Toxicology GC/MS Performance Monitoring and Maintenance

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Toxicology GC/MS Performance Monitoring and Maintenance

1 INTRODUCTION

This document addresses the performance monitoring and maintenance of the Agilent GC/MS system prior to the analysis of toxicological samples. The Agilent GC/MS system consists of an Agilent Gas Chromatograph (GC) with a single quadrupole Mass Selective Detector (MSD) Mass Spectrometer (MS). The system may also be equipped with an additional detector, such as a Thermal Conductivity Detector (TCD). Definitions and guidelines are outlined in IOSS-701.

2 SCOPE

This document applies to personnel using the associated instrument(s)/equipment in the Toxicology discipline.

3 EQUIPMENT

- Instrumentation
    - Agilent 7890 Gas Chromatograph, Agilent 5977 Mass Selective Detector with EI Source, Agilent TCD (if equipped) and Chemstation/Masshunter Software (or equivalent)
    - Gerstel MPS2 automated sampler, accessories, and Gerstel Maestro Software (or equivalent)
- Materials
    - Agilent J&W DB-5 MS, 30 m, 0.25 mm i.d., 0.25 µm film (or equivalent) (MSD)
    - Agilent Molsieve, 30 m, 0.32 mm i.d., 12 µm film (or equivalent) (TCD – if equipped)
    - Helium, 99.99% (high purity)
    - Perfluorotributylamine (PFTBA, FC-43) (Agilent or equivalent)
    - 2, 10 or 20 mL GC vials, crimp or screw top, with or without 100-500 µL inserts (Agilent or equivalent)
    - 4 mm split-splitless tapered injection port liners and o-rings (Agilent or equivalent)
    - Low-bleed 11 mm injection port septa (Agilent or equivalent)
    - Hamilton 701ASN 10 µL autosampler syringes (or equivalent) (for liquid injection)
    - 1 mL or 2.5 mL Gerstel headspace syringe (or equivalent) (for headspace sampling)
    - Methanol (Optima Grade or equivalent)
    - Caffeine Stock Standard (1 mg/mL):
      A methanolic solution purchased from Cerilliant or other approved vendor. Stability and storage are determined by the manufacturer.
    - Fentanyl Stock Standard (1 mg/mL):
      A methanolic solution purchased from Cerilliant or other approved vendor. Stability and storage are determined by the manufacturer.
    - MDEA Stock Standard (1 mg/mL):

A methanolic solution purchased from Cerilliant or other approved vendor. Stability and storage are determined by the manufacturer.

- Oxycodone Stock Standard (1 mg/mL):
  A methanolic solution purchased from Cerilliant or other approved vendor. Stability and storage are determined by the manufacturer.
- Secobarbital Stock Standard (1 mg/mL):
  A methanolic solution purchased from Cerilliant or other approved vendor. Stability and storage are determined by the manufacturer.
- Trazodone Stock Standard (1 mg/mL):
  A methanolic solution purchased from Cerilliant or other approved vendor. Stability and storage are determined by the manufacturer.
- Formic acid (~89%, reagent grade)
- 0.05 M formic acid solution (GC/TCD performance check):
  Dilute 215 µL of formic acid to 100 mL with deionized water in a graduated cylinder or flask. Mix well and store in glass at room temperature. Stable for at least one year.
- Concentrated Sulfuric Acid
- General laboratory supplies

4 STANDARDS AND CONTROLS

4.1 PFTBA Tuning Solution (MSD)

The PFTBA tuning solution is used for tuning the mass spectrometer and verifying mass calibration. It is supplied by the instrument manufacturer and does not expire. It is stored in a glass container attached to the MSD.

4.2 Tox Testmix (10 µg/mL each of Caffeine, Fentanyl, MDEA, Oxycodone, Secobarbital; 40 µg/mL of Trazodone)

The testmix is used to assess daily operating performance, mass assignment, and continued integrity of the system. This preparation may be appropriately scaled up.

A. Pipet 250 µL each of Caffeine, Fentanyl, MDEA, Oxycodone, Secobarbital Stock Standards, and 1 mL Trazodone Stock Standard into a 25-mL volumetric flask.
B. Bring to the mark with methanol and mix well.
C. Store the solution in the refrigerator. It has a shelf-life of three years.

