

10.2.2.6.1 Mills, T. and Robinson, J.C. Instrumental Data for Drug Analysis. 2nd Ed. Volumes 1-7, New York: Elsevier, 1987.

10.2.2.6.2 Knapp, D.R., Handbook of Analytical Derivatization Reactions. John Wiley, New York, 1979.

10.2.3 Demonstration

10.2.3.1 The following techniques will be observed from beginning to end and notes will be taken by the Trainee: qualitative drug screening by GC/MS, qualitative and quantitative drug screening and confirmation using GC/MS SIM, quantitative drug confirmation by GC/NPD, qualitative and quantitative drug screening/confirmation by LCMSMS, and drug identification by GC/NPD, GC/MS, and LCMSMS.

10.2.4 Laboratory Exercises

10.2.4.1 Determine the retention time and relative retention time (using the GC/NPD and methapyrilene as the internal standard) of basic drug mixes (may use data collected from previous exercises or from previously analyzed casework data).

10.2.4.2 Use GC/MS and mass spectral libraries to identify drugs and metabolites in 20 drug screens (20 total of base and acid/neutral). Review all cases with a qualified forensic scientist/toxicologist to ensure all drugs and metabolites were correctly identified. Additional screens may be assigned by T.C. or designee as necessary.

10.2.4.3 Analyze the data for a basic drug mix quantitation on the GC/NPD (may use data collected from previous exercises or from previously analyzed casework data). Use the GC quantitative software to create a calibration curve and determine the quality control values. Determine LOQ, ULOQ, and pass/fail of the threshold control for each drug in the mix.

10.2.4.4 Analyze the data for either a fentanyl derivative or NPS analysis (may use data collected from previous exercises or from previously analyzed casework data).

10.2.4.5 Analyze LCMSMS data for both qualitative and quantitative analyses (may use data collected from previous exercises or from previously analyzed casework data). Determine LOQ, ULOQ, and pass/fail of the threshold control for each drug in the mix.

10.3 Evaluation

10.3.1 Completion of written study questions.

10.3.2 Laboratory Competency Testing

10.3.2.1 Perform two GC quantitations that are available in the current Toxicology Procedures Manual. One quantitation should be a GC/NPD quantitation and one should be a GC/MS SIM quantitation (dependent upon laboratory availability of instrumentation and methodology). The quantitations shall be chosen by the TC of designee. The quantitations should each

include 5 samples for analysis and the results must agree within ±20% of the established target value (reported or reanalysis) or of the target analyte's uncertainty of measurement for the current year.

10.3.2.2 Perform one LCMSMS quantitation of 5 samples. The quantitation is chosen by the TC or designee. Results must agree within ±20% of the analyte's target value (reported or reanalysis) or of the target analyte's uncertainty of measurement for the current year.

10.3.3 Oral presentation followed by technical question/answer session.

10.4 Study Questions

10.4.1 What are the advantages of using relative retention time for drug identification rather than retention time?

10.4.2 Describe the use of drug reference materials in the identification process.

10.4.3 Describe the difference between full mass scans and selective ion monitoring.

10.4.4 What is an extracted ion profile? How would you use it in drug identification?

10.4.5 How does the probability-based-matching library search work?

10.4.6 Explain the following:

10.4.6.1 Threshold control (as applied to both quantitative and qualitative analyses)

10.4.6.2 LOD

10.4.6.3 LOQ

10.4.6.4 ULOQ

10.4.7 Explain the SOP criteria concerning rejecting calibrator concentrations in a calibration curve.

10.4.8 Define and explain the following:

10.4.8.1 Blank and negative control

10.4.8.2 Internal standard

10.4.8.3 External standard

10.4.8.4 Positive control

10.4.8.5 Calibrator

10.4.9 Describe silylation and methylation.

10.4.10 Describe and/or draw the derivative formed using the Toxicology Procedures Manual for a barbiturate (e.g., butalbital) and an analyte derivatized utilizing BSTFA + 1% TMCS.

10.4.11 How would the following be reported:

10.4.11.1 Drug concentration is greater than the ULOQ.

10.4.11.2 Drug concentration is below LOQ but above the threshold control and has acceptable ion ratios.

10.4.11.3 Drug concentration is below LOQ and the threshold control and the drug has acceptable ion ratios.

10.4.11.4 Drug concentration is below LOQ but above the threshold control and one ion ratio is unacceptable.

10.4.11.5 The LOQ changed (i.e. the lowest calibrator was removed). Drug concentration is below the new LOQ but above the threshold control and has acceptable ion ratios.

11 GAS AND LIQUID CHROMATOGRAPHY

11.1 Objectives

11.1.1 Understand the theory, practical aspects, and components of gas chromatography (GC) and high performance liquid chromatography (HPLC or LC) instrumentation.

11.1.2 Perform routine maintenance on the GC and LC systems.

11.2 Methods of Instruction

11.2.1 Lectures and/or Self-Directed Study

11.2.1.1 Principles of chromatography (GC and LC).

11.2.1.2 Components and operation of GC and LC.

11.2.1.3 Parameters affecting the separation process and resolution of peaks in GC.

11.2.1.4 Types of injectors and injection techniques (GC).

11.2.1.5 Types of GC columns.

11.2.1.6 GC optimization.

11.2.1.7 Parameters affecting the separation process and resolution of peaks in LC.

11.2.1.8 Types of LC columns and mobile phases.

11.2.1.9 Optimization of LC.

11.2.2 Required Reading

11.2.2.1 Toxicology Procedures Manual.

11.2.2.2 Moffatt, A.C., editor. Clarke's Analysis of Drugs and Poisons. 3rd Ed. London: The Pharmaceutical Press. 2004 pp 80-108, 379-391, 425-499 (edition specific).

11.2.2.3 Willard, H.H. et al. Instrumental Methods of Analysis. 7th Ed. 1988. Wadsworth Pub. Co. pp 540-578 (edition specific).

