Achieving Peak Performance

For LTQ Velos, Velos Pro, LTQ Orbitrap Velos, Orbitrap Velos Pro, and Orbitrap Elite

This quick reference guide describes how to achieve peak performance from the Thermo Scientific™ LTQ Velos™, Velos Pro™, LTQ Orbitrap Velos™, Orbitrap Velos Pro™, and Orbitrap Elite™ mass spectrometers, including when to tune and calibrate, when and how to maintain the instrument, how to diagnose simple problems, and when it is necessary to contact your local field service engineer for assistance.

For additional information, refer to the *LTQ Series Getting Started Guide*, the *LTQ Series Hardware Manual*, the *Achieving Peak Performance Reference Manual*, and the Tune Plus application Help.

Contents

Stable Ionization Spray

Before you perform any tune, calibration, or diagnostic procedure, make sure that you establish a consistent, stable ionization spray. Always stabilize the spray before you begin, whether you infuse a sample or use a liquid chromatography (LC) system.

❖ To evaluate the spray stability

1. Attach the atmospheric pressure ionization (API) source housing to the mass spectrometer:
   - For the tuning procedure, install an electrospray ionization (ESI), heated-electrospray ionization (HESI), or nanospray ionization (NSI) probe.
   - For the calibration procedure, install an ESI or HESI probe.

   For instructions, refer to Chapter 2 in the *LTQ Series Getting Started Guide*.
2. Attach the syringe adapter from the Syringe Adapter Kit (P/N 70005-62011), which is part of the MS Accessory Kit, and then install a 250 µL or 500 µL syringe for direct infusion.
3. Open the Tune Plus application, choose **Setup > Syringe Pump**, and then set the flow control and syringe type.

Revision A 97655-97002

Thermo SCIENTIFIC

4. In the Tune Plus application, ensure that the following are at their optimum settings:
   - API gas flow rates (refer to Chapter 1 in the *LTQ Series Getting Started Guide*)
   - API probe position (refer to Chapter 2 in the *LTQ Series Getting Started Guide*)
   - Electrospray voltage—Use the following values.

ESI or HESI probe	EASY-Spray™ nanospray ion source	Nanospray Flex™ ion source or NSI-1 dynamic nanospray probe
4.5 kV	1.4–2.4 kV	1.5–2.5 kV

5. In the Tune Plus application, choose **Diagnostics > Diagnostics**, click **Tools**, select **System Evaluation**, and then run the **API Stability Evaluation** procedure.

   Note Because the API stability evaluation runs indefinitely, when you are ready to end the evaluation, click Stop and then click OK, which closes the dialog box.

   The API stability evaluation generates a real-time graph of the relative standard deviation (RSD) of the total ion current (TIC). When you change the parameters, such as the spray voltage, the RSD value and graph move up or down, depending on whether the change makes the spray more or less stable. The actual RSD value and its rating (for example, excellent or good) appear above the graph. The goal is a 15 percent or less RSD of the TIC.

Tuning for Peak Performance

Ensure that you periodically check the tune of the mass spectrometer to maintain peak performance over time. Proper tuning provides for longer periods between cleaning cycles and more stable, long-term performance.

Follow these procedures:

- To set the ion optic parameters
- To optimize the front lens

❖ To set the ion optic parameters

1. Attach the source housing to the mass spectrometer, and then install an ESI, HESI, or NSI probe, whichever is appropriate for data acquisition.

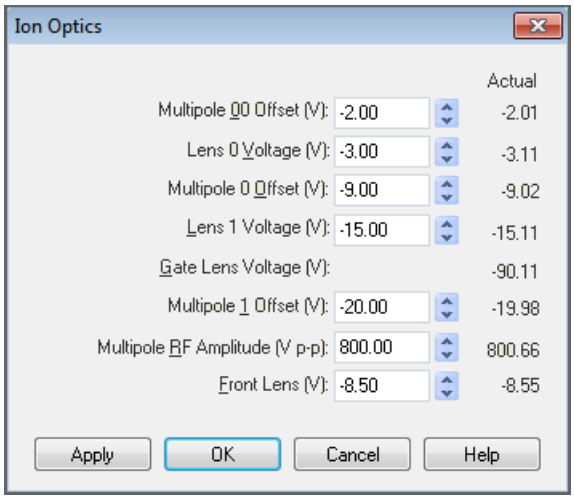
   For ESI and HESI modes, refer to Chapter 2 in the *LTQ Series Getting Started Guide*. For NSI mode, refer to the product documentation.