4.3 TCD Testmix (Carbon Monoxide Performance Standard)

A. To a 20 mL autosampler vial, add 1 mL of concentrated sulfuric acid.
B. Add 50 µL of a 0.05 M formic acid solution.
C. Immediately crimp-seal the autosampler vial and vortex for 10 seconds.
D. Incubate autosampler vial at 100°C for 60 minutes in a laboratory heating block. Carbon Monoxide (CO) is produced quantitatively from the dehydration of formic acid in sulfuric acid.

5 PROCEDURE

5.1 Daily Checks

The following steps will be performed daily, regardless of the detector to be used. Enter the appropriate information in the instrument log.

A. Check to ensure that the GC wash vials are filled, the waste vials are empty, and all are in the appropriate positions.
B. Record the remaining disk space on the hard drive. Verify that the hard disk has at least 100 MB of free disk space. Do not use if less than 100 MB remain.
C. Record the line pressure of the building helium supply (carrier gas). The regulator should read 50 p.s.i. or above. If it cannot maintain this pressure, contact appropriate instrument support personnel. If the instrument is supplied by a gas cylinder, record the tank pressure. Change the tank if less than 100 p.s.i. is remaining.

5.1.1 MSD Daily Checks

The following steps will be performed daily when using the MSD. Enter the appropriate information in the instrument log.

A. Check the Ion Gauge to ensure that there are no significant leaks in the system. Do not use if the pressure is higher than 6×10^{-5} torr.
B. Perform a tune of the instrument. If Autotune (ATUNE) is selected, the mass spectrometer will tune itself using PFTBA. Evaluate the results using the 'Acceptance Criteria' section. If the results are acceptable, save and print the tune file (ATUNE) when completed.
C. Perform an analysis of the Tox Testmix. Alternatively, if a specific analyte or analyte class is of interest, an appropriate testmix may be substituted. Open the appropriate testmix instrument method, and verify the parameters as listed in the 'Instrumental Conditions' section. Set up a sequence, load the autosampler with a vial containing the Tox Testmix, and start the analysis. Evaluate the results using the 'Acceptance Criteria' section. If the results are acceptable, print the TIC, RICs, and spectra for all six components in the Tox Testmix.
D. If all requirements are within specification, prepare the documentation as outlined in IOSS-701. If any requirements fail, contact appropriate instrument support personnel.

5.1.2 TCD Daily Checks

The following steps will be performed daily when using the TCD. Enter the appropriate information in the instrument log.

A. Ensure a 1.0 or 2.5 mL Gerstel headspace syringe or equivalent is loaded into the autosampler.
B. Ensure that the headspace autosampler injects into the back inlet of the GC.
C. Check the status of the TCD using the front panel controls of the GC.

D. Perform an analysis of the TCD Testmix. Open the appropriate testmix instrument method, and verify the parameters as listed in the 'Instrumental Conditions' section. Set up a sequence, load the autosampler with a vial containing the TCD Testmix, and start the analysis. Evaluate the results using the 'Acceptance Criteria' section. If the results are acceptable, print the chromatogram.
E. If all requirements are within specification, prepare the documentation as outlined in IOSS-701. If any requirements fail, contact appropriate instrument support personnel.

5.2 Yearly Scheduled Maintenance/Checks

The following steps will be performed once in a calendar year. Enter the information in the appropriate instrument log to indicate completion. Refer to IOSS-701 for more information on instrument maintenance and documentation.

A. Replace the GC split vent trap(s).
B. Check the overall column performance. Clip the inlet side of the GC column(s) if needed.
C. Check the MSD calibrant level. Fill if needed.
D. Check the MSD filaments and overall MSD performance. Replace blown filaments and clean source if needed.

5.3 As Needed Maintenance/Checks

The following steps are to be performed as needed. Enter the information in the appropriate instrument log to indicate completion. Refer to IOSS-701 for more information on instrument maintenance and documentation.

A. Replace the septum in the GC injection port(s).
B. Replace the liner and o-ring within the GC injection port(s).
C. Replace the gold seal within the GC injection port(s).
D. Check the GC syringe in the autosampler. Replace if needed.
E. Check the TCD. Bake out if needed.