11.2.2.4 Levine, B. Principles of Forensic Toxicology. 2003. Pp 67-78, 89-116, 139-153 (edition specific).

11.2.2.5 McLafferty, F.W. Interpretation of Mass Spectra. 3rd Ed., Chapter 1.

11.2.2.6 Additional Resources

11.2.2.6.1 Mills, T and Robinson, JC. Instrumental Data for Drug Analysis. 2nd Ed. Vol. 1-7. New York: Elsevier. 1987.

11.2.2.6.2 Knapp, D.R. Handbook of Analytical Derivatization Reactions. John Wiley. New York. 1979.

11.2.3 Demonstration

11.2.3.1 The following will be observed from beginning to end and notes will be taken by the Trainee: routine maintenance, non-routine maintenance, and troubleshooting of GC/MS, GC/NPD and/or FID, and LCMSMS.

11.2.4 Laboratory Exercises

11.2.4.1 Perform daily routine maintenance of the GC/MS to include (but not limited to) changing or adjusting the liner, septum, seals, gap column, transfer lines, and gold seal.

11.2.4.2 Perform maintenance of the LCMSMS to include (but not limited to) replacing the column, changing the guard column, installing a new capillary (if needed), removing air bubbles from the LC lines, and preparing new solvent.

11.2.4.3 Use Lab Advisor to perform both leak and pressure tests of at least one channel of the LC system.

11.3 Evaluation

11.3.1 Completion of written study questions.

11.3.2 Laboratory Competency Testing

11.3.2.1 Perform two GC quantitations that are available in the current Toxicology Procedures Manual. One quantitation should be a GC-NPD quantitation and one should be a GC/MS SIM quantitation (if either are used in the Trainee's home laboratory). The quantitations shall be chosen by the TC or designee. The quantitations should include 5 samples for each analysis. Quantitative results must agree within ±20% of the analyte's target value (reported or reanalysis) OR of the target analyte's uncertainty of measurement for the current year.

11.3.2.2 Perform an LCMSMS quantitation of 5 samples. The quantitation shall be chosen by the TC or designee. Quantitative results must agree within ±20% of the analyte's target value (reported or reanalysis) OR of the target analyte's uncertainty of measurement for the current year.

11.3.3 Oral presentation followed by a technical question/answer session.

11.4 Study Questions

11.4.1 Provide a brief overview of gas chromatography.

11.4.2 Draw a schematic diagram of a gas chromatograph and describe the function of each component. Provide detailed explanations of the injection system and column(s) commonly used in the Toxicology section.

11.4.3 Describe the different types of GC stationary phases used in the Toxicology section.

11.4.4 What are the possible causes and remedies for the following GC problems:

11.4.4.1 No peaks

11.4.4.2 Tailing peaks

11.4.4.3 Leading peaks

11.4.4.4 Split peaks

11.4.4.5 Baseline drift

11.4.5 What is column bleed?

11.4.6 When and why are columns conditioned? Describe the process.

11.4.7 Define the following terms as related to GC:

11.4.7.1 Carrier gas.

11.4.7.2 Mobile phase.

11.4.7.3 Stationary phase.

11.4.7.4 Partition coefficient.

11.4.7.5 Retention time.

11.4.7.6 Relative retention time.

11.4.7.7 Resolution.

11.4.7.8 Theoretical plates.

11.4.7.9 Height equivalent theoretical plate.

11.4.7.10 Column efficiency.

11.4.7.11 Make-up gas.

11.4.7.12 Van Deemter plot.

11.4.7.13 Phase ratio.

11.4.7.14 Selectivity.

11.4.7.15 Flow rate.

11.4.7.16 Signal to noise ratio.

11.4.8 Draw a schematic diagram of an HPLC system and describe the function of each component.

11.4.9 Describe factors that can affect peak resolution (e.g., particle size, column choice, mobile phase) in LC analysis. Using these factors, describe the steps you would take to resolve two co-eluting peaks.

11.4.10 Define the following as related to LC:

11.4.10.1 Mobile phase.

11.4.10.2 Capacity factor.

11.4.10.3 Isocratic elution.

11.4.10.4 Gradient elution.

11.4.10.5 Normal phase HPLC

11.4.10.6 Reverse phase HPLC

11.4.10.7 Ion chromatography

11.4.10.8 Resolution

11.4.11 Discuss the use of various buffers and acid additives within the mobile phase with respect to LCMSMS.

12 MASS SPECTROMETRY, MASS SELECTIVE DETECTORS, AND ALTERNATE DETECTORS

12.1 Objectives

12.1.1 Understand the theory, practical aspects, and components of mass spectrometry (MS) and tandem mass spectrometry (MSMS).

12.1.2 Understand the theory and maintenance of alternate detectors (e.g., nitrogen phosphorus detectors (NPD) and flame ionization detectors (FID)).

12.1.3 Perform routine maintenance on the NPD and/or FID.

12.1.4 Perform routine maintenance and tuning of the GC/MS and LCMSMS.

12.1.5 Understand and explain the operation of the LCMSMS user interface.

12.1.6 Understand and explain ion formation (electron impact and electrospray ionization).

12.2 Methods of Instruction

12.2.1 Lectures and/or Self-Directed Study

12.2.1.1 Types of detectors.

12.2.1.2 Principles of mass spectrometry: ionization, source, detection.

12.2.1.2.1 MS components (sample inlets, ion sources, mass filters, detectors, vacuum systems).

12.2.1.3 Principles of tandem mass spectrometry.

12.2.1.3.1 Ion sources: ESI, APCI.

12.2.1.3.2 Ion focusing optics/lenses.

12.2.1.3.3 MSMS: quadrupoles, collision cells

12.2.1.4 MS components (sample inlets, ion source, focusing components, quadrupoles, collision cell, HED).

12.2.1.5 Optimization of targets: purpose, parameters.

12.2.2 Required Reading

12.2.2.1 Toxicology Procedures Manual

12.2.2.2 Moffatt, A.C., editor. Clarke's Analysis of Drugs and Poisons. 3rd Ed. London: The Pharmaceutical Press. 2004. pp 80-108, 379-391, 425-499 (edition specific).

12.2.2.3 Willard, H.H. et al. Instrumental Methods of Analysis. 7th Ed. 1988. Wadsworth Pub. Co. pp 540-578 (edition specific).

