Q-Exactive LC/MS Performance Monitoring and Maintenance

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Q-Exactive LC/MS Performance Monitoring and Maintenance

1 INTRODUCTION

This document addresses the performance monitoring and maintenance of the Q-Exactive LC/MS system consisting of a Thermo Electron Q-Exactive Mass Spectrometer (MS) and a Waters Ultra-High Performance Liquid Chromatograph (UPLC). This system can be used for high resolution accurate mass analyses. The instrument is configured with an API source that is capable of electrospray ionization (ESI), atmospheric pressure chemical ionization (APCI), or atmospheric pressure photoionization (APPI). The instrument is primarily used in ESI mode. However, in can be used for APCI or APPI provided the method of ionization is clearly labeled in the resulting data and documentation. Definitions and guidelines are outlined in IOSS-701.

2 SCOPE

This document applies to personnel using the associated instrument(s)/equipment in the following disciplines/subdisciplines: General Chemistry, Explosives Chemistry, and Seized Drugs.

3 EQUIPMENT

- Instrumentation
  - Thermo Electron Q-Exactive MS, API Source, and Xcalibur software (or equivalent)
  - Waters Acquity UPLC (or equivalent)
- Materials
  - Nitrogen, 99.99% (high purity or equivalent)
  - Methanol (Optima grade or equivalent)
  - Deionized Water, 18.2 MΩ·cm (Milli-Q or equivalent)
  - Acetone (HPLC grade or equivalent)
  - Ammonium Nitrate (NH_4NO_3) (reagent grade or equivalent)
  - Caffeine (Sigma or equivalent)
  - Codeine (Sigma or equivalent)
  - Brucine (Sigma or equivalent)
  - Reserpine (Sigma or equivalent)
  - γ-Aminobutyric Acid (GABA),(Sigma or equivalent)

Redacted

  - 3.125 mM Ammonium Nitrate Mobile Phase (250 mg to 1 Liter water)
  - Waters Cortecs UPLC C18 1.6 µm, 2.1 mm X 50 mm (or equivalent)
  - 10 µL LC syringe (Hamilton or equivalent)
  - 100, 250 or 500 µL LC syringe (Hamilton or equivalent)
  - Pierce LTQ ESI Positive Ion Calibration Solution (Thermo or equivalent)
  - Pierce ESI Negative Ion Calibration Solution (Thermo or equivalent)
  - General laboratory supplies

4 STANDARDS AND CONTROLS

4.1 Testmix (General Chemistry, Seized Drugs)

The testmix is used to assess daily operating performance, mass assignment, and continued integrity of the system. Record preparations in the appropriate discipline/subdiscipline reagent log.

- Stock Solution
  Weigh 1.5 mg GABA, 5.0 mg caffeine, 1.0 mg codeine, 1.0 mg brucine, and 1.0 mg reserpine into a 100-mL volumetric flask. Bring to the mark with methanol and mix well. Shelf life is three years when stored refrigerated in brown glass. This preparation may be appropriately scaled.
- Testmix Solution
  Pipette 4.0 mL of the Stock Solution into a 100-mL volumetric flask. Dilute to the mark with methanol and mix well. Shelf life is three years when stored refrigerated in brown glass. This preparation may be appropriately scaled.

4.2 Testmix (Explosives Chemistry)

The testmix is used to assess daily operating performance, mass assignment, and continued integrity of the system. Record preparations in the appropriate discipline/subdiscipline reagent log.

- 100 µg/mL Stock Solution
  Place 1 mL of each Redacted standards in a separate 10 mL volumetric flask and dilute to the mark with acetone to achieve a final concentration of 100 µg/mL. Shelf life is two years when stored refrigerated in colored glass. This preparation may be appropriately scaled.
- 10 µg/mL Stock Solution
  Pipette 1 mL of each Redacted into a 10 mL volumetric flask and dilute to the mark with acetone to achieve a concentration of 10 µg/mL. Shelf life is two years when stored refrigerated in colored glass. This preparation may be appropriately scaled.
- Testmix Solution
  For daily use, dilute 20 µL of the 10 µg/mL stock solution to 1 mL with a 50:50 solution of methanol and water.