6 INSTRUMENTAL CONDITIONS

6.1 GC/MS

6.1.1 Gas Chromatograph

Oven	
Initial Temperature:	60°C
Initial Time:	2.0 min
Ramp:	35°C/min
Final Temperature:	280°C
Hold Time:	26.71 min
Inlet/Injector	
Injection Volume:	1 µL
Inlet Temperature:	220°C
Mode:	Split
Split Ratio:	10:1
Flow Mode:	Constant flow
Initial Flow:	1.2 mL/min
Average Linear Velocity:	40 cm/sec
Carrier Gas:	Helium
Column	
Type:	DB-5 MS
Length:	30 m
Diameter:	0.25 mm
Film Thickness:	0.25 µm

6.1.2 Mass Spectrometer

Scan Mode:	Full Scan
Scan Range:	35-500 m/z
Transfer Line Temp:	280°C
Quad Temperature:	150°C
Source Temp:	200°C

6.2 GC/TCD

6.2.1 Autosampler

Oven Temperature:	60°C
Valve Temperature:	115°C
Transfer Line Temp:	115°C
Oven Stabilization Time:	0.1 min
Sample Shaking Rate:	Low
Sample Equilibration Time:	0.6 min
Vial Pressurization Time:	0.3 min
Loop Fill Time:	0.04 min
Loop Equilibration Time:	0.02 min
Sample Injection Time:	0.2 min

6.2.2 Gas Chromatograph

Oven	
Temperature:	40°C
Run Time:	4.0 min
Equilibration Time:	0.2 min
Inlet	
Inlet Temperature:	250°C
Mode:	Split
Split Ratio:	3:1
Flow Mode:	Constant flow
Carrier Flow:	13.2 mL/min
Carrier Gas:	Helium
Column	
Type:	Molsieve
Length:	30 m
Diameter:	0.32 mm
Film Thickness:	12 µm

6.2.3 TCD

Temperature:	250°C
Reference Flow:	20.0 mL/min
Makeup Flow:	2.5 mL/min
Makeup Gas:	Helium

7 ACCEPTANCE CRITERIA

7.1 Autotune

Compare the results of the Autotune to previous tune results. Significant voltage increases, changes in the isotope ratios or elevated air/water indicate the need to initiate corrective maintenance procedures. The following are typical ATUNE values for the MSD:

A. PFTBA tune:
   - m/z ±0.4 for m/z 69, 219, and 502

B. Peak width:
   - 0.45-0.65

C. Relative abundance:
   - 69 greater than 50%
   - 219 greater than 50%
   - 502 greater than 1%
   - 18 and 28 (water & nitrogen) each 5% or below

7.2 Testmix

7.2.1 Tox Testmix

Verify the results of the Tox Testmix.

A. In order for the instrument to be considered in good operating condition, all six components should generate well resolved, symmetrical peaks with baseline separation.
B. A SNR of 3:1 will be the minimum response necessary to consider a response a peak.
C. There should be no significant extraneous peaks in the chromatogram.
D. When analyzing the Tox Testmix, the following ions should be observed in the mass spectra of the RICs (in order of elution) and their mass assignments should be within ± 0.5 m/z:
    - MDEA 72
    - Caffeine 194
    - Secobarbital 168
    - Oxycodone 315
    - Fentanyl 245
    - Trazodone 205

7.2.2 TCD Testmix

Verify the results of the TCD Testmix.

A. The CO peak should be well separated from the nitrogen and oxygen peaks (>0.5 min baseline separation), and have a readily detectable peak.
B. The GC column used in this procedure is a molecular sieve column, which may retain water. The column may be reconditioned by heating the GC oven to 225°C for >4 hours or overnight. Insufficient column conditioning results in poor chromatographic separation between the CO and air peaks.

8 LIMITATIONS

Only properly trained personnel will perform duties involved in the operation, maintenance, or troubleshooting of this instrument.

9 SAFETY

Many instrument components are held at temperatures of 250°C and higher. Precautions should be taken to prevent the contact of skin with heated surfaces and areas.

10 REVISION HISTORY

Revision	Issued	Changes
01	10/04/2018	Section 1- Scope to include disciplines/categories of testing. Deleted Calibration section and renumbered. Section 5- Updated heading. Section 6- Added 'appropriate instrument support personnel' throughout. Added alternative Testmix. Section 8- Updated to account for instrument variation and maintenance. Section 13- Updated 'Instrument Operation and Systems Support'.
04	09/15/2022	Revised to match new format requirements. Section 5- Added annual maintenance and expanded as-needed maintenance.