12.2.2.4 Levine, B. Principles of Forensic Toxicology. 2003. pp 67-78-89-116, 139-153 (edition specific).

12.2.2.5 McLafferty, F.W. Interpretation of Mass Spectra. 3rd Ed. Chapter 1.

12.2.2.6 Additional Resources

12.2.2.6.1 Mills, T. and Robinson, J.C. Instrumental Data for Drug Analysis. 2nd Ed. Vol. 1-7. New York: Elsevier. 1987.

12.2.2.6.2 Knapp, D.R. Handbook of Analytical Derivatization Reactions. John Wiley. New York. 1979.

12.2.3 Demonstration

12.2.3.1 The following will be observed from beginning to end and notes will be taken by the Trainee: routine maintenance, non-routine maintenance, and troubleshooting of MSD, MSMS, and NPD/FID.

12.2.4 Laboratory Exercises

12.2.4.1 Perform daily routine maintenance of the GC/MS to include (but not limited to) changing or adjusting the autotune.

12.2.4.2 Perform daily, weekly, and monthly maintenance procedures. This is to include draining the demister (if applicable), evaluation of a Checktune, cleaning of the ion source, manually adjusting one or more tune parameters, and performing an Autotune.

12.2.4.3 Review the results of Autotune and Checktune reports. Evaluate the reports.

12.2.4.4 Change the bead on an NPD. (*Due to the expensive nature of replacing the bead, the training coordinator may instead have the trainee describe in detail how to do so, including conditioning of the new bead, without requiring the trainee to perform this maintenance to complete the training section. However, the trainee should observe/participate in replacing a bead at the earliest opportunity.*)

12.2.4.5 Use Lab Advisor to perform both leak and pressure tests of at least one channel of the LC system.

12.3 Evaluation

12.3.1 Completion of written study questions.

12.3.2 Laboratory Competency Testing

12.3.2.1 Clean a GC/MS source, including correct reassembly and installation of the source. Once the MS has been pumped down, the autotune must provide acceptable results for the competency test to be considered successful.

12.3.2.2 Troubleshoot one or more leaks on an LCMSMS system. Acceptable performance of an injected test mix (either extracted or non-extracted) must be achieved for successful completion of the competency test.

12.3.3 Oral presentation followed by technical question/answer session.

12.4 Study Questions

12.4.1 List all the types of detectors used in the Toxicology section of DFS. Briefly the functions and benefits of the NPD and FID.

12.4.2 Draw a schematic diagram for each of the detectors commonly used in the Toxicology section. Label and describe the functions of each component.

12.4.3 Provide a brief overview of mass spectrometry.

12.4.4 What is “make-up” gas? How and why is it used?

12.4.5 Explain the following statement: *response is proportional to the number of carbon atoms in the sample*. What type(s) of detector is this statement applicable to?

12.4.6 Describe how a quadrupole mass filter operates.

12.4.7 Diagram and explain the functions of the components of a common EI source.

12.4.7.1 Are the ions formed positive or negative?

12.4.7.2 Do they have an even or odd number of electrons?

12.4.7.3 What is the ionization efficiency of this technique?

12.4.8 What vacuum conditions are necessary in the ionization source and the analyzing regions of an MS and why?

12.4.8.1 Describe how a rough pump works?

12.4.8.2 Describe how a turbomolecular pump works?

12.4.9 Describe the importance of autotuning and explain the Autotune report.

12.4.10 Explain the following MS terms:

12.4.10.1 Mass to charge ratio.

12.4.10.2 Molecular ion.

12.4.10.3 Parent ion.

12.4.10.4 Base peak.

12.4.10.5 Total ion chromatogram.

12.4.10.6 Extracted ion chromatogram.

12.4.10.7 SIM.

12.4.10.8 Mass resolution.

12.4.10.9 Relative abundance.

12.4.10.10 Scan rate.

12.4.10.11 Spectral tilting.

12.4.11 Define the term transition in LCMSMS analysis and relate it to GC/MS SIM analysis. Explain how ion transitions and ion fragmentation provides appropriate specificity and quantitative information for the two types of analyses.

12.4.12 Diagram an electrospray ionization source (can use previously drawn schematic for LCMSMS).

12.4.12.1 Explain the ionization process.

12.4.12.2 What is coulombic explosion?

12.4.12.3 What is the purpose of the drying gas?

12.4.13 Discuss ion suppression and how can it affect LCMSMS analysis?

12.4.14 Discuss the following with examples from the Agilent 6400 Series LCMSMS. When does the operator perform each of these activities?

12.4.14.1 Checktune.

12.4.14.2 Autotune.

12.4.15 Discuss the advantage of the following comparisons:

12.4.15.1 LCMS v. LCMSMS.

12.4.15.2 GC/EI-MS v. LCMSMS.

13 ALCOHOLIC BEVERAGE ANALYSIS

13.1 Objectives

13.1.1 Demonstrate proficiency in the analysis of beverages for alcohol content.

13.2 Methods of Instruction

13.2.1 Lectures and/or Self-Directed Study

13.2.1.1 Chemical formulations and compositions of alcoholic beverages.

13.2.2 Required Readings

13.2.2.1 Lembeck, H. Grossman's Guide to Wine, Beers, and Spirits. New York: Charles Scribner's Sons. 1983.

13.2.2.2 Lichine, A. Alexis Lichne's Encyclopedia of Wines and Spirits. New York: Alfred Knopf, Inc. 1983.

13.2.2.3 Caplan, Y.H. and Goldberger, B.A. Garriott's Medicolegal Aspects of Alcohol. 6th Ed. 2015. Lawyers and Judges Publishing Co, Inc. Chapter 1.

13.2.2.4 Code of Virginia Title 4.1 Alcoholic Beverage Control Act, 4.1-100.

13.2.3 Demonstration

13.2.3.1 Alcoholic beverage analyses will be observed from beginning to end and notes will be taken by the Trainee.

13.2.4 Laboratory Exercises

13.2.4.1 Perform ethanol content analyses on 20 different alcoholic beverages.

13.3 Evaluation

13.3.1 Completion of written study questions.

13.3.2 Laboratory Competency Testing.

13.3.2.1 A series of at least 20 different alcoholic beverages will be presented to the Trainee for a routine alcohol content determination. Quantitative results must agree within ±10 of the target value (reported or reanalysis).

13.3.3 Oral presentation followed by technical question/answer session.

13.4 Study Questions

13.4.1 Explain when calibration or recalibration of the HS-GC is necessary. How is recalibration accomplished?

13.4.2 What is NIST? Why is it important?

13.4.3 Describe the ranges of alcohol content for the following alcoholic beverages: table wines, fortified wines, light beer, premium beer, malt liquors, special stouts, and distilled spirits.

