Q Exactive GC

Mass Spectrometer

Software Manual

1R120706-0003 Revision B December 2015

Thermo SCIENTIFIC

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Release History: Revision A released in June 2015; Revision B released in December 2015.

Software Version: Q Exactive GC Tune 2.7

For Research Use Only. Not for use in diagnostic procedures.

Contents

Read This First

Welcome to the Thermo Scientific™ Q Exactive™ GC system!
Q Exactive GC is a member of the Thermo Scientific family of mass spectrometer (MS) detectors that are powered by Orbitrap™ technology.

About This Guide

This *Q Exactive GC Software Manual* provides reference information about the parameters in the Instrument Configuration window, the Instrument Setup window, and the Q Exactive GC Tune window. For information about the operating procedures for the Q Exactive GC mass spectrometer, we recommend that you read the *Q Exactive GC Operating Manual* in its entirety.

Who Uses This Guide

This *Q Exactive GC Software Manual* is intended for all personnel that need to perform measurements with the Q Exactive GC mass spectrometer, especially the key operator. This manual should be kept near the instrument to be available for quick reference.

Scope of This Guide

The *Q Exactive GC Software Manual* includes the following chapters:

- Chapter 1: "Instrument Configuration Window" describes the Instrument Configuration window, which displays configuration information.
- Chapter 2: "Q Exactive GC Readback Status Page for the Information View" describes the page that allows controlling the instrument from the Roadmap view of the Xcalibur™ Home Page window.
- Chapter 3: "Instrument Setup" describes Instrument Setup, which is used to specify instrument settings.
- Chapter 4: "Explore Q Exactive GC Tune" provides information about Q Exactive GC Tune, its views, functions, and features.
- Chapter 5: "Procedures in Q Exactive GC Tune" provides information about procedures in Q Exactive GC Tune.

- Chapter 6: "Reference Information" provides information about various file types and other supplemental information.

Related Documentation

In addition to this guide, Thermo Fisher Scientific provides the following documents for the Q Exactive GC mass spectrometer:

- *Q Exactive GC Preinstallation Requirements Guide*
- *Q Exactive GC Quick Start Guide*
- *Q Exactive GC Operating Manual*
- Manuals for the delivered ion sources and other software.

You can access PDF files of the documents listed above and of this guide from the data system computer. The Q Exactive GC Tune software also provides Help.

❖ **To view product manuals**

Go to **Start > Programs > Thermo Exactive Series > Manuals**.

❖ **To open Help**

- From the Q Exactive GC Tune window, choose **Help > Help Content**.
- If available for a specific window or dialog box, click **Help** or press **F1** for information about setting parameters.

For more information, including upcoming application notes, visit www.thermoscientific.com.

Contacting Us

There are several ways to contact Thermo Fisher Scientific.

Assistance

For technical support and ordering information, **visit us on the Web**:

www.gc-gcms-customersupport.com/WebPage/Share/Default.aspx

Service contact details are available under:

www.unitylabservice.com

For brochures, application notes and other material, please visit:

www.thermoscientific.com

For operating manuals, please contact your local Service organization.

Changes to the Manual

❖ **To suggest changes to this manual**

- Please send your comments to:

  Technical Documentation
  Thermo Fisher Scientific
  2215 Grand Avenue Parkway

  Austin, Texas 78728

  United States

- Send an e-mail message to the Technical Publications Editor at techpubs-austin@thermofisher.com.

You are encouraged to report errors or omissions in the text or index. Thank you.

Typographical Conventions

This section describes typographical conventions that have been established for Thermo Fisher Scientific manuals.

Signal Word

Make sure you follow the precautionary statements presented in this manual. The special notices appear different from the main flow of text:

NOTICE Points out possible material damage and other important information in connection with the instrument. ▲

Data Input

Throughout this manual, the following conventions indicate data input and output via the computer:

- Messages displayed on the screen are represented by capitalizing the initial letter of each word and by italicizing each word.
- Input that you enter by keyboard is identified by quotation marks: single quotes for single characters, double quotes for strings.
- For brevity, expressions such as "choose **File > Directories**" are used rather than "pull down the File menu and choose Directories."
- Any command enclosed in angle brackets < > represents a single keystroke. For example, "press **<F1>**" means press the key labeled *F1*.
- Any command that requires pressing two or more keys simultaneously is shown with a plus sign connecting the keys. For example, "press **<Shift>** + **<F1>**" means press and hold the <Shift> key and then press the <F1> key.
- Any button that you click on the screen is represented in bold face letters. For example, "click **Close**".

Topic Headings

The following headings are used to show the organization of topics within a chapter:

Chapter 1 Chapter Name

Second Level Topics

Third Level Topics

Fourth Level Topics

Chapter 1 Instrument Configuration Window

NOTICE **Never change the settings in this window except during installation of the instrument.** Any changes in this window affect the data acquisition or the functionality of your instrument. A later change will rarely solve problems users have to face. ▲

The Instrument Configuration window displays Q Exactive GC configuration information. See Figure 1-1.

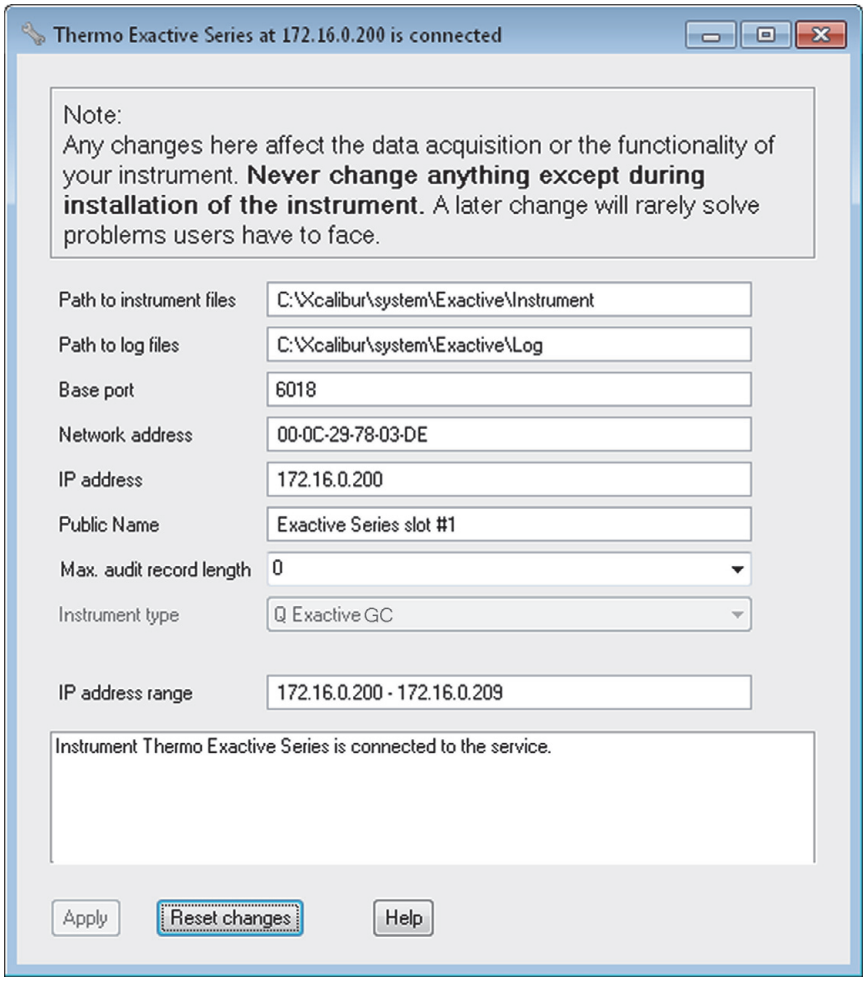


Figure 1-1. Instrument Configuration window

The Instrument Configuration window has the following parameters:

Parameter	Description
Path to instrument files	Displays the location of the instrument files. To change the path, click the folder button on the right side of the text box. A dialog box appears where you can select another location.
Path to log files	Displays the location of the log files. For information about the content of the log files, see "Log Files" on page 6-2. To change the path, click the folder button on the right side of the text box. A dialog box appears where you can select another location
Base port	Displays the first TCP/UDP port in use for this particular instrument. Leave the field empty for an automatic assignment.
Network address	Displays the hardware address of the network card installed in the instrument. Leave the field empty for an automatic assignment.
IP address	Displays the IP address. Leave the field empty for an automatic assignment.
Public Name	Displays the name that is shown in the About dialog box of Q Exactive GC Tune.
Max. audit record length	Use this list box to select the maximum audit record length. The following options are available: • **0** disables auditing • **50** is common for an Access audit database initialized by Platform's Database Configuration • **255** is common for an Oracle audit database initialized by Platform's Database Configuration • **1000** is common if auditing is done by Chromeleon. It is a compromise between size and readability
Instrument type	Displays the configured instrument type, that is Q Exactive GC.
IP address range	Displays the IP address or IP address range which should be assigned to the instrument during its starting phase. Use the numeric form, for example "172.16.2.1-172.16.2.15". **NOTICE** Change only when advised by your network administrator. ▲
Message box	Displays information about the instrument and connection status.
Buttons	
Apply	Saves your changes in this window.
Reset changes	Discards your changes in this window.
Help	Displays the Help for this window.

❖ **To display this window**

1. Choose **Programs > Thermo Foundation 3.1 > Instrument Configuration**.

2. From the Instrument Configuration window, click Q Exactive GC - Orbitrap MS in the Configured Devices area.
3. Click **Configure**.

Chapter 2 Q Exactive GC Readback Status Page for the Information View

The readback status of each Xcalibur-configured instrument appears on the Status page of the Information view. When you click an instrument, Xcalibur™ displays current readings for the instrument on a page below the Run Manager pane. See Figure 2-1.

Right-click any of the instruments to display a shortcut menu where you can switch your instrument to On, Off, or Standby mode.

NOTICE This view is normally displayed on the left side of the Home Page window. If this view is not displayed, the view has been turned off. ▲

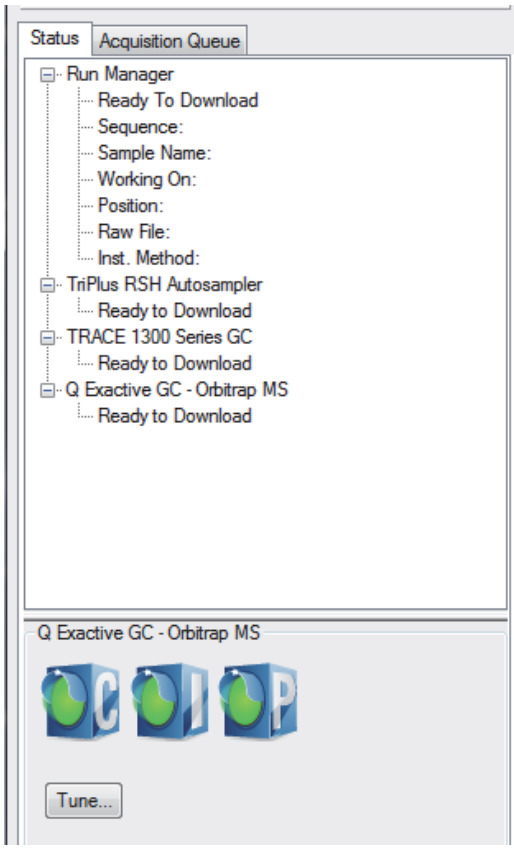


Figure 2-1. Q Exactive GC Readback Status Page for the Information View

The following functions are available:

Button		Description
	Communication Status	Shows the actual communication status of the system: • Green: communication with instrument is ok. • Yellow: only service is accessible (no instrument). • Red: communication is broken (no instrument, no service).
	Instrument Status	Shows the actual hardware status of the system (top instrument tree state): • Green: all readbacks are in specifications (green hooks). • Red: one or more readbacks are out of range.
	Performance Status	Shows the actual performance status of the system: • Green: the last evaluation/calibration was successful. • Yellow: last evaluation/calibration was successful, but is out of date. • Red: the evaluation/calibration was not successful.
Tune		Opens the Q Exactive GC Tune window.

❖ **To open this page**

1. From the Home Page window, choose **View > Info View** to display the Information view.
2. Click the **Status** tab.
3. Click **Q Exactive GC - Orbitrap MS**.

Chapter 3 Instrument Setup

After you have selected in the Instrument Configuration program which instruments you want Xcalibur to control, use Instrument Setup to specify your instrument settings.

Contents

Instrument Setup Window

The Instrument Setup window displays the icons of the instruments that you have selected using the Instrument Configuration window. (See the View bar on the left side of the window.) See Figure 3-1. If you have configured more instruments than can be displayed on your screen, a vertical scroll bar appears in the View bar. So, you can access all of the instruments.

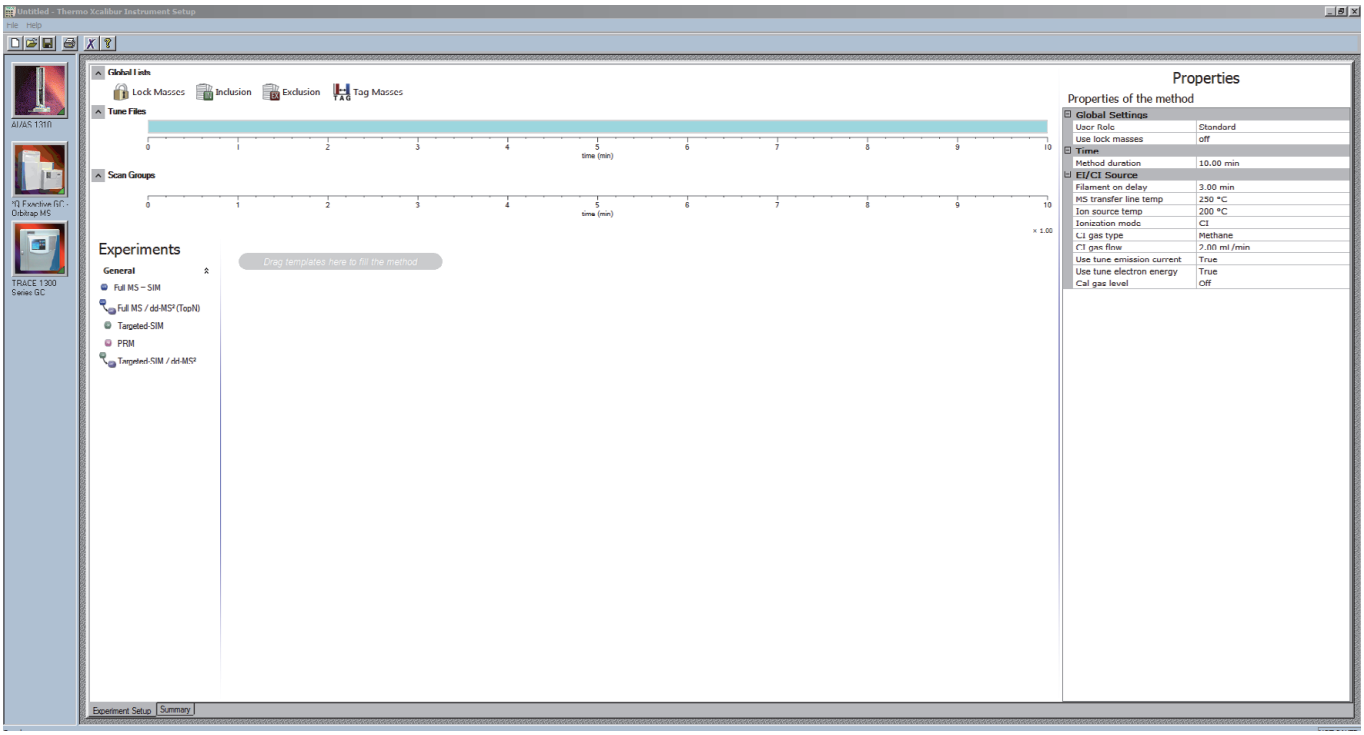


Figure 3-1. Instrument Setup window

To enter the setup parameters for a particular instrument, click the icon for that instrument. Xcalibur displays one or more pages of parameters to be set for the one you selected.

