Training Procedure for Drug Chemistry Gas Chromatography-Mass Spectrometry

1.0 Purpose - Gas Chromatography (GC) dates back to the early 1900's and may be linked to distillation, which dates to antiquity. The beginnings of Mass Spectrometry (MS) date to before 1920, but mass spectrometers were not produced commercially until around 1940. Today, the mass spectrometer is one of the most widely used instruments in analytical chemistry. It has both qualitative and quantitative applications in Forensic Drug Chemistry. There are different types of mass spectrometers in use. Ion Trap Mass Spectrometers, Time of Flight Mass Spectrometers (TOF), Quadrupole Mass Spectrometers, Laser Ionization Mass Spectrometers, Chemical Ionization Mass Spectrometers, and Liquid Chromatograph Mass Spectrometers are among many examples. This section will familiarize the Forensic Scientist with GC-MS basics and the hardware configuration of the Electron Impact (EI) Mass Selective Detector (MSD) through the answer of study questions. The trainee shall gain knowledge of the operation and maintenance of the GC-MS used in the Drug Section through experimental/practical exercises.

2.0 Scope - This procedure applies to trainees in the Drug Chemistry Sections of the State Crime Laboratory.

3.0 Procedure

3.1 Objectives

3.1.1 Review the Drug Chemistry Technical Procedure for Gas Chromatograph-Mass Spectrometry (GC-MS) and the Drug Chemistry Technical Procedure for Drug Chemistry Analysis, as it pertains to GC-MS.

3.1.2 Become familiar with the components of the GC-MS.

3.1.3 Understand basic GC-MS theory and concepts.

3.1.4 Gain practical knowledge of the use, operation, and maintenance of the GC-MS and successfully perform all calibration and quality control procedures pertaining to the GC-MS contained in the procedures listed in the Drug Chemistry Technical Procedure for Gas Chromatograph-Mass Spectrometry (GC-MS).

3.1.5 Observe Forensic Scientists use the GC-MS (load samples, retrieve data, etc.), and practice retrieving data from training samples run by the trainee and samples run by other Forensic Scientists.

3.1.6 Complete a written practical exercise on spectral identification using parametric retrieval software.

3.1.7 Obtain data from runs of both methamphetamine hydrochloride and methamphetamine base in order to compare quality of chromatography.

3.1.8 Complete a practical exercise using infrared spectroscopy, extraction techniques, and mass spectral data to identify two unknowns.

3.1.9 Collect mass spectral data and complete final identification of unknowns from the Infrared Training Module as needed.

3.1.10 Review the GC-MS section of the Drug Chemistry worksheet in FA with the GC-MS Coordinator(s) (or his/her designee).

3.1.11 Successfully complete a written exam on Gas Chromatography-Mass Spectrometry (GC-MS).

3.2 Study Questions

3.2.1 Name the basic components of a Gas Chromatograph-Mass Selective Detector (MSD) system and briefly describe how each component works.

3.2.2 Explain how the GC stationary phase and a mobile phase function to separate components of a sample. Give one example of each.

3.2.3 What is the difference between a split and splitless injection?

3.2.4 Explain what is meant by the term "split ratio." Give an example and describe how this can be helpful with over and under-concentrated samples.

3.2.5 Why are Drug Chemistry samples run with split injections?

3.2.6 Describe the function of the septum purge vent on a GC injector.

3.2.7 What is the function of an injection liner?

3.2.8 What would happen if the sample vapor volume exceeded the volume of the injection liner?

3.2.9 Describe two general types of GC columns.

3.2.10 What is the chemical composition of a DB-5 or comparable stationary phase?

3.2.11 Explain what is meant by constant flow and constant pressure. Which one does the Drug Chemistry Section use for solid dosage drug analysis?

3.2.12 Explain the difference between an isothermal program and a temperature program. Why does the Drug Chemistry Section use temperature programs?

3.2.13 Can decomposition occur in gas chromatography? If so, how can it be avoided?

3.2.14 The theory surrounding separation via gas chromatography is well studied and can be described mathematically. Define the following:

3.2.14.1 Signal to noise ratio. What value would differentiate between analyte and noise?

3.2.14.2 Resolution.

3.2.15 What is the difficulty in interfacing a Gas Chromatograph with a Mass Selective Detector?

3.2.16 Name the three major functional components of the quadrupole mass selective detector, and describe how each functions.

3.2.17 Explain the term "mean free path." How is this achieved in a mass spectrometer?

3.2.18 Define the term "base peak" with respect to a mass spectrum.

3.2.19 Define the term "molecular ion" with respect to a mass spectrum.

3.2.20 Explain the term mass defect.

3.2.21 What does tuning the mass spectrometer do? Give specifics for standard spectral tune versus autotune.

3.2.22 Explain the phenomenon of "spectral tilting."

3.2.23 Most MS systems have sophisticated search algorithms which perform mass spectral searches of unknown mass spectra. No search routine can provide conclusive identification 100 % of the time. Interpretation and identification is the responsibility of the Forensic Scientist. What are some factors that would affect a library search?

3.2.24 A Total Ion Chromatogram (TIC) is a plot of what type of data on the X and Y axes?

3.2.25 A Mass Spectrum (MS) is a plot of what type of data on the X and Y axes?

3.2.26 What are the requirements for retention time matches in the Drug Chemistry Section and when are they required?

3.3 Practical/Laboratory Exercises

3.3.1 Describe the effect of the change in oven temperature on a chromatogram.

3.3.2 Describe the effect of change in flow rate on a chromatogram.

3.3.3 Under the direct supervision of the GC-MS Coordinator (or his/her designee) or the Training Coordinator, successfully perform all calibration and quality control procedures contained in the Drug Chemistry Technical Procedure for Gas Chromatograph-Mass Spectrometry (GC-MS).

