Name of Procedure:

Hewlett-Packard/Agilent 6890 GC interfaced to the Hewlett-Packard/Agilent 5973 and 5975 Series MSD/DS

Suggested Uses:

The gas chromatograph/quadrupole mass selective detector/data system is used to qualitate and quantitate compounds present in items of evidence.

The gas chromatograph/mass spectrometer separates mixtures of compounds and produces mass spectra of compounds. The mass spectrum of a compound can be compared to a reference spectrum for identity confirmation. If necessary, mass spectral libraries can be searched through computer-based matching software to aid in identifying unknown compounds.

Apparatus Used to Perform Procedure:

Hewlett-Packard/Agilent Gas Chromatograph 6890 (GC)
Hewlett-Packard/Agilent 5973 Series Mass Selective Detector (MSD) or
Hewlett-Packard/Agilent 5975 Series Mass Selective Detector (MSD)
Hewlett-Packard/Agilent Automatic Liquid Sampler
PC with Hewlett-Packard Analytical MSD Productivity ChemStation Software, or equivalent
Computer Printer or other data output device
Methanol
Hexane
Chloroform
Ethyl Acetate
Sample vials and caps
(Crimper/decrimper tools if not using snap-top sample vials)
10*u*l syringe
DB-5 GC Column (or other appropriate column)
Helium Gas
Perfluorotributylamine [FC-43]

Calibration of the Hewlett Packard 6890 GC/ 5973MSD and 6890 GC/5975MSD:

1. The GC-MS is kept on at all times.
2. Calibration is performed daily when in use with the Autotune program, using the Standard Spectra Tune option.
3. This procedure uses Perfluorotributylamine (PFTBA) as a tuning standard and the resulting data file is kept in a notebook near each instrument.
4. Compare this Standard Spectra Tune file to previous ones and address any major variations which may indicate instrument problems.

    a. The three tuning masses in the upper profile part of the report should be within +/- 0.1 amu of 69.00, 219.00, and 502.00. Any deviation larger than +/- 0.1 amu indicates a problem. Correct the problem and re-tune the instrument.
    b. The peak widths of these three peaks should be 0.55 +/- 0.05 amu.
    c. The relative abundances should show that the peak at 69 amu corresponds to 100%.
        1) Relative to that peak, the peak at 219 amu should be in the 40-85% range.
        2) The peak at 502 amu should be greater than 2% but less than or equal to 5%.
    d. An air leak within the system may be indicated if the mass-to-charge ratio (m/z) at 28 is greater than m/z 18, or if either of these m/z ratios are greater than 10%.
        1) If an air leak is detected, the air leak should be isolated and corrected.
        2) The instrument then needs to be tuned again.

5. If any of the above stated parameters are out of specification, correct the problem, and tune the instrument again. If the problem persists, contact the service engineer.
6. The RTE integrator will be used with default settings to integrate peaks on sample and standard data.
7. Inject two standard solutions on a weekly basis to verify GC performance and GC retention times. (Run these solutions within a 7-day period prior to casework.)

    a. The High Temperature Standard will be run on the HIGH temperature program and the solution shall consist of:
        - •Cocaine
        - •Diazepam
        - •Hydrocodone
        - •Oxycodone
        - •Heroin
        - •Alprazolam

    b. The Low Temperature Standard solution will be run on the LOW temperature program and consist of:
        - •Amphetamine
        - •Phentermine
        - •Methamphetamine
        - •3,4-Methylenedioxyamphetamine (MDA)
        - •3,4-Methylenedioxymethamphetamine (MDMA)

    c. If the Pseudoephedrine Standard is needed for retention time comparison, run it

on the LOW temperature program on an as needed basis, within a 7-day period before or after the case sample.

d. Weekly standard solutions should be named with the designation LO or HI and the date on which they are run. The corresponding blank should have a similar name with a designation that it is a blank.

8. If maintenance is performed that may affect retention times, the standard solutions will be re-run to reflect new retention times.

9. The instrument will be evaluated to determine if it is in compliance before performing casework if retention times shift greater than 2.0% between standard drug runs.

10. A copy of the corresponding blanks, along with the chromatograms from the Low Temperature Solution and the High Temperature Solution injections displaying integrated peaks and retention times will be placed in the "GC RT" section of the notebook for each instrument.

Application of Procedure:

These procedures do not cover every aspect of the instrument. The operator of the instrument should consult the manual(s) for the instrument.

A. Sample Preparation (suggested)

1. Powders and residues: Filter with appropriate solvent to prevent particulate matter and undesired compounds from being introduced onto the column (example: sugars). Derivatizing agents may be used when needed.

2. Tablets:
   a. Alprazolam, Lorazepam, Diazepam, etc.: Add several drops of solvent to an intact (uncrushed) tablet(s). Allow the tablet to soaks for a short time. Transfer the solvent through a filter to a sample vial or insert and add more solvent for analysis.
   b. Coated tablets: Remove coating before adding several drops of solvent to the remaining intact tablet(s), prepare as described above. Pharmaceutical tablets should be extracted to remove large amounts of Acetaminophen or Aspirin prior to running on the GC/MS.
   c. Sulfates need to be extracted/converted before they can be introduced into the instrument.

3. Syringes: Wash with methanol and extract if necessary. (If excessive quantities of blood or other liquids are present in a syringe then an extraction is required).

B. GC/MS Methods:

The following are standard GC/MS methods used in the Drug Chemistry Section:

1. **HIGH**
   2 minutes initial time, 120°C initial temperature, 15°C/minute, 275°C final temperature, 15 minutes final time, 27.33 minutes total time.
   Used for cocaine, opiates, and general use.

2. **LOW**
   2 minutes initial time, 70°C initial temperature, 15°C/minute, 275°C final temperature, 15 minutes final time, 27.67 minutes total time.
   Used for phenethylamines and other volatile compounds.