NOTICE Before using the Instrument Setup window, use the Instrument Configuration program to select the instruments to be used for your experiment. ▲

Use Instrument Setup to specify settings for your instruments after you have selected with the Instrument Configuration program the instruments that you want Xcalibur to control. The Instrument Setup window displays the setup parameters required for each instrument that you select on the View bar. These might include your autosampler, GC, mass spectrometer, and/or all other Xcalibur supported instruments that you have configured.

You can create new methods, modify existing methods, and save method files.

❖ **To display this window**

- Click  from the Roadmap view of the Home Page window.
- Alternatively, choose **GoTo > Instrument Setup**.

View Bar

The View bar is a vertical bar on the left of the Instrument Setup window. It contains buttons for each of the instruments that you have selected by using the Instrument Configuration program.

Menus

Instrument Setup contains the following menus:

- File Menu for the Instrument Setup Window
- Help Menu for the Instrument Setup Window

Toolbar

Instrument Setup Window Toolbar

File Menu for the Instrument Setup Window

The File menu provides commands for file and program operations. It has the following commands:

	Command	Description
	New	Create a new method file having the appropriate extension.
	Open	Find and open a file that already exists.
	Save	Save the active method. Changes will be recorded in Audit Trail after the method is saved.
	Save As	Edit user and description information, and view header information about the active file. Xcalibur opens the Save As dialog box. Changes will be recorded in Audit Trail after the method is saved.

	Command	Description
	Summary Information	Edit user and description information, and view header information about the active file.
	Change Study Name	Select a dataset from a predefined list of names. The text of this menu item might be different if the administrator chose to use another name for a dataset. For example, this menu item might be Change Job Name.
	Audit Trail	View all auditable events and changes made to data files in the current application.
	Print	Print the parameters in your instrument method.
	Print Preview	View your page setup so that you can see what it looks like before printing it.
	Print Setup	Select the following printing options: printer, form, orientation, and one-sided or two-sided printing.
	Most Recently Used Files	View the paths and names of the last four files used. These are located above the **Exit** command. Both open and closed files are displayed. Click a displayed file to load it. If the selected file was closed, it will be opened.
	Exit	Close the active window. If you exit before clicking **OK** from an active dialog box, Xcalibur asks if you want to save your changes.

Help Menu for the Instrument Setup Window

The Help menu lists the following commands:

Command	Description
Q Exactive GC - Orbitrap MS Help	Open the Configuration and Setup Help for the instrument.
Instrument Setup Help	Open Xcalibur Help and display Help for the Instrument Setup window.
Help On Current Item	View Help for the Instrument Setup page that is currently displayed.
Q Exactive GC - Orbitrap MS Contents and Index	View the Contents, Index, and Find Help pages for the selected instrument.
Xcalibur Help	Open Xcalibur Help.
How to use Online Help	Open Help that describes how to use the Help viewer.
About Instrument Setup	View the version number of the Instrument Setup program and the Thermo Fisher Scientific copyright notice.

Instrument Setup Window Toolbar

The toolbar provides symbol shortcuts for frequently used commands. The following functions are available:

	Button	Description
	New	Create a new instrument method.
	Open	Find and open an existing file.
	Save	If your method has not been saved before, clicking **Save** opens the Save As dialog box. Select the name and location for your instrument method. When you click **Save**, the File Summary Information Dialog Box opens. Enter header information for your instrument method. The file is saved when you click **OK**.
	Print	Print the parameters in your instrument method.
	Home Page	View the Home Page window—Roadmap view.
	Help	View Help for the Instrument Setup page that is currently displayed.

Method Editor

Use the Method Editor to specify the type of mass spectrometer experiment you plan to perform. See Figure 3-2.

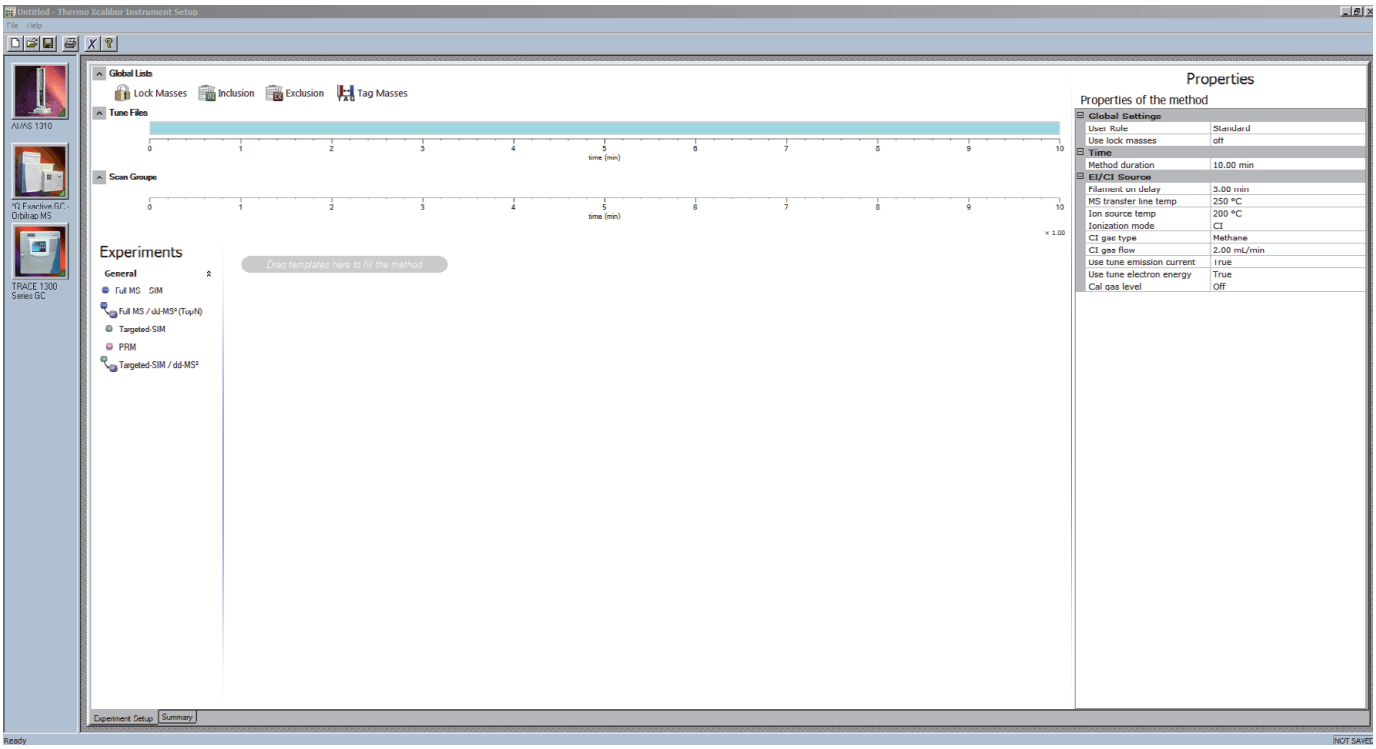


Figure 3-2. Method Editor (Xcalibur)

The Method Editor comprises the following pages:

- Experiment Setup Page
- Summary Page

❖ **To display this view**

Click **Instrument Setup** from the Roadmap View of the Home Page window or choose **GoTo > Instrument Setup**.