13.4.4 Define the following:

13.4.4.1 Congeners.

13.4.4.2 Proof.

13.4.4.3 Fermentation.

13.4.4.4 Mash.

13.4.4.5 Distillation.

13.4.5 What is the purpose of the Virginia Department of Alcoholic Beverage Control?

13.4.6 What are the common investigations in which ABC evidence is submitted?

13.4.7 Describe the accessioning process for ABC evidence.

13.4.8 Describe any differences between the Blood Alcohol method and the ABC Alcohol method.

14 DATA REVIEW AND CASE EXAMINATION

14.1 Objectives

14.1.1 To learn the process and documentation involved in data review, case examination, and technical review.

14.1.2 To learn the process for creating and releasing cases using LIMS.

14.2 Methods of Instruction

14.2.1 Data review and case examination training primarily learned by observing multiple certified examiners and performing training examinations that are critiqued by certified examiners.

14.2.2 Trainee will observe and take notes of data (batch) review process with at least two experienced data reviewers.

14.2.3 Trainee will observe and take notes of case examination and review process with at least two experienced examiners.

14.2.4 Trainee will observe and take notes on LIMS Certificate of Analysis (CoA) creation, technical review, and release with at least two experienced examiners.

14.2.5 Trainee will review the Toxicology Procedures Manual (Ch. 2) and the Quality Manual (Ch. 16, 17).

14.3 Laboratory Exercises

14.3.1 Perform practice data review (do NOT mark up the data) on alcohol, immunoassay, drug screens, GC quantitation, GC/MS quantitation, and LCMSMS quantitation batches with at least two different examiners. These task may be completed throughout the training program during the completion of specific modules.

14.3.2 Perform practice case examinations (do NOT mark up case data) on 10 non-implied consent cases with at least two different examiners (20 total cases minimum). Cases should be a variety to include (if available) homicide, drug overdose, drug-facilitated crime, and decomposition cases. Medical examiner ethanol-only cases are not included.

14.3.3 Perform 10 DUI/DUID case examinations with at least two different examiners (20 cases total minimum). No more than 5 ethanol only cases and 2 negative cases can be included.

14.3.4 The Trainee should document the review of at least five case files using the appropriate technical review form (TRF) or process. TRFs shall be marked as Training and retained in the Training File. Case files should be generated by multiple examiners, if possible. Any findings from the Trainee will be discussed with the TC before discussions with the case examiner. The case files shall be technically reviewed by a certified examiner prior to release.

14.4 Evaluation

14.4.1 Non-implied consent case: The TC will select 20 cases that have not had final case examination performed. The Trainee will perform final case examination using a Toxicology Summary Worksheet (or other named document/process used for report generating purposes) marked as a “Training” case and submit cases for evaluation.

14.4.2 DUI/DUID cases: The TC will select 20 DUI/DUID cases that have not had final case examination performed. Trainee will perform final case examination using a Toxicology Summary Worksheet (or other named document/process used for report generating purposes) marked as a “Training” case and submit cases for evaluation.

14.5 Study Questions

14.5.1 What do the analyst date and time notations on the batch chain-of-custody indicate?

14.5.2 How many controls must be acceptable in a drug quantitation batch?

14.5.3 How is carryover monitored in a drug quantitation? What is the response when carryover is suspected?

14.5.4 Describe occasions when a drug may be reported as “present”? “Present greater than,…”?

14.5.5 An OCME case history states “suspected overdose” and they requested an Abused panel. The blood morphine quantitation is 0.10 mg/L and 6AM is below the threshold level. As the case examiner, is this case complete? What other questions/analyses might you consider?

14.5.6 A methadone quantitation was performed on femoral blood and heart blood. Would you expect the methadone concentrations to be different and if so why?

14.5.7 A sexual assault case has immunoassay blood benzodiazepine negative, urine benzodiazepine pending and benzodiazepine quantitation none detected. As final case examiner is benzodiazepine testing complete?

14.5.8 Hospital blood and urine are submitted in a DUI manslaughter case. The blood alcohol was 0.10% on two separate aliquots. Would you order a urine alcohol and why or why not?

15 COURTROOM TESTIMONY

15.1 Objectives

15.1.1 To familiarize the trainee with the functions of a criminal courtroom proceeding.

15.1.2 To have the Trainee prepare a current curriculum vitae (or resume) and properly answer voir dire questioning.

15.1.3 To familiarize the Trainee with proper methods of presenting expert testimony.

15.2 Methods of Instruction

15.2.1 Required Reading

15.2.1.1 Kuzmack, N.T., JD, MA. *Legal Aspects of Forensic Science* in Saferstein's *Forensic Science Handbook*. Englewood Cliffs, NJ: Prentice Hall, 1982 pp 1-27.

15.2.1.2 Babitsky S. and J. Mangraviti. *How to Excel during Cross-Examination. Techniques for Experts that Work*. Falmouth, MA: SEAK, 1997.

15.2.1.3 Kogan, J. *Being a Good Expert Witness in a Criminal Case*. J For Sci 23(1): 190-200, 1978.

15.2.1.4 Kates, James H. and Henry K. Guttenplan, Ph.D. *Ethical Considerations in Forensic Science Services* J For Sci 28(4): 972-976, 1983.

15.2.1.5 Keefe, J.F. *Forensic Sciences: Criminal Justice System Viewed by the Defense.* 12(2):59, 1980.

15.2.1.6 Lucas, Douglas M., M.Sc. *The Ethical Responsibilities of the Forensic Scientist: Exploring the Limits* J For Sci 34(3):719-729, 1989.

15.2.1.7 Saks, Michael J., Ph.D., M.S.L. *Prevalence and Impact of Ethical Problems in Forensic Science* J For Sci 34(3): 772-793, 1989.

15.2.1.8 Schroeder, Oliver C., J.D. *Ethical and Moral Dilemmas Confronting Forensic Scientists* J For Sci 29(4): 966-986, 1984.

15.2.1.9 Wu, A., Hill, D., Crouch, D., Hodnett, N., and H. McCurdy. *Minimal Standards for the Performance and Interpretation of Toxicology Tests in Legal Proceedings.* J For Sci 44(3): 516-522, 1999.