3.3.3.1 Refer to the Technical Procedure for Drug Chemistry Gas Chromatography-Mass Spectrometry (GC-MS) Appendix A or B (depending on your laboratory). Discuss with the instructor what could happen to a mass spectrum if it were collected while each of the listed parameters was out of specification.

3.3.4 Under the direct supervision of the GC-MS Coordinator (or his/her designee), replace the septum, syringe and injection liner of a GC. Discuss how to clip or change a column, and the required post-maintenance checks.

3.3.5 Review the data system settings and temperature programs used in the Drug Chemistry Section with the GC-MS Coordinator (or his/her designee) or the Drug Chemistry Training Coordinator.

3.3.6 Observe the Drug Chemistry Training Coordinator or his/her designee prepare to use a GC-MS, setup a sequence, run a sequence and analyze data files. Using the GC-MS, review the data files provided by the Drug Chemistry Training Coordinator. The data

files consist of sets of substances that produce similar mass spectra. Attempt to group the substances into pairs and identify each substance. Describe the criteria used to differentiate between the substances.

3.3.7 Perform analyses on known samples provided by the Drug Chemistry Training Coordinator using the Drug Chemistry Technical Procedure for Gas Chromatography Mass Spectrometry (GC-MS). These may include extracted samples from the Training Section on Origins, Extractions and Separations of Drugs, or samples from training standards issued by the Training Coordinator.

3.3.7.1.1 Sample preparation techniques, including proper blank preparation, should be discussed with and approved by the GC-MS Coordinator (or his/her designee) or the Training Coordinator prior to loading samples on the instrument.

3.3.7.1.2 Demonstrate the following for the GC-MS Coordinator (or his/her designee) or Training Coordinator:

- Change the scale of the TIC (e.g. from largest peak to 200,000 and then back to largest peak)
- Change libraries that are used for the spectrum search. Discuss the use of each available library (e.g. certified versus non-certified)
- Use parametric retrieval to look at a library entry of the instructor's choosing.
- Use the Extracted Ion Chromatogram software to check for the presence of a specific compound in a sample.

3.3.8 Review the mass spectra for dextromethorphan/levomethorphan and pseudoephedrine/ephedrine. Is it possible to identify optical isomers using mass spectral data?

3.3.9 What change would occur in the TIC and the MS if the multiplier voltage were increased? The standard energy for the beam of ionizing electrons in EI MS is 70 eV. What would be the effect if the voltage of the ionizing source were changed?

3.3.10 Propose molecular structures for m/z ions in the following mass spectra, and answer the questions:

3.3.10.1 Blank / baseline – 44. What is the origin of the 207 m/z ion?

3.3.10.2 PFTBA - 69, 219, 502. The m/z 32 and 28 m/z ions are not ions of PFTBA. What would cause these ions?

3.3.10.3 Cocaine - 105, 182

3.3.10.4 Methamphetamine - 58, 91. What is the name of the 91 m/z ion?

3.3.10.5 Amphetamine – 44, 91

3.3.10.6 Alprazolam – 77. Explain the significance of the 279 / 281 m/z pair and the 308 / 310 m/z pair.

3.3.10.7 4-bromo-2,5-dimethoxyphenethylamine (BDMPEA/2C-B) - 215. Explain the significance of the 215/217 m/z, 230/232 m/z, and 259/261 m/z pairs.

3.3.10.8 Procaine – 86, 120

3.3.10.9 Benzocaine – 120, 92

Required Reading

3.4 *Agilent GC-MSD ChemStation and Instrument Operation Student Manual Course Number H4043A* Volume 1, Revision E.02.xx, Agilent Technologies: printed February 2008, pages 1-32, 39-73, 81-122.

4.0 References

Hewlett Packard / Agilent Technologies. *GC MSD ChemStation and Instrument Operation Student Manual*, Vol. I & II, (Manual Part Number H4043-90000). Hewlett Packard: April 1997.

McLafferty, F.W. *Interpretation of Mass Spectra.* 2nd Ed. W.A. Benjamin and Sons: 1973.
http://www.chem.agilent.com

5.0 Records

- Drug Chemistry Training Checklist
- Section Completion Summary

6.0 Attachments – N/A

Revision History		
Effective Date	Version Number	Reason
04/18/2014	1	Original Document - Combined the Drug Chemistry Training Procedure for Gas Chromatography and the Drug Chemistry Training Procedure for Mass Spectrometry to create a new Training Procedure
10/19/2015	2	**Header –** Revised issuing authority **Purpose** – corrected typo **3.1.6 through 3.1.9** – Added new objectives **3.2** – Edited study questions.
08/17/2018	3	**3.1.10** – Added new objective **3.2.4** – Expanded question to encompass over or under-concentrated samples **3.2.12** – Corrected "in" to "between" **3.2.21** – Clarified question and added specifics for standard spectral tune versus autotune **3.3.3, 3.3.5, 3.3.7.1.2** – Changed "Key Operator" to "Coordinator (or his/her designee)" **3.3.4** - Changed "Key Operator" to "Coordinator" **3.3.7.1.1** – Added proper blank preparation statement and changed "Key Operator" to "Coordinator (or his/her designee)" **3.3.8** – Changed "dextropropoxyphene/levopropoxyphene" to "pseudoephedrine/ephedrine" **3.3.10** – Removed all images, changed "m/e" to "m/z" **3.3.10.1, 3.3.10.2, 3.3.10.4, 3.3.10.6, 3.3.10.7** – Changed "m/e" to "m/z" **3.3.10.7** – Corrected typo and added additional names