2. Establish a stable spray (see "Stable Ionization Spray" on page 1).

3. In the Tune Plus application, choose **Setup > Ion Optics** to open the Ion Optics dialog box (Figure 1).

   (For LTQ 2.7 SP1 or later only) The default ion optics values typically provide an optimum tune for both sensitivity and long-term instrument performance. Therefore, use the default values unless you determine that they are insufficient.

Figure 1. Ion Optics dialog box showing the default values



4. Ensure that the ion optic values are as shown in Figure 1. If they are not, manually type the values.

   Use negative voltages for positive ion mode and positive voltages for negative ion mode.

5. Click **OK**.

6. Click the **Save** button.

> **Tip** Save the tune method while the mass spectrometer is on. Because a tune file is source-type dependent, when you change the source type (for example, from HESI to NSI), you must repeat this tune procedure and save the tune file to another name.

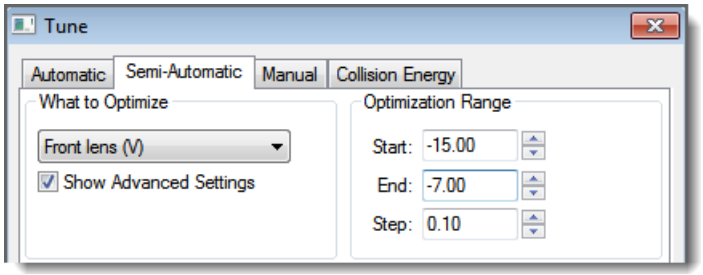
❖ To optimize the front lens

> **Note** Because the front lens has a sharp tuning curve, tune the lens.

1. In the Tune Plus application, click the **Tune** button to open the Tune dialog box, and then click the **Semi-Automatic** tab.

2. In the What to Optimize list, select **Front Lens (V)** (Figure 2).

Figure 2. Tune dialog box showing the Semi-Automatic page (partial)



3. Under Optimization Range, ensure that the Start, End, and Step values are as shown in Figure 2.

   The optimum front lens voltage is directly dependent on the multipole MP0 offset, which has a default value of –8.5 V. Typically, you set the front lens voltage to a value within a few volts of the MP0 voltage. For an MP0 offset of –8.5 V, the front lens voltage is usually in the range of –7.0 to –10.0 V.

4. Under What to Optimize On, do one of the following as applicable for the experiment:

   - Select the **Base Peak** option.

   –or–

   - Select the **Mass (*m/z*)** option, and then enter the appropriate mass of the analyte.

5. Click **Start**.

6. Click the **Save** button.

Tip Usually, a full automatic tune is unnecessary. However, you can run the automatic tune to ensure that the default values yield optimum results. If the automatic tune's performance results are within ±20 percent of the default values, Thermo Fisher Scientific recommends that you use the default values.

Calibration Schedule

You must periodically calibrate the mass spectrometers to maintain peak performance over time. Run all of the calibration checks once a week. Run the actual calibration for any calibration check that fails.

Make sure that you check the calibrations for the electron multipliers and the transfer lens because these calibrations change most often with usage. Check these calibration parameters in both positive and negative ion modes. Because the electron multipliers age more rapidly when they are new, to ensure proper gain, calibrate the new multipliers every 3 days for the first month or so of operation or until the multiplier voltage reaches the following upper limits:

- Approximately 1000 V for the LTQ Velos and LTQ Orbitrap Velos
- Approximately 1800 V for the Velos Pro, Orbitrap Velos Pro, and Orbitrap Elite

Instrument Maintenance

Peak performance requires periodic maintenance of the mass spectrometers. Thermo Fisher Scientific recommends the procedures in Table 1 as part of routine maintenance in between scheduled preventive maintenance (PM) visits from your Thermo Fisher Scientific field service engineer. Other tasks should be performed only if a performance issue arises with the instrument, as described in "Troubleshooting" on page 5.