15.2.1.10 Saady, J. *Ethics for Toxicologists: An Examination of Conscience* J Anal Tox 25:390 - 392, 2001.

15.2.2 Demonstration

15.2.2.1 The trainee will observe expert courtroom testimonies. Discuss testimony with each examiner. Document each observed testimony with the name of the examiner, date, court, and notes reflecting the testimony and discussion.

15.2.3 Practical Exercises

15.2.3.1 Complete curriculum vitae or resume.

15.2.3.2 Mini mock trials (may be completed throughout other modules).

15.3 Evaluation

15.3.1 Completion of written study questions.

15.3.2 Courtroom exercises.

15.3.3 The trainee must be capable of answering questions on this module such as would be expected in a courtroom scenario. This may be completed through mini mock trials in other modules.

15.4 Study Questions

15.4.1 Discuss the role of the following during a trial:

15.4.1.1 Expert witness.

15.4.1.2 Judge.

15.4.1.3 Prosecutor.

15.4.1.4 Defendant.

15.4.1.5 Defense counsel.

15.4.1.6 Jury.

15.4.2 Define the following:

15.4.2.1 Voir dire.

15.4.2.2 Direct examination.

15.4.2.3 Cross examination.

15.4.2.4 Redirect.

15.4.2.5 Rebuttal witness.

15.4.2.6 Chain of custody.

15.4.3 Define the word “ethics”.

15.4.3.1 Why is it important in forensic science?

15.4.3.2 Investigate and describe the Code of Ethics for DFS, AAFS, ANAB, SOFT, and ABFT.

15.4.3.3 Give some examples of ethical violations and sanctions imposed by forensic organizations.

15.4.4 Verbally answer the following questions to the TC or designee:

15.4.4.1 What is your name?

15.4.4.2 What is your occupation?

15.4.4.3 Who are you employed by?

15.4.4.4 How long have you been so employed?

15.4.4.5 What are your duties in this position?

15.4.4.6 What education and training do you possess that qualifies you to perform your duties?

15.4.4.7 What specific courses have you taken that are directly related to toxicology analysis?

15.4.4.8 How are these courses related? For example, what did you learn in your general chemistry course that aids you in the analysis of forensic toxicology samples?

15.4.4.9 What is the definition of an expert witness?

15.4.4.10 Is the university/college you graduated from accredited, and if so, by whom?

15.4.4.11 Who conducted your training?

15.4.4.12 What are his/her/their qualifications?

15.4.4.13 What literature do you read relating to your job?

15.4.4.14 How many analyses have you done on forensic cases?

15.4.4.15 Do you belong to a professional organization related to your occupation?

15.4.4.16 Have you written any articles or published materials dealing with your work?

16 ALCOHOL PHARMACOLOGY, IMPAIRMENT, AND COURTROOM TESTIMONY

16.1 Objectives

16.1.1 To familiarize the Trainee with alcohol pharmacology (pharmacokinetics and pharmacodynamics) to include retrograde extrapolation and the use of the Widmark's equation.

16.1.2 To familiarize the trainee with testimony regarding ethanol effects and calculations.

16.1.3 Successful completion of a technical examination, a practical test, and a mock trial.

16.2 Required Reading

16.2.1 Jones, A.W. (2011) Pharmacokinetics of Ethanol – Issues of Forensic Importance. Forensic Science Review, 23 (2) (July 2011), 92-132.

16.2.2 Jones, A.W. (2010) Evidence-based survey of the elimination rates of ethanol from blood with applications in forensic casework. Forensic Science International, 200, 1-20.

16.2.3 Goodman and Gilman's *The Pharmacological Basis of Therapeutics*, ethanol specific chapter (edition dependent).

16.2.4 Garriott *Medicolegal Aspects of Alcohol*, Ch 2-4 (pharmacology), 13-15 (impairment), 17-19 (testimony) (edition dependent).

16.2.5 Dubowski, K.M. (1985) Absorption, Distribution, and Elimination of Alcohol: Highway Safety Aspects. Journal of Studies on Alcohol, Supplement No. 10.

16.2.6 Winek, C.L. et al (1996) Determination of absorption time of ethanol in social drinkers. Forensic Science International, 77, 196-177.

16.2.7 Jones, A.W. (1993) Disappearance Rate of Ethanol from the Blood of Human Subjects: Implications in Forensic Toxicology. Journal of Forensic Science, 38 (1), 104-118.

16.2.8 Jones, A.W. and Anderson, L. (1996) Influence of Age, Gender, and Blood-Alcohol Concentration on the Disappearance Rate of Alcohol from Blood in Drinking Drivers. Journal of Forensic Science, 41(6), 922-926.

16.2.9 Stowell, A.R. and Stowell, L.I. (1998) Estimation of blood alcohol concentrations after social drinking. Journal of Forensic Science, 43(1), 14-21.

16.2.10 Gullberg, R.G. and Predmore, D.B. (1982) Variation in blood alcohol concentration following the last drink. Journal of Police Science Administration, 10(3), 289-296.

16.2.11 Jones, A.W. and Jonsson, K.A. (1994) Food-induced lowering of blood-ethanol profiles and increased rate of elimination immediately after a meal. Journal of Forensic Science, 39(4), 1084-1093.

16.2.12 Jakus, J.T., Shajani, N.K., Image, B.A. (1992) Consumption of a large dose of alcohol in a short time span. Forensic Science International, 56(2), 113-125.

16.2.13 Jones, A.W., Jonsson, K.A., Neri, A. (1991) Peak blood-ethanol concentration and the time of its occurrence after rapid drinking on an empty stomach. Journal of Forensic Science, 36(2), 376-385.

16.2.14 Watkins, R.L. and Adler, E.V. (1993) The effect of food on alcohol absorption and elimination patterns. Journal of Forensic Science, 38(2), 285-291.

16.2.15 Moskowitz, H., Burns, M., Fiorentino, D., Smiley, A., Zador, P. (2000) Driver characteristics and impairment at various BACs. DOT Technical Document.

16.2.16 Moskowitz, H., Burns, M., Williams, A. (1985) Skills performance at low blood alcohol levels. Journal of Studies on Alcohol, 46(6), 482-485.