4.3 Calibration Solutions

The calibration solutions are used for coarse tuning and calibrating the mass spectrometer over the entire mass range. This procedure only needs to be performed when warranted based on system performance or if the system has been significantly moved, down for a long period of time, or undergone a major repair. The calibration solutions are purchased from Thermo Fisher Scientific or equivalent.

5 PROCEDURE

5.1 Daily Checks

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

A. 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.
B. Record the line pressure of the building nitrogen supply (API gas). The regulator should read between 70 and 100 psi. If it cannot maintain this pressure, contact appropriate instrument support personnel. If the nitrogen is supplied by a gas cylinder, record the tank pressure. Change the tank if less than 250 psi remaining.
C. Check the oil level of the vacuum pump.
D. Check the vacuum pressure under instrument status on the tune page. If a green circle with a white check mark in it is present, the system is ready.
E. Prime the UPLC system:
    1. Open the Acquity UPLC Console and select Acquity UPLC system in the menu on the left side of the screen.
    2. From the control drop down menu select ‘System Start Up.’
    3. Confirm for the sample manager (SM) that the strong wash, weak wash, and sample syringe are checked and 3 is entered in the cycles box.
    4. Confirm for the binary solvent manager (BSM) that all the boxes are checked and that the duration time is set to 5 minutes.
    5. Click the start button to start priming the system.
F. Analyzed the testmix:
    - General Chemistry, Seized Drugs
      If a column is installed, remove it from that system and replace it with a zero-dead-volume union. Perform an analysis of the appropriate testmix prior to the analysis of case samples. Use parameters listed in the 'Instrumental Conditions' section. Select the appropriate mobile phase. Start the UPLC pump. Ensure the ESI probe is engaged and turn on the MS. Start an acquisition using a filename such as 'TMyymmdd' (or equivalent). Make three 5 µL injections of the testmix solution at least 10 seconds apart by using the manual loop injector, and then stop the data collection. Evaluate the results using the 'Acceptance Criteria' section. If the results are acceptable, print the TIC, RICs, and spectra for components in the testmix.
    - Explosives
      Conduct a performance verification of the appropriate testmix through the column. Evaluate the results using the 'Acceptance Criteria' section. If the results are acceptable, print the TIC, RICs, and spectra for components in the testmix.
G. 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 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.

5.2.1 Orbitrap

A. Check the faceplate for debris, clean as needed.
B. Change the capillary.
C. Check level and color of rough pump oil. Top off or change oil as needed.
D. Bakeout the analyzer as needed.
E. Perform a full evaluation/calibration. If any steps do not pass, repeat. Perform a full calibration if warranted.

5.2.2 Liquid Chromatograph

A. Replaced solvent frits as needed.
B. Replace drain valve as needed.
C. Replace degasser pump as needed.
D. Inspect pump plungers, diaphragms and check valves. Replaced if needed.
E. Inspect oven fan.
F. Inspect degasser filters, replace if needed.
G. Check autosampler injection count and performance. Replace components (seals, rotors, stators, needle) if warranted.

5.3 Mass Calibration

The mass calibration procedure should be performed as needed based on system performance, when the instrument has been significantly moved, down for a long period of time, or undergone a major repair.

A. Load a 100, 250 or 500 µL syringe with either the LTQ Velos ESI Positive Ion Calibration Solution for Positive Mode, or Pierce ESI Negative Ion Calibration Solution for Negative Mode.
B. Connect the infusion syringe to the ESI probe assembly, and place in the syringe pump.
C. Set the syringe pump to the correct syringe type and set the pump rate to 5 µL/minute. The flow rate can be adjusted as needed.
D. Verify that the ESI heater is turned off.
E. Ensure a stable spray is achieved (approximately 2-5% variation)
F. On the tune page click "Calibrate" and confirm that MS Mass Calibration (pos) is checked for Positive Mode, or MS Mass Calibration (neg) for Negative Mode.
G. Turn on the syringe pump and verify that the solution is flowing out the ESI needle.
H. Ensure the ESI probe is engaged and turn on the MS.
I. Click the "Calibrate" button to start the calibration.

J. When the calibration is complete, it will display whether or not the calibration was successful.
    - If the procedure fails, repeat the calibration.
    - When the procedure passes, save the report and evaluate the calibration solution spectrum using the 'Acceptance Criteria' section.
    - If all requirements are within specification, prepare the documentation as outlined in IOSS-701. If any requirements fail, contact appropriate instrument support personnel.