If the signal intensity decreases gradually with time, check the calibration and tune, and follow the maintenance schedule in Table 1. If the signal intensity remains lower than expected, the ion optics might require cleaning. For instructions, refer to the *Achieving Peak Performance Reference Manual* and the *LTQ Series Hardware Manual*.

Table 1. Instrument components and recommended maintenance frequency (Sheet 1 of 2)

Component	Procedure	Frequency
Forepump	Open the forepump's ballast inlet.	30 min duration, once a week
	IMPORTANT To minimize the risk of oil contamination in the vacuum system, make sure that the purging ballast is closed when venting the system to atmosphere.	
	Change the oil.	Every 3 months or if the oil is cloudy or discolored

Table 1. Instrument components and recommended maintenance frequency (Sheet 2 of 2)

Component	Procedure	Frequency
ESI Probe	Trim the fused-silica sample tube, especially when you use acetonitrile as one of the chromatography solvents and when the polyimide coating on the tube has elongated past the end of the electrospray needle.	Weekly
HESI probe	Replace the needle insert for the metal sample tube if it becomes plugged or damaged.	Check weekly
NSI probe	Maintain the emitter per the manufacturer's instructions. Replace the emitter as needed.	Check the manufacturer's documentation
Ion transfer tube	Remove and clean the ion transfer tube.	Weekly or if the ion transfer tube bore is contaminated or obstructed

Trouble-shooting

Table 2 lists some problems that might occur with the mass spectrometer and their possible solutions.

Table 2. Troubleshooting solutions (Sheet 1 of 2)

Problem	Possible solutions
Oscillating signal intensity	• Follow the procedure "Stable Ionization Spray" on page 1. • If the problem persists, do the following: – Run the Transfer Efficiency Evaluation diagnostic. – If the evaluation fails, run the Transfer Lenses calibration for the appropriate ion mode. – If the calibration consistently fails, contact your local Thermo Fisher Scientific field service engineer for assistance.
Loss of signal intensity	• Make sure that the ion transfer tube is not restricted; clean or replace it if needed. • Check the tune. Make sure that the MP0–MP00 gradient is at least –5.5 V. • Check the signal by using direct infusion of the calibration solution. • Check the electron multiplier gain calibration. • Run the following charging diagnostics[a], and then clean the ion optics as indicated by the results: – Multipole Gradient Evaluation – Multipole MP0 Flight Time Evaluation – Source Optics Flight Time Evaluation – Ion Optics Charging Evaluation (run last) • If the problem persists, check the LC system.

Table 2. Troubleshooting solutions (Sheet 2 of 2)

Problem	Possible solutions
Complete loss of signal	• In the Tune Plus application, check the Status View for any faults. If necessary, run the appropriate diagnostics tests. • Make sure that the ion transfer tube is not restricted; clean or replace it if needed. • Check the source spray conditions by using direct infusion of the calibration solution. • Open a different tune file. It is possible that the current tune file has become corrupt. • Press the reset button on the mass spectrometer (refer to Chapter 3 in the *LTQ Series Hardware Manual*). • If the problem persists, check the LC system.
Failure of the electron multiplier gain calibration—the signal is too weak	• Make sure that the spray is stable (see page 1). • Make sure that the calibration mixture (Calmix) is fresh and that you have the correct solution for either the stand-alone or Orbitrap instrument (www.thermo.com/pierce). – Stand-alone: Pierce™ LTQ Velos ESI Positive Ion Calibration Solution (P/N 88323) – Orbitrap: Pierce ESI Negative Ion Calibration Solution (P/N 88324) • Manually set the electron multiplier gain to a higher voltage through the Diagnostics dialog box, and then repeat the calibration. • If the gain is above 2500 V, contact your local Thermo Fisher Scientific field service engineer to replace the electron multipliers.
Failure of the transfer lenses calibration	• Repeat the transfer lenses calibration in the mode that failed. • If the problem persists, contact your local Thermo Fisher Scientific field service engineer for assistance.
Failure of the ejection and multiplier gain evaluation	• Make sure that the spray is stable (see page 1). • Repeat the electron multiplier gain calibration. • If the problem persists, contact your local Thermo Fisher Scientific field service engineer for assistance.

[a] Available with LTQ 2.7 SP1 or later

Trademarks