16.3 Study Questions

16.3.1 Describe zero and first order elimination. Diagram each.

16.3.2 Mr. Jones was in an accident at 0015 hrs. He admitted to drinking 3 beers rapidly at 2330hrs. He submitted to a breath test at 0200 hrs and the result was 0.20%w/v. What would his blood alcohol concentration have been at the time of the accident?

16.3.2.1 Further investigation revealed that he had his last drink at 2200hrs, but the accident still occurred at 0015hrs. Estimate his blood alcohol concentration at 0015hrs.

16.3.2.2 At trial, Mr. Jones claimed that after the accident, but before the officers arrived at the scene, he had consumed an unknown quantity of whiskey that he kept in his car. Estimate his blood alcohol concentration at 0015hrs.

16.3.3 How many beers would Mr. Jones (Height: 5'10", Weight: 170lb, Age: 45, known alcoholic) have had to consume to reach 0.20%w/v? Assume two scenarios: 1) very rapid consumption (within 30 minutes) and 2) consumption over three hours.

16.3.4 Describe the effects of alcohol on human performance and how that correlates to driving skills.

16.3.5 Approximately how long would it take someone with a BAC of 0.31 g/210L to metabolize all the alcohol in the body?

16.3.6 Mrs. Brown (Age: 29, Height: 5'3", Weight: 214lbs) was stopped at 2335hrs for weaving in her lane. Upon investigation, the officer charged her with DUI and conducted a breath alcohol analysis at 0017hrs with a result of 0.23g/210L. Mrs. Brown stated that she stopped drinking during happy hour which was at approximately 1800hrs and only consumed three glasses of wine.

16.3.6.1 How many standard drinks would Mrs. Brown had to have consumed at happy hour (assume all drinks consumed from 1700-1800 hours) to produce a result of 0.23g/210L approximately six hours later.

16.3.6.2 Is the scenario provided by the defendant consistent with the information provided by the officer and the blood alcohol measurement? Please explain.

16.4 Technical Examination

16.4.1 Prior to the mock trial, a technical oral examination of the trainee will be conducted to ascertain the alcohol pharmacological and impairment knowledge of the individual. This will be limited to 3 hours.

16.4.2 After the examination, the evaluating members of the audience (minimally, the Program Manager and TC) will discuss the trainee's performance.

16.4.3 The outcome of the examination will be satisfactory or not satisfactory.

16.4.4 If the panel determines that the trainee's performance was not satisfactory, steps must be taken to determine and enact an appropriate action.

16.5 Practical Test

16.5.1 Following successful completion of the technical examination, the Trainee will be given a practical test.

16.5.2 The practical test will be a scenario which will require a retrograde extrapolation and the use of Widmark's equation.

16.5.3 Acceptable performance is obtaining the expected result for the calculations.

16.6 Mock Trial

16.6.1 This mock trial may be combined with the final mock trial for forensic toxicologist trainees.

16.6.2 A recorded mock trial will be conducted using the scenario and calculation for the practical test.

16.6.3 The Toxicology Program Manager must agree with the selection of all participants.

16.6.4 The atmosphere will be formal, that is, it will be conducted in the same manner as a real courtroom situation. This includes dress, conduct, protocol and all other aspects. Answers and explanations are to be directed as to a lay jury or judge.

16.6.5 The mock trial will not exceed 2 hours.

16.6.6 The role of the prosecutor will be assumed by the training coordinator or designee.

16.6.7 The mock trial may be stopped at any time upon request of any of the involved parties.

16.6.8 After the court, supervision/management will assess the trainee's performance.

16.6.9 The outcome of the mock trial will be satisfactory or not satisfactory.

16.6.10 If the panel determines that the trainee's performance was not satisfactory, steps must be taken to effect the appropriate action.

16.6.11 This evaluation will be immediately followed by a short performance critique.

16.6.12 The training coordinator will review the recording of the trial with the trainee as soon as possible. Other participants/observers should provide comments to the training coordinator as soon as possible.

17 PHARMACOLOGY AND TOXICOLOGY

17.1 Objectives

17.1.1 Display a working knowledge of the various categories of drugs encountered in toxicological analyses.

17.1.2 Understand the differences in interpretation for medical examiner (OCME) cases vs. driving under the influence of drugs (DUID) cases. Explain how the same drug concentration may be interpreted differently.

17.1.3 Know and understand the pharmacodynamic (PD) and pharmacokinetic (PK) properties of major drug classes.

17.1.4 Understand how the therapeutic, toxic, and lethal blood concentrations are assigned and used for populations, but may vary for an individual.

17.1.5 Explain the PD effects on human behavior and performance using blood drug concentrations as it pertains to court testimony and DUID cases.

17.1.6 Understand the process of postmortem redistribution, the interpretation of cases where this occurs, and which drugs are expected to undergo this process.

17.2 Methods of Instruction

17.2.1 Lectures and/or Self-Directed Study

17.2.1.1 SOFT Forensic Toxicology Review Course Lectures (2003)

17.2.1.2 Specific topics for each class of drugs.

17.2.1.2.1 General PK parameters (e.g., Vd, $t_{1/2}$, metabolism).

17.2.1.2.2 Major therapeutic and/or illicit uses.

17.2.1.2.3 Therapeutic effects.

17.2.1.2.4 Side effects.

17.2.1.2.5 Effects on driving.

17.2.1.2.6 Concentrations at which effects are observed.

17.2.1.2.7 Comparison of concentrations in DUID vs. postmortem cases.

17.2.1.2.8 Potential drug interactions.

17.2.1.2.9 Postmortem redistribution.

17.2.1.2.10 Practice trial testimony.

17.2.1.3 Literature review.

17.2.1.3.1 Levine, B. Principles of Forensic Toxicology. 2003.

17.2.1.3.2 Goodman and Gilman's The Pharmacological Bases of Therapeutics.

17.2.1.3.3 Garriott's Medicolegal Aspects of Alcohol.

17.2.1.3.4 SOFT Forensic Toxicology Review Course, Raleigh-Durham, NC. 2003.

17.2.1.3.5 National Highway Safety Traffic Administration. Drugs and Human Performance Fact Sheets. 2004.

17.2.1.3.6 The Effects of Drugs on Human Performance and Behavior. Forensic Science Review. 14. January 2002.

17.2.1.4 Discussion of interpretation and testimony.

17.2.1.5 Practice testimony on each drug class (mini mock trials).

17.2.1.6 Attend the Robert F. Borkenstein Effects of Drugs on Human Performance and Behavior Course and the Postmortem Interpretive Toxicology Course provided by the Center for Forensic Science Research and Education. These are considered mandatory contingent on resources (funding, availability).