Experiment Setup Page

Use the Experiment Setup page to set up Q Exactive GC mass spectrometer experiments. You use this page to specify values for the Q Exactive GC parameters and save the parameters in an instrument method. See Figure 3-3.

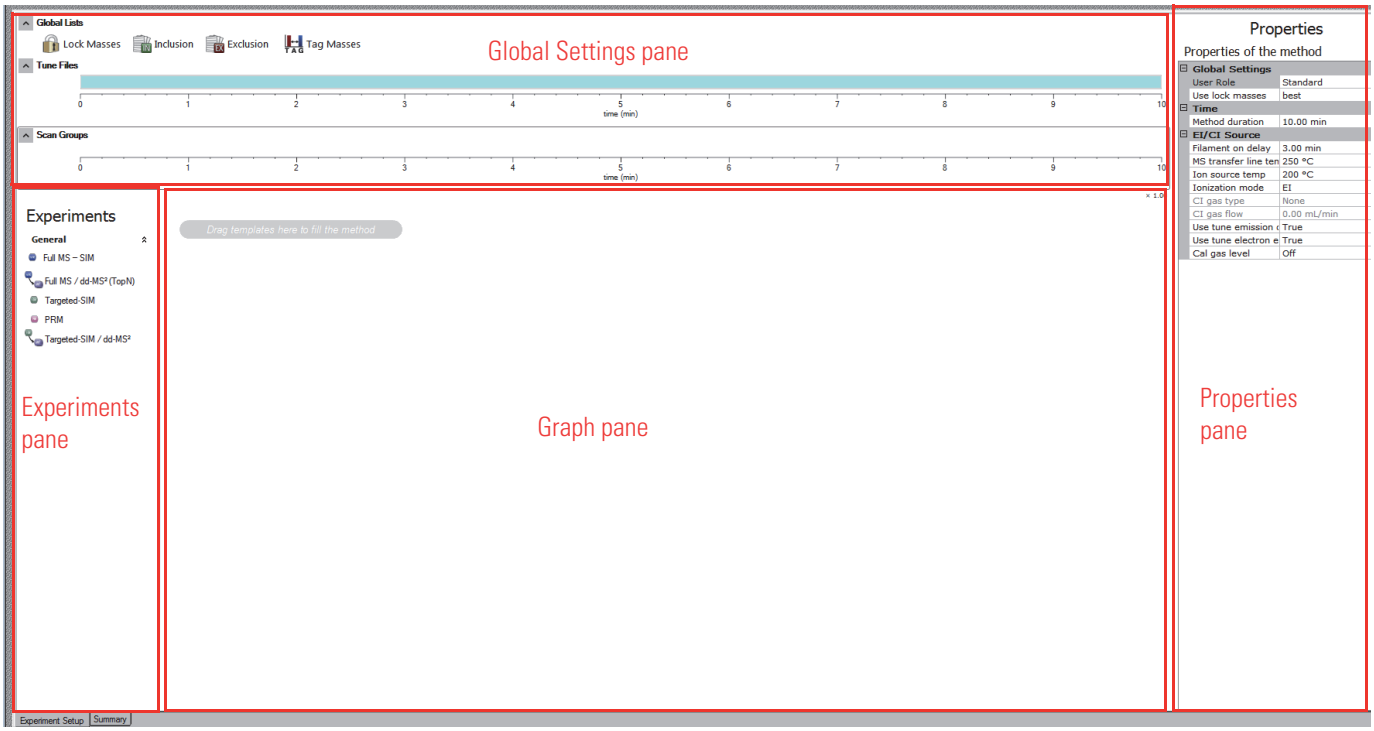


Figure 3-3. Method Editor—Experiment Setup page

The Experiment Setup page comprises four major windows:

- Global Settings Pane
- Experiments Pane
- Graph Pane
- Properties Pane

Global Settings Pane

The Global Settings pane is located on the top left side of the Experiment Setup page. It gives access to global lists, tune files, external hardware, chromatogram, and scan groups. See Figure 3-4.

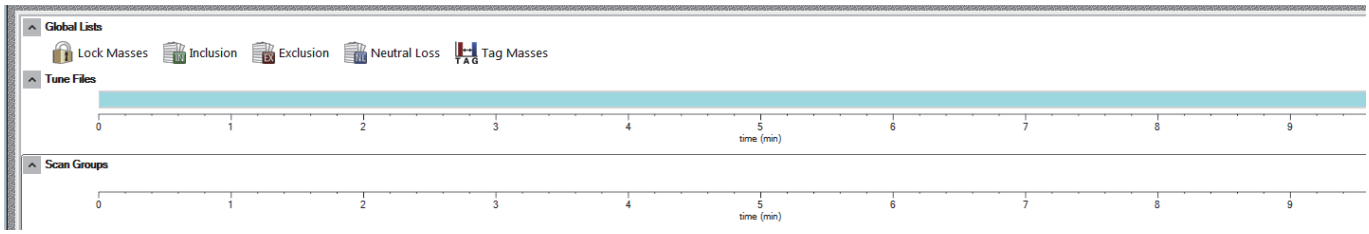


Figure 3-4. Method Editor—Global Settings pane

The Global Settings pane contains the following panes:

- Global Lists
- Tune Files
- Right-click in the pane to display the shortcut menu. Choose **Unzoom**.Scan Groups

Click the title bar of an individual pane to display it. Click again to hide it.

Global Lists

Use the Global Lists pane of the Experiment Setup page to perform intelligent automated MS/MS and SIM type experiments by efficiently utilizing the mass spectrometer to characterize parent ions independent of the "current segment" model. Use the Global Lists pane to specify parent, reject, and/or non-data dependent MS/MS and SIM masses with customizable and independent time windows. See Figure 3-5. Scan events that reference the mass lists that you create here run only if there is a corresponding mass/time window at that particular retention time.

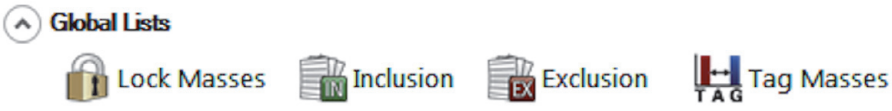


Figure 3-5. Method Editor—Global Lists pane

The following dialog boxes are available:

- Lock Masses Dialog Box
- Inclusion List Dialog Box
- Exclusion List Dialog Box

- Tag Masses Dialog Box

To display a dialog box, click the respective icon.

Tune Files

The Tune Files pane shows a graphical representation of the tune files usage during the various phases of the experiment. See Figure 3-6. In addition to the base tune file, you can specify up to 50 other tune files.