6 INSTRUMENTAL CONDITIONS

6.1 Testmix (General Chemistry, Seized Drugs)

6.1.1 UPLC

Mobile Phase:	(From discipline-specific procedure)
Flow Rate:	0.15 mL/min
Column:	N/A
Injection Volume:	5 µL
Number of Injections:	3

6.1.2 Mass Spectrometer

Ionization:	ESI
Polarity:	Positive
Tune File:	testmix_pos (or equivalent)
Sheath Gas Flow:	9 (arb)
Aux Gas Flow:	3 (arb)
Sweep Gas Flow:	0 (arb)
Scan Mode:	Full Scan
Scan Range:	100–650 m/z
Resolution:	75,000

6.2 Testmix (Explosives)

6.2.1 UPLC

Mobile Phase:	(From discipline-specific procedure)
Flow Rate:	0.5 mL/min
Column:	Waters Cortecs UPLC C18
Injection Volume:	8 µL

6.2.2 Mass Spectrometer

Ionization:	ESI
Polarity:	Negative
Tune File:	exp_tune (or equivalent)
Sheath Gas Flow:	20 (arb)
Aux Gas Flow:	5 (arb)
Sweep Gas Flow:	0 (arb)
Scan Mode:	Full Scan
Scan Range:	200–400 m/z (minimum)
Resolution:	17,500

6.3 Mass Calibration

6.3.1 Mass Spectrometer

Ionization:	ESI
Polarity:	Positive or Negative (select as appropriate)
Scan Mode:	Full Scan
Scan Range:	100-2000 m/z

7 ACCEPTANCE CRITERIA

7.1 Testmix

The testmix components should be observed within the ranges list below from their expected monoisotopic masses.

7.1.1 General Chemistry, Seized Drugs

	Formula	Expected Mass	Acceptable Mass Range
Caffeine	$C_8H_{11}O_2N_4$	195.0877	195.0847 - 195.0907
Codeine	$C_{18}H_{22}O_3N$	300.1594	300.1564 - 300.1624
Brucine	$C_{23}H_{27}O_4N_2$	395.1965	395.1935 - 395.1995
Reserpine	$C_{33}H_{41}O_9N_2$	609.2807	609.2777 - 609.2837

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7.2 Mass Calibration

Verify the results of the calibration. The calibration will indicate if the procedure was successful. For reference, the individual ions for the calibration solution are listed below for each mode.

7.2.1 Positive Mode

Verify the results of the calibration. The calibration will indicate if the procedure was successful. For reference, the individual ions for the calibration solution are:

- Caffeine 195.0877 m/z
- MRFA 524.2650 m/z
- Ultramark 1022.0033 m/z
  1121.9969 m/z
  1221.9905 m/z
  1321.9841 m/z
  1421.9777 m/z
  1521.9713 m/z
  1621.9649 m/z
  1721.9585 m/z
  1821.9521 m/z

7.2.2 Negative Mode

- Sodium dodecyl sulfate 265.1479 m/z
- Sodium taurocholate 514.2844 m/z
- Ultramark 1279.9972 m/z
  1379.9908 m/z
  1479.9844 m/z
  1579.9781 m/z
  1679.9717 m/z
  1779.9653 m/z

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
03	10/04/2018	Section 1- Updated scope to include applicable disciplines/categories of testing. Section 4- Changed 'all' to 'stock solution' Section 8- Changed 'subunit' to 'discipline' mobile phase. Added '(minimum)' to scan range. Updated mobile phase and flow rate. Section 9- Reduced decimal places from five to four. Section 14- Updated to 'Instrument Operation and Systems Support' Added 'appropriate instrument support personnel' throughout.
04	09/30/2022	Revised to match new format requirements. Section 5- Expanded as-needed maintenance.
05	02/18/2025	Section 2 - Scope updated to remove toxicology. Section 4.1 – Removed toxicology. Section 5.2 – Removed check water chiller filters. Section 5.2 – merged with section 5.3, renumbered to 5.2. Section 5.4 – Changed calibration from print to save report. Section 6.1 – Removed toxicology. Section 7.1 – Removed toxicology. Sections 7.2.1 and 7.2.2 – Updated masses.