17.3 Evaluation

17.3.1 Written study questions on each class of drugs.

17.3.2 Mini mock trials on each class of drugs.

17.3.3 The Trainee must be capable of answering questions on each class of drugs such as would be expected in courtroom scenario.

17.4 Pharmacodynamics and Pharmacokinetics

17.4.1 Lectures and/or Self-Directed Study

17.4.1.1 Neurotransmission, drug-receptor interactions, and dose/response.

17.4.2 Required Reading

17.4.2.1 Levine, B. Principles of Forensic Toxiclogy. 2003. Ch. 4 (PK/PD, edition specific).

17.4.2.2 Goodman and Gilman's The Pharmacological Basis of Therapeutics. Ch. 1-4, 12 (edition specific chapters; PK, PD, principles of therapeutics, principles of toxicology, neurotransmission).

17.4.3 Study Questions

17.4.3.1 Define pharmacokinetics and pharmacodynamics.

17.4.3.2 What factors influence absorption?

17.4.3.3 Will a weak base be absorbed primarily in the stomach or small intestine? Why? What about a weak acid?

17.4.3.4 Define bioavailability?

17.4.3.5 What is Vd? How is it calculated?

17.4.3.6 Describe zero and first order elimination. Diagram each (may use answer for Alcohol Pharmacology module).

17.4.3.7 Define first pass effect.

17.4.3.8 Give 5 examples of different routes of administration and a drug example for each. Describe how each route of administration would affect onset of action and peak blood concentration.

17.4.3.9 Give two examples of phase I and II reactions. Give a drug example for each.

17.4.3.10 Diagram a dose/response curve. What would be the effect of adding an antagonist? Add a non-competitive antagonist?

17.4.3.11 Diagram an neuronal synapse. Describe how reuptake inhibitors influence this environment.

17.4.3.12 Discuss the major structures of the brain that could be affected by drugs acting on the central nervous system.

17.4.3.13 What is therapeutic index? How is it calculated? Give an example of a drug with a high therapeutic index. Give an example of a drug with a low therapeutic index.

17.5 Opioids

17.5.1 Opioids to include, but not limited to, natural and synthetic opioids including fentanyl and fentanyl derivatives.

17.5.2 Required Reading

17.5.2.1 Levine, B. Principles of Forensic Toxicology. Ch 12 (edition specific, opioid chapter).

17.5.2.2 Goodman and Gilman's The Pharmacological Basis of Therapeutics, Ch. 23-24 (edition specific, opioid related chapters).

17.5.2.3 NHTSA: Methadone, morphine.

17.5.3 Study Questions

17.5.3.1 Differentiate between the terms opiate, opioid, and narcotics.

17.5.3.2 Discuss the structure-activity relationship of morphine and its opiate analogs vs. the opiate antagonist, naloxone.

17.5.3.3 Which of the following are used to synthesize opioids? Give specific products.

17.5.3.3.1 Morphine.

17.5.3.3.2 Codeine.

17.5.3.3.3 Papaverine.

17.5.3.3.4 Noscopine.

17.5.3.3.5 Thebaine.

17.5.3.4 Discuss absorption, distribution, metabolism, and elimination (ADME) of heroin and fentanyl.

17.5.3.5 Discuss the role of codeine and 6AM in the determination of whether a death involved heroin.

17.5.3.6 What is the classical clinical presentation of acute opiate toxicity?

17.5.3.7 Discuss the pharmacological CNS effects of opiates that would be relevant in a DUID case.

17.6 Cocaine/Benzoylecgonine

17.6.1 Required Reading

17.6.1.1 Levine, B. Principles of Forensic Toxicology. Ch. 13 (edition specific, cocaine).

17.6.1.2 NHTSA: cocaine.

17.6.1.3 FSR: cocaine.

17.6.2 Study Questions

17.6.2.1 What is contraction band necrosis?

17.6.2.2 What are the effects of cocaine on catecholamines?

17.6.2.3 What is neurotransmitter depletion? How is it related to cocaine use?

17.6.2.4 What are the effects of cocaine on drivers at the following concentrations?

17.6.2.4.1 Cocaine 0.02 mg/L, benzoylecgonine 0.3 mg/L

17.6.2.4.2 Cocaine ND, benzoylecgonine 2.0 mg/L

17.7 Cannabinoids

17.7.1 Required Reading

17.7.1.1 Levine, B. Principles of Forensic Toxicology. Ch. 14 (edition specific, cannabinoids).

17.7.1.2 NHTSA: cannabinoids.

17.7.1.3 FSR: cannabinoids.

17.7.2 Study Questions

17.7.2.1 A Commonwealth Attorney calls to discuss the following cases. What would you say?

17.7.2.1.1 THC 0.001 mg/L, THC-COOH 0.02 mg/L. Driver pulled over for bad driving, officer witnessed suspect throw joint out of the window, performed poorly on SFSTs.

17.7.2.1.2 THC 0.001 mg/L, THC-COOH 0.02 mg/L. Driver pulled over for broken taillight, defendant admitted to smoking a joint the night before, performed well on SFSTs.

17.7.2.2 Is there an established relationship between THC blood concentration and driving impairment? Discuss why or why not.