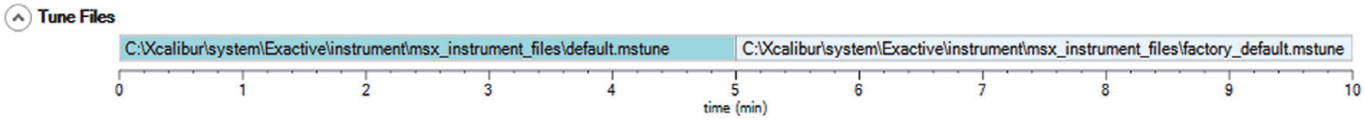


Figure 3-6. Method Editor—Tune Files

❖ **To assign tune files**

1. Do one of the following:

   - Point to a position on the time bar and right-click. Choose **Change to another tunefile at *<time at pointer position>*** in the shortcut menu.

   - Change the value of the Switch Count parameter in the Properties of Tunefiles pane.

2. In the Properties of Tunefiles pane, click the button next to the respective box of the Base Tunefile or New Tunefile parameter and browse for the new tune method file. See Figure 3-7. Select the tune files and assign them to the experiment phases.

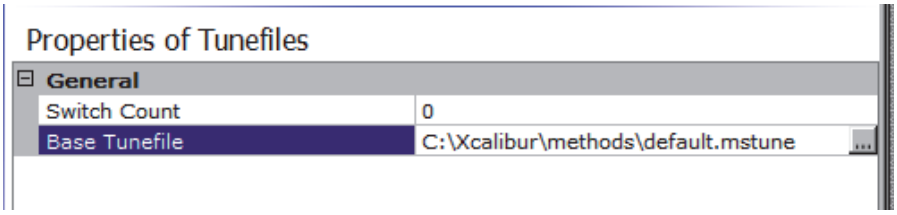


Figure 3-7. Properties of Tunefiles

The name and path way of the tune files will be shown on the respective time bar on the Tune Files pane.

❖ **To change the starting time for tune files**

- Position the cursor between adjacent tune files and drag the scan event along the time bar.

- In the Properties of Tunefiles pane, enter the time in the box of the At parameter of the respective Element.

❖ **To remove a tune file**

1. Position the cursor between adjacent tune files
2. Right-click to display the shortcut menu and choose **Remove change to tunefile at <*time at pointer position*>**. The tune file to the right is removed.

❖ **To zoom on the Tune Files pane**

- Position the mouse pointer within the time axis area and drag the mouse across the time area of interest.
- To zoom in, position the mouse pointer within the pane and roll the mouse wheel forward.
- To zoom out, roll the wheel backward.
- To increase the zooming factor by two, keep the **<Shift>** key pressed while using the mouse wheel.

❖ **To unzoom the Tune Files pane**

Right-click in the pane to display the shortcut menu. Choose **Unzoom**.**Scan Groups**

In the Scan Groups pane, time bars represent the scan events during the acquisition. See Figure 3-8. The Scan Groups pane is filled when you drag experiment symbols from the experiments pane to the Graph pane.

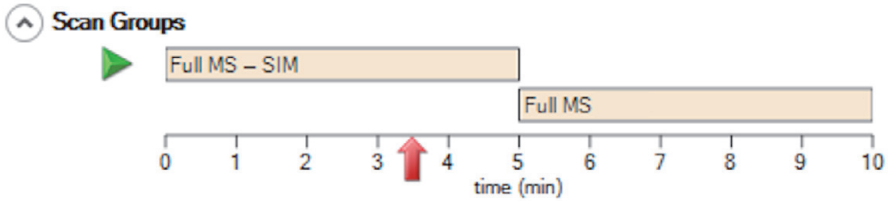


Figure 3-8. Scan Groups

Use the Properties pane of a scan event to control the activities during individual experiments. Red triangles ( ) to the left side of a time bar indicate active scan events.

❖ **To display the active scan events at a certain time**

Click the time line at the position. A red arrow ( ) indicates the selected time. A green triangle ( ) to the left side of the time bar indicates the active scan event. If scan events overlap at the selected time, green triangles appear for each active scan event. The bottom part of the Properties pane shows the parameters for the selected scan event.

❖ **To change start and end time of a scan event**

- Drag the left and right edges of the time bar to the desired positions on the time line.
- Enter the times in the Minimum and Maximum boxes of the runtime parameter on the Properties pane.

❖ **To zoom on the Scan Groups pane**

- Position the mouse pointer within the time axis area and drag the mouse across the time area of interest.
- To zoom in, position the mouse pointer within the pane and roll the mouse wheel forward.
- To zoom out, roll the wheel backward.
- To increase the zooming factor by two, keep the **<Shift>** key pressed while using the mouse wheel.

❖ **To unzoom the Scan Groups pane**

Right-click in the pane to display the shortcut menu. Choose **Unzoom**.

Experiments Pane

The Experiments pane is located on the bottom left side of the Experiment Setup page. It gives access to predefined experiment templates. You cannot delete the experiment templates.

Every single experiment is associated with a descriptive icon. This icon can be dragged from the Experiments pane to the Graph pane. When selected, the icon gives an overview to the operator, which experiment and which associated options are active currently. Therefore, this icon changes dependent on the settings. Use the Properties pane to specify the values for your experiment parameters.

General Experiment Templates

The General folder contains predefined templates for experiments. See Figure 3-9.

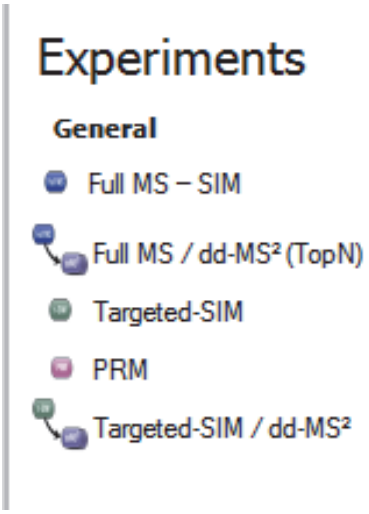
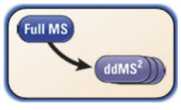


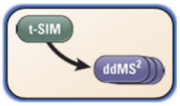
Figure 3-9. Available general templates

In Xcalibur, the Experiment Setup page allows using several system templates in an instrument method.

Experiment Symbols

The following experiment templates are available on the experiments pane:

Template	Experiment Symbol	Description
General Experiment Templates		
Full MS – SIM		This experiment comprises a full MS scan or SIM scan without HCD fragmentation. Special features: • Segmented Master Scans
Full MS / dd-MS^2 (TopN)		This experiment comprises a full MS scan (without collision energy) followed by a set of Data Dependent™ scans with a fragmentation energy applied. Ions of the second scan event enter the HCD collision cell, ions of the first do not. Special features: • Auto value option: Dynamic Exclusion • Segmented Master Scans • Spectral Multiplexing (MSX) • Stepped Collision Energy

Template	Experiment Symbol	Description
Targeted-SIM		This experiment acquires SIM scans depending on the entries of the inclusion list, which is therefore mandatory and always activated. Special features: • Spectral Multiplexing (MSX)
PRM		This Parallel Reaction Monitoring (PRM) experiment comprises MS/MS scans depending on the entries of the inclusion list. The mandatory inclusion list will be processed from first to last row without any automated sorting. Special features: • Spectral Multiplexing (MSX) • Stepped Collision Energy
Targeted-SIM / dd-MS^2		This experiment comprises a targeted-SIM scan on precursor ions followed by a set of Data Dependent triggered MS/MS scans. Special features: • Auto value option: Dynamic Exclusion • Spectral Multiplexing (MSX) • Stepped Collision Energy

Graph Pane

The Graph pane is located at the middle bottom side of the Experiment Setup window. It shows a graphic representation of the experiment during the acquisition. See Figure 3-10.

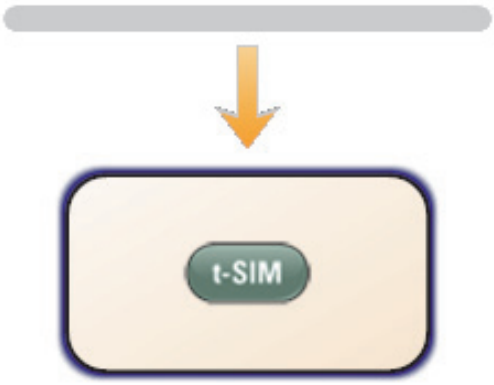


Figure 3-10. Graphic representation of the experiment

❖ **To add an experiment to the active method**

1. Drag an experiment symbol from the experiments pane to the gray bar in the Graph pane.

   In Xcalibur, the Scan Groups pane shows a corresponding time bar for each symbol.

2. Use the Properties pane of a experiment to control the activities during individual experiments.

❖ **To delete an experiment**

1. Right-click the experiment symbol to display the shortcut menu.
2. Choose **Delete this <Name of experiment>**.

❖ **To zoom in or out on the Graph pane**

- Position the mouse pointer within the pane and roll the mouse wheel forward to zoom in.
- Roll the wheel backward to zoom out.

The zoom factor ranges from 0.25 to 2. The actual zoom factor is displayed in the top right corner of the Graph pane.

Properties Pane

The Properties pane is located at the right side of the Experiment Setup page. See Figure 3-11.

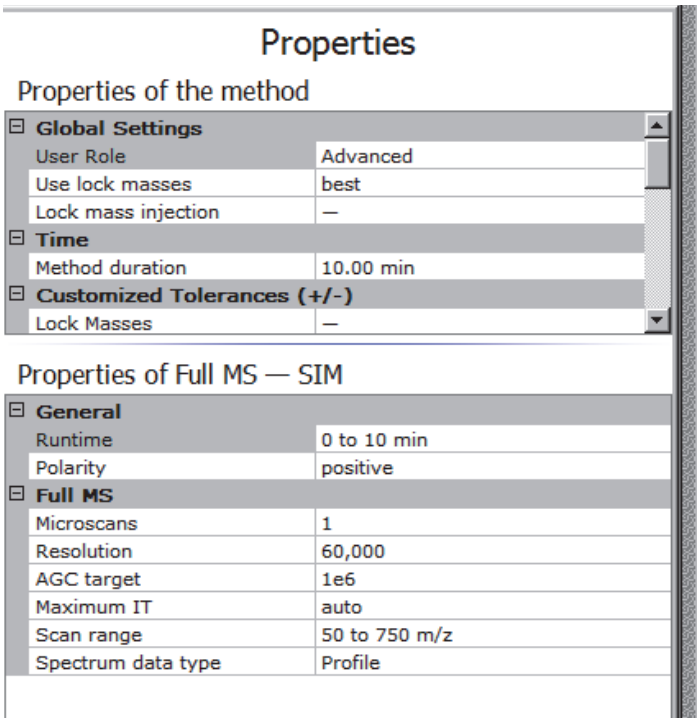


Figure 3-11. Properties pane

The pane consists of two parts:

- Properties of the Method

  The upper part is always available. It shows the properties of the active method.

NOTICE Some properties are only available in Advanced user mode.

The lower part depends on the item that is selected on the left side of the Experiment Setup page. One of the following tables is displayed:

In Xcalibur:

- Properties of Full MS – SIM
- Properties of Full MS / dd-MS^2 (TopN)
- Properties of Targeted-SIM
- Properties of PRM
- Properties of Targeted-SIM / dd-MS^2

Properties of the Method

The properties of the method include the following parameters:

Parameter	Description
Global Settings	
User Role	Use this field to specify whether the advanced parameters for scan events are displayed or not. To show the advanced parameters, set the field to **Advanced**. To hide the advanced parameters, set the field to **Standard**. Double-click into the field to change the status.
Use lock masses	Use the list box to specify how lock masses are used in the active instrument method. The following options are available: • off No lock masses are used. • best Only the most intense lock mass of the global list that is found in the spectrum is used for a calibration. • if all present Lock masses are used only when all active masses of the global lock mass list are present at the same time. **NOTICE** If timed lock masses are used, the Q Exactive GC mass spectrometer takes into account only those lock masses whose time windows cover the current retention time. ▲
Lock mass injection	If lock masses are used in the active instrument method, use this field to select the scan types for which lock mass will be additionally injected. The mass spectrometer will inject all lock masses of the active lock mass list to provide additional signals to improve mass accuracy. The decision whether found lock masses will be used for the mass correction is not affected and depends on the settings for the “use lock masses” parameter. Enter the scan types manually by using commas as separators for the strings. Alternatively, click the down arrow to display a dialog box and select the corresponding check boxes. The following options are available: • Full MS • MS^2 • SIM
Time	
Method duration	Use this box to specify the total mass spectrometer acquire time, in minutes, for the run. Q Exactive GC Tune rescales the Segments bar to correspond to the specified acquire time. To change the time, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter the time in the spin box text field. The valid range for the time is from 0.1 to 10000 minutes.
Customized Tolerances (+/-)	

Parameter	Description
Lock Masses Inclusion Exclusion Mass Tags Dynamic Exclusion	Use the boxes in this group to adjust mass tolerances for global lists in different instrument methods. The tolerance can be set independently for each list type. To change a tolerance, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter the tolerance in the spin box text field. Available options are: • – (use instrument default) • 0.1–1000 ppm • 0.1–50000 amu When you directly enter a tolerance in the text field, you can use additional units. The software will convert it to ppm or amu depending of its nature (absolute or relative definition). Accepted units are (for absolute definition: Da, Th, u, amu; for a relative definition: ppt, ppb, ppm, %). An empty property value field as well as a set value of **0**, **-**, or **off** will result in using the instrument default (displayed as –).

Parameter	Description
EI/CI Source	
Filament on Delay MS Transfer Line Temp Ion Source Temp Ionization Mode CI Gas Type CI Gas Flow Use Tune Emission Current Emission Current Use Tune Electron Energy Cal Gas Level	Use the boxes in this group to adjust the EI or CI ion source settings for global lists in different instrument methods. • Filament on Delay: Set the initial time delay to account for the solvent front. The delay will begin after the run starts (contact closure) before turning on the filament. • MS Transfer Line Temp: Set the temperature for the part of the transfer line that is inside the vacuum manifold. Temperatures from 200 – 280 °C are recommend for most applications. • Ion Source Temp: This allows the user to set the ion source temperature. Temperatures from 200 – 350 °C are recommend for most applications. • Ionization Mode: Set to EI or CI ionization mode depending on analysis. The mode must be set to CI to turn on CI gas. **NOTICE** The CI or EI/CI ion volume must be installed in the ion source cartridge to use CI ionization mode. ▲ • CI Gas Type: Select the type of CI gas to use for the analysis. Selection options are methane, isobutane, ammonia, carbon dioxide, or other. Configure the selected gas on Port A or Port B within Tune. • CI Gas Flow: Set the CI gas flow rate. • Use Tune Emission Current: Select True to set the filament emission current to the value saved in the tune file during the acquisition. Select False to set the emission current in the method and override the value in the tune file. • Emission Current (Only visible if “Use Tune Emission Current” is set to “False”): Set the emission current to be used during acquisition instead of the emission current in the tune file. • Use Tune Electron Energy: Select True to set the filament voltage to the value saved in the tune file during the acquisition. Select False to set the filament voltage in the method and override the value in the tune file during acquisition. When setting low electron energies, the electron lens may also need to be set to a new value in the tune file. • Cal Gas Level: Set the calibration gas to the EI or the higher CI level during acquisition.

NOTICE In some cases the instrument will not allow the filament to be turned on to protect against damage. This can happen when the electron energy is set low and the read value for the filament voltage has not returned a negative voltage, or the electron lens is not at least 45 V more positive than the filament. In these cases, make sure the filament and electron lens are at the appropriate values in Tune before starting an acquisition. Also notice that in some cases, when the electron energy is set low, the instrument must override the user value in the tune file for the electron lens in order to meet the required conditions to protect the filament. The filament voltage is calculated by the instrument from the ion source offset and the electron energy settings. ▲

Properties of Full MS – SIM

The properties of a Full MS – SIM experiment include the following parameters:

Parameter	Description
General	
Runtime	Use this box to specify the duration in minutes of the active scan event. Click into the field to display the spin boxes for the start time (Minimum) and the end time (Maximum) of the scan event. To change a value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range for the start time is from 0 to (end time minus 0.01) minutes. The valid range for the end time is from (start time plus 0.01) to 10000 minutes. Alternatively, drag the left and right edges of the corresponding time bar to the desired positions on the time line in the Scan Groups pane.
Polarity	Use this list box to toggle between positive ion and negative ion polarity.
Full MS – SIM	
Use the parameters in this group to specify the properties of the master scan(s).	
Microscans	Use this spin box to select the number of microscans to be performed. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 to 10 microscans.
Resolution	Use this list box to select the mass resolution used during the selected scan event. The mass resolution of the Orbitrap analyzer is proportional to 1/sqrt (*m/z*). The scan time increases with increasing resolution and detect time. Available options are 15000, 30000, 60000, or 120000. Resolution is calculated according to the IUPAC peak width definition. The available settings represent m/Δm or the *m/z* value of an ion at *m/z* 200 divided by the full width at half its maximum height (FWHM).

Parameter	Description
AGC target	Use this list box to select the AGC target value for the selected scan event. The AGC target value controls the number of ions that are injected into the Orbitrap analyzer. Available options are 2e4, 5e4, 1e5, 2e5, 5e5, 1e6, 3e6, or 5e6.
Maximum IT	Use this list box to type or click a maximum allowed injection time to reach the AGC target value. For example, type **160**. The valid range is from 1 to 3000 milliseconds.
Number of scan ranges	Use this spin box to enter the number of segments in a segmented master scan. The segments can be defined independently from each other. They can be used to get a better S/N for specific areas and exclude matrix signals. The m/z ranges are acquired one after another and will result one scan for each range. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. The valid range is from 1 to 8 scans.
Scan range *n* (*n*=1–8)	Use the spin boxes in this field to set values for Minimum, Maximum, Center, and Width for the current scan range. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. • Minimum Select the minimum value for the scan range (in mass-to-charge ratio units). You can enter any value from 30.0 to 2500.0. **NOTICE** Scanning below 50 *m/z* might reduce instrument performance. ▲ • Maximum Select the maximum value for the scan range (in mass-to-charge ratio units). You can enter any value from 30.4 to 3000.0. • Center Select the center mass (in mass-to-charge ratio units) of the scan range. You can enter any value from 30.2 to 2750.0. • Width Select the width of the scan range (in mass-to-charge ratio units). You can enter any value from 0.4 to 2800.0.
Spectrum data type	Use this list box to toggle between Profile data format and Centroid data format.

❖ **To show this view**

Click the Full MS symbol in the Graph pane.

Properties of Full MS / dd-MS^2 (TopN)

The properties of a Full MS / dd-MS^2 experiment include the following parameters:

Parameter	Description
General	
Runtime	Use this box to specify the duration in minutes of the active scan event. Click into the field to display the spin boxes for the start time (minimum) and the end time (maximum) of the scan event. To change a value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range for the start time is from 0 to (end time minus 0.01) minutes. The valid range for the end time is from (start time plus 0.01) to 10000 minutes. Alternatively, drag the left and right edges of the corresponding time bar to the desired positions on the time line in the Scan Groups pane.
Polarity	Use this list box to toggle between positive ion and negative ion polarity.
Inclusion	Use this field to enable the inclusion masses list that is attached to the active instrument method. Available options are on (inclusion list enabled) and – (inclusion list disabled). Default setting is –.
Exclusion	Use this field to enable the exclusion masses list that is attached to the active instrument method. Available options are on (exclusion list enabled) and – (exclusion list disabled). Default setting is –.
Tags	Use this field to enable the tag masses list for the active instrument method. Available options are on (list enabled) and – (list disabled). Default setting is –.
Full MS	
Microscans	Use this spin box to enter the number of microscans to be performed. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 to 10 microscans.
Resolution	Use this list box to select the mass resolution used during the selected scan event. The mass resolution of the Orbitrap analyzer is proportional to 1/sqrt(*m/z*). The scan time increases with increasing resolution and detect time. Available options are 15000, 30000, 60000, or 120000. Resolution is calculated according to the IUPAC peak width definition. The available settings represent m/Δm or the *m/z* value of an ion at *m/z* 200 divided by the full width at half its maximum height (FWHM).
AGC target	Use this list box to select the AGC target value for the selected scan event. The AGC target value controls the number of ions that are injected into the Orbitrap analyzer. Available options are 2e4, 5e4, 1e5, 2e5, 5e5, 1e6, 3e6, or 5e6.

Parameter	Description
Maximum IT	Use this list box to type or click a maximum allowed injection time to reach the AGC target value. For example, type **160**. The valid range is from 1 to 3000 milliseconds.
Number of scan ranges	Use this spin box to enter the number of segmented master scans. Segmented master scans can be defined independently from each other. They can be used to get a better S/N for specific areas and exclude matrix signals. The scan ranges are acquired one after another and will result one scan for each range. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. The valid range is from 1 to 8 scan ranges.
Scan range *n* (*n*=1–8)	Use the spin boxes in this field to enter values for Minimum, Maximum, Center, and Width for the current scan range. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. • Minimum Select the minimum value for the scan range (in mass-to-charge ratio units). You can enter any value from 30.0 to 2500.0. **NOTICE** Scanning below 50 *m/z* might reduce instrument performance. ▲ • Maximum Select the maximum value for the scan range (in mass-to-charge ratio units). You can enter any value from 30.4 to 3000.0. • Center Select the center mass (in mass-to-charge ratio units) of the scan range. You can enter any value from 30.2 to 2750.0. • Width Select the width of the scan range (in mass-to-charge ratio units). You can enter any value from 0.4 to 2800.0.
Spectrum data type	Use this list box to toggle between Profile data format and Centroid data format.
dd-MS^2 / dd-SIM	
Microscans	Use this spin box to enter the number of microscans to be performed. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 to 10 microscans.
Resolution	Use this list box to select the mass resolution used during the selected scan event. The mass resolution of the Orbitrap analyzer is proportional to 1/sqrt(*m/z*). The scan time increases with increasing resolution and detect time. Available options are 15000, 30000, 60000, or 120000. Resolution is calculated according to the IUPAC peak width definition. The available settings represent m/Δm or the *m/z* value of an ion at *m/z* 200 divided by the full width at half its maximum height (FWHM).
AGC target	Use this list box to select the AGC target value for the selected scan event. The AGC target value controls the number of ions that are injected into the Orbitrap analyzer. Available options are 2e4, 5e4, 1e5, 2e5, 5e5, 1e6, 3e6, or 5e6.

Parameter	Description
Maximum IT	Use this list box to type or click a maximum allowed injection time to reach the AGC target value. For example, type **160**. The valid range is from 1 to 3000 milliseconds.
Loop count	Use this spin box to enter the number of repetitions of the corresponding scan event before continuing with the next scan event or experiment cycle. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 (no repetition) to 100 repetitions.
MSX count	Use this spin box to enter the maximum number of precursors to be multiplexed in an scan event. In spectra multiplexing, multiple preselected precursors are collected in the C-Trap for simultaneous detection in the Orbitrap analyzer. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 (no spectral multiplexing) to 10 fillings.
TopN	This field displays the product of the selected values for Loop count and MSX count as maximum number of the most abundant precursors that will be selected.
Isolation window	Use this list box to enter the isolation width (in mass-to-charge ratio units) for the parent ion of interest. The mass range for the peak is centered at the parent mass and ranges one-half of the isolation width to either side of the parent mass. You can set the isolation window to any value from *m/z* 0.4 to 50 times the first mass of the Data Dependent scan. The default value is *m/z* 4.0. Too low a value results in loss of sensitivity because not all of the ions in a mass peak or in neighboring isotopic peaks are isolated. Too high a value causes interference from neighboring peaks.
Isolation offset	Use this spin box to define an absolute offset by which the isolation window is shifted. The resulting window is non-symmetric with regard to the precursor *m/z*. Thus, more isotopes of a cluster might be covered, without the need of expanding the isolation window. Example: Precursor = *m/z* 524.265; Isolation window = 2.0 *m/z*; Isolation offset = +0.5 *m/z*; Resulting isolation window = *m/z* 523.765–525.765 To change the isolation offset, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set the isolation offset to any value from *m/z* -50 to +50. Default is *m/z* 0.

Parameter	Description
Fixed first mass	Use this box to specify whether to use a first mass of the Data Dependent scan. After an automated determination of the detection mass range, the first mass will be set to the defined fixed first mass. Because only masses up to 15 times the first mass can be trapped in the C-Trap, loss of ions in the upper detection range may occur. To change the Fixed first mass, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set Fixed first mass either to – (no Fixed first mass) or to any value from *m/z* 30 to 750. Default is –.
CE / Stepped CE	Use this box to specify the collision energy (CE). This dimensionless number is approximately equivalent to the HCD collision energy (in eV) for a reference ion of mass 500 and charge 1. The actual HCD energy is calculated on basis of mass and charge of the selected precursor ion. If more than one CE value is entered, the Q Exactive GC mass spectrometer will perform a stepwise fragmentation on the precursor ion. All fragments created in the steps are collected and sent to the Orbitrap analyzer for one scan detection. Enter up to three CE values manually separated by blanks or the locally defined "list separator" (defined for Microsoft™ Windows™ via the "Region and Language" settings). Alternatively, click the down arrow to display a dialog box and select the corresponding number of check boxes. To change an CE value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set CE to any value from 10 to 200. Default is 30.
Spectrum data type	Use this list box to toggle between Profile data format and Centroid data format.
dd Settings	
Use the parameters in this group to specify criteria for selecting one or more ions of interest on which to perform subsequent scans.	
Underfill ratio	Use this box to specify a minimum percentage of the AGC Target value. If the mass peak of interest reaches this intensity within the maximum IT, the Q Exactive GC mass spectrometer initiates a Data Dependent scan. The valid range is from 0 to 100%. Example: AGC Target = 1e5, maximum IT = 250 ms, underfill ratio = 1%: trigger threshold is 1000 charges in 250 ms.
Intensity threshold	This field displays the minimum intensity that a mass peak requires to initiate a Data Dependent scan.

Parameter	Description
Apex trigger	To obtain the highest quality data in the shortest period of time, it is best to defer data acquisition until near the apex of a chromatographic peak. Using the chromatographic peak apex detection provides numerous benefits for Data Dependent GC/MS/MS, including: • Improved MS/MS quality and signal levels • Shorter ion injection times • Reduced MS/MS acquisition of chemical background • Less dependence on static or dynamic exclusion lists • Reduced retriggering of scans on tailing chromatographic peaks Use this box to enter start (Minimum) and end (Maximum) of a retention time window in seconds relative to the occurrence of a precursor. If the Q Exactive GC mass spectrometer finds an apex within the window, it triggers a Data Dependent acquisition for the corresponding *m/z* or range of *m/z*. The mass spectrometer also triggers a Data Dependent scan when the end of the time window is reached without detecting an apex. The apex trigger always depends on other selection criteria (for example, intensity threshold). • Minimum Enter the minimum value for the time window (in seconds). You can enter any value from – (0) to 360. The default value is –. • Maximum Enter the maximum value for the time window (in seconds). You can enter any value from – (0) to 360. The default value is –.
Exclude isotopes	Use this field to define a window around a reference Data Dependent mass (including dynamic exclusion list masses) inside which other peaks in the window will not trigger a subsequent data-dependent scan. Use this feature, for example, to exclude isotope clusters. Available options are on (isotopes exclusion enabled) and – (isotopes exclusion disabled). Default setting is **on**.

Parameter	Description
Dynamic exclusion	Use this box to enter a time (in seconds) to prevent an ion from triggering a subsequent Data Dependent scan after it has already triggered a Data Dependent scan. The following inputs are possible: • – (no dynamic exclusion) • 0.1 to 50000 Dynamic exclusion is valuable if the Q Exactive GC mass spectrometer has obtained enough data on an ion or if the base peak corresponds to a contaminant or an ion of non interest. If you enable dynamic exclusion, and if the mass spectrometer performs a Data Dependent scan on an ion, that ion is placed on the exclusion list (a temporary reject mass list) for a user-specified period of time. Then, when the mass spectrometer performs another Data Dependent scan on another ion, that ion is also placed on the exclusion list, and so on.
if idle	Use this field to select the behavior of the Q Exactive GC mass spectrometer when it has finished the Data Dependent scans that were triggered by a mass that meets the established criteria for Data Dependent actions. Available options are do not pick others and pick other. Default setting is **do not pick others**. This parameter is available only when at least one of the parameters Inclusion or Tags is set to **on**.

❖ **To show this view**

Click the Full MS / dd-MS^2 symbol in the Graph pane.

Properties of Targeted-SIM

The properties of a Targeted-SIM experiment include the following parameters:

Parameter	Description
General	
Runtime	Use this box to specify the duration in minutes of the active scan event. Click into the field to display the spin boxes for the start time (minimum) and the end time (maximum) of the scan event. To change a value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range for the start time is from 0 to (end time minus 0.01) minutes. The valid range for the end time is from (start time plus 0.01) to 10000 minutes. Alternatively, drag the left and right edges of the corresponding time bar to the desired positions on the time line in the Scan Groups pane.
Polarity	Use this list box to toggle between positive ion and negative ion polarity.

Parameter	Description
Inclusion	Because this experiment requires inclusion masses, this field is always set to on (inclusion list enabled).
SIM	
Microscans	Use this spin box to enter the number of microscans to be performed. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 to 10 microscans.
Resolution	Use this list box to select the mass resolution used during the selected scan event. The mass resolution of the Orbitrap analyzer is proportional to 1/sqrt(*m/z*). The scan time increases with increasing resolution and detect time. Available options are 15000, 30000, 60000, or 120000. Resolution is calculated according to the IUPAC peak width definition. The available settings represent m/Δm or the *m/z* value of an ion at *m/z* 200 divided by the full width at half its maximum height (FWHM).
AGC target	Use this list box to select the AGC target value for the selected scan event. The AGC target value controls the number of ions that are injected into the Orbitrap analyzer. Available options are 2e4, 5e4, 1e5, 2e5, 5e5, 1e6, 3e6, or 5e6.
Maximum IT	Use this list box to type or click a maximum allowed injection time to reach the AGC target value. For example, type **160**. The valid range is from 1 to 3000 milliseconds.
MSX