3. **HVYHI** (Heavy High)
   2 minutes initial time, 120°C initial temperature, 15°C/minute, 275°C final temperature, 70 minutes final time, 82.33 minutes total time.
   Used for steroids and other high molecular weight compounds.

Each of these methods can be used at split ratios of 5:1, 20:1, or 100:1.
Splitless injections can be used when needed.
Numbers or an abbreviation in front of the method name indicates the split ratio.

C. Sample Injection:

1. Prior to the injection of a sample, a ◘blank◘ solvent injection shall be made. The solvent shall be prepared by the individual chemist at the time of sample preparation and be the same solvent from the same bottle used in the sample preparation.

2. The temperature program for the “blank” solvent injection will be the same one used for the injection of the corresponding sample. The split ratio should be the same for both injections.

3. The syringe must be flushed at least 10 times with clean solvent between injections to ensure the sample integrity between injections and that no sample

transfer is made between sample vials. Methanol will be used in the first wash solvent bottle and hexane or chloroform in the second solvent wash bottle.

D. Data:

After the data system has collected the data, examine the chromatogram and spectra for the peaks of interest, print (or electronically transfer) the following for the case file:

1. Total Ion Chromatogram (TIC) for the corresponding blank, including the case number and item number.
2. The Total Ion Chromatogram (TIC) for the sample, including the case number and item number.
3. The mass spectrum of peaks of interest.
4. The expanded mass spectrum of phenethylamines, labeled as such.

E. Reporting:

The requirements for drug/chemical identification using the GC/MS system are the approximate relative retention time for the column and method used, and a reasonable comparison between a standard and the identified drug/chemical□s mass spectra.

1. If the retention time is being used as a confirmatory test for identification, a standard of the drug must be run to show retention times are consistent within +/- 2.0%. The standard can be run before or after the case file sample, within a seven day period, assuming no column maintenance has been performed that would change the retention times.

2. The standard data must be included in the case file and consist of:
   a. Total Ion Chromatogram for corresponding blank, including data file name.
   b. Total Ion Chromatogram for sample, including data file name.
   c. Mass spectrum of peaks of interest.

F. Instrument Logbook:

A logbook consisting of four sections will be maintained adjacent to each instrument.

1. The □Log□ section will document injections made on the instrument beside the appropriate date. The log will include the date, sample identification, initials of

operator, GC/MS method used, and comments. Weekly septum changes will be noted on the right side of the row beside the appropriate date. Any unusual error messages or the like should be recorded in this section. This will assist the Key Operator and the GC/MS Coordinator when diagnosing instrument problems.
Use the attached form to document injections and other data listed above.

2. If samples are rerun for any reason, a new entry will be placed in the logbook. (Solvent runs used for maintenance purposes need not be recorded.)
3. The ◘Tune◘ section will contain daily tune reports.
4. The ◘Maintenance◘ section will document the date, descriptions of work performed, parts replaced, and the initials of the person performing the maintenance.
   (Weekly septum changes do not need to be logged in the maintenance section. These entries can be recorded in the ◘Log◘ section as outlined above.)
   Use the attached form to document maintenance performed.
5. The ◘GC RT◘ section will contain all GC retention time verification data.
   All instrument logbooks will be restarted with blank activity log forms on or about the first workday of the new calendar year.
   Activity logs, Daily Tunes, and GC Retention Data from each instrument notebook will be compiled yearly, labeled with instrument serial number, and placed in a packet near the instrument.
   Data may be archived to the packet as the year progresses and the notebooks become full.

G. Sequence Files:

The current date can be used to name the first sequence of the workday. These sequences can be deleted after approximately one week and do not need to be archived.

H. Data Files:

1. Data file names should include the year designation and the case file number. This will ensure data from different years with the same file number will not be overwritten.
2. Data files associated with casework and standard solutions will not be deleted or overwritten. Data will be archived and labeled with the instrument serial number and dates as needed. Notify the Key Operator or the GC/MS Coordinator if the disk drive(s) become full; they are responsible for resolving this situation.

I. Weekly Standard Solutions:

1. Chemists may add samples to the sequence behind the Standard Solutions. It will be the Key Operator□s responsibility to ensure the daily tune data is reviewed, printed and filed in the □Tunes□ section of the Activity Log. A note will be placed on the □log□ page if any problems occur to prevent starting new samples.
2. It will be the Key Operator□s responsibility to run the weekly Standard Solutions. However, chemists are responsible to ensure the standards are acceptable for the week before running samples. If a problem exists, whoever finds the problem will notify all those who have run samples that day. Case file samples will be re-run after acceptable standard data is collected.

J. Instrument Problem Reporting:

1. Key Operators should be notified of any problems related to instrument performance.
2. The key operator should notify the GC/MS Coordinator of any performance problems they cannot solve.
3. Schedule requests for outside service through the GC/MS Coordinator, SAC or ASAC On-Call.
4. Key operators are responsible for ensuring the above actions are carried out.

Safety Concerns:

A. Avoid syringe punctures of hands and fingers.
B. Use extreme caution handling organic solvents to avoid contact with skin and eyes.
C. Use extreme caution dismantling/installing/transporting compressed gas cylinders.
D. Caution: Gas Chromatograph and Mass Spectrometer may be extremely hot.

Literature References:

Agilent 6890 GC Instrument Manuals

Hewlett-Packard 5973 and 5975 Instrument Manuals

Moffat,Jackson, Moss and Widdop, **□Clarke□s Isolation and Identification of Drugs□**; 2nd Ed., Vol. 1, 1986.

Skoog,Holler, Nieman, ◘**Principles of Instrumental Analysis**◘;5th Ed., 1998.