17.7.2.3 What are the major metabolites of THC? Are the active/inactive? Which ones does DFS analyze and why?

17.7.2.4 Describe ADME of THC.

17.7.2.5 THC has a broad spectrum of pharmacological effects. Describe each. Can THC be classified in one drug category?

17.7.2.6 Describe the effects of THC on driving.

17.8 CNS Depressants

17.8.1 CNS depressants to include, but not limited to, benzodiazepines, barbiturates, carisoprodol, zolpidem, GHB.

17.8.2 Required Reading

17.8.2.1 Levine, B. Principles of Forensic Toxicology. Ch. 11 (edition specific, CNS depressants).

17.8.2.2 Goodman and Gilman's The Pharmacological Basis of Therapeutics. Ch. 17 (edition specific, CNS depressants).

17.8.2.3 NHTSA: Benzodiazepines, GHB.

17.8.2.4 FSR: Carisoprodol, GHB, zolpidem.

17.8.3 Study Questions

17.8.3.1 Make a table listing major CNS depressant drugs analyzed in DUID cases. Include the following information for each drug:

17.8.3.1.1 Dosage form.

17.8.3.1.2 Therapeutic uses.

17.8.3.1.3 Therapeutic, toxic, lethal ranges.

17.8.3.1.4 Half-life

17.8.3.1.5 Detection time in blood, urine.

17.8.3.1.6 Typical adverse side effects.

17.9 Sympathomimetic Amine

17.9.1 Sympathomimetic amines to include, but not limited to, methamphetamine, amphetamine, MDMA, ephedrine, and methylphenidate.

17.9.2 Required Reading

17.9.2.1 Levine, B. Principles of Forensic Toxicology. Ch. 15 (edition specific, sympathomimetic amines).

17.9.2.2 Goodman and Gilman's The Pharmacological Basis of Therapeutics. Ch. 10 (edition specific, sympathomimetic amines).

17.9.2.3 NHTSA: Methamphetamine.

17.9.2.4 FSR: methamphetamine, MDMA.

17.9.3 Study Questions

17.9.3.1 What are the common neurotransmitter involved in sympathomimetic pathways?

17.9.3.2 What are the common structural properties of these neurotransmitters?

17.9.3.3 How does hydroxylation affect their action?

17.9.3.4 Compare ADME for methamphetamine and MDMA. Include concentrations that contribute to observed effects and discuss tolerance.

17.9.3.5 What "rave" accessory is used to provide protection from a common MDMA side effect?

17.9.3.6 Discuss the noted effects of methylone (or other novel psychoactive substances like methylone for which DFS provides testing).

17.9.3.7 Discuss the effects of methamphetamine and MDMA on driving.

17.9.3.8 Sympathomimetic amines are usually present in racemic mixtures. Describe the different properties of d and l methamphetamine and MDMA.

17.10 **Hallucinogens**

17.10.1 Hallucinogens to include, but not limited to, LSD, PCP, ketamine, and psilocybin.

17.10.2 Required Reading

17.10.2.1 Levine, B. Principles of Forensic Toxicology. Ch. 16 (edition specific, hallucinogens).

17.10.2.2 NHTSA: ketamine, LSD, PCP.

17.10.2.3 FSR: ketamine.

17.10.3 Study Questions

17.10.3.1 Which neurotransmitters are responsible for the hallucinogenic properties of compounds?

17.10.3.2 Compare ADME of LSD and PCP. Include dosage and detection times.

17.10.3.3 Discuss significant adverse effects of hallucinogenic drugs on driving.

17.10.3.4 What is the prevalence of hallucinogenic drug use in the general population?

17.11 Neuroleptics

17.11.1 Neuroleptics are also known as antipsychotics.

17.11.2 Required Reading

17.11.2.1 Levine, B. Principles of Forensic Toxicology. Ch. 19 (edition specific, neuroleptics).

17.11.2.2 Goodman and Gilman's The Pharmacological Basis of Therapeutics. Ch. 18 (edition specific, neuroleptics).

17.11.3 Study Questions

17.11.3.1 Give 2 examples each of old and new generation neuroleptics and describe ADME for each example.

17.11.3.2 What are some of the side effects of old and new generation neuroleptics?

17.11.3.3 What are some of the advantages of the new generation neuroleptics?

17.12 Antidepressants

17.12.1 Antidepressants to include, but not limited to, MAO, TCA, SSRI.

17.12.2 Required Reading

17.12.2.1 Levine, B. Principles of Forensic Toxicology. Ch. 18 (edition specific, antidepressants).

17.12.2.2 Goodman and Gilman's The Pharmacological Basis of Therapeutics. Ch. 19 (edition specific, antidepressants).

17.12.3 Study Questions

17.12.3.1 What are some of the side effects that would result from tricyclic antidepressant combined concentrations of 0.1 mg/L amitriptyline and 0.5 mg/L nortriptyline.

17.12.3.2 Compare and contrast mechanisms of actions, ADME, and side effects of TCAs, SSRIs, and MAOs.

17.13 Anticonvulsants

17.13.1 Anticonvulsants to include, but not limited to, phenytoin, carbamazepine, valproic acid, gabapentin, lamotrigine, and topiramate.

17.13.2 Required Reading

17.13.2.1 Levine, B. Principles of Forensic Toxicology. Ch. 17 (edition specific, anticonvulsants).

17.13.2.2 Goodman and Gilman's The Pharmacological Basis of Therapeutics. Ch. 21 (edition specific, anticonvulsants).

17.13.3 Study Questions

17.13.3.1 Drugs used to control seizures have varied chemical structures. Describe the following: phenytoin, carbamazepine, valproic acid, gabapentin, lamotrigine, and topiramate.

17.13.3.2 Describe the neurological pathways of seizure control.

17.13.3.3 Describe lethal toxicities associated with seizure medications.

17.13.3.4 Describe the metabolism of carbamazepine and its significance.

17.13.3.5 Describe the adverse effects of seizure medication on driving.

17.13.3.6 In OCME cases, what is the most important reason for the analysis of seizure medication?

17.14 Antihistamines/NSAIDs

17.14.1 Antihistamines/NSAIDs to include, but not limited to, diphenhydramine, promethazine, dextromethorphan, acetaminophen, and acetylsalicylic acid.

17.14.2 Required Reading

17.14.2.1 Goodman and Gilman's The Pharmacological Basis of Therapeutics. Ch. 25, 27 (edition specific, antihistamines and NSAIDs).

17.14.2.2 NHTSA: diphenhydramine, dextromethorphan.

17.14.3 Study Questions

17.14.3.1 Make a table of histamine receptors including localization within the body, antagonists associated with each and therapeutic uses, therapeutic/toxic/lethal levels, therapeutic effects and effects on driving for each antagonist.

17.14.3.2 Why do antihistamines have anticholinergic effects?

17.14.3.3 Describe postmortem redistribution of antihistamines.

17.14.3.4 What antihistamines can be used in a DFC? What screening method is used to detect them? What is their detection time in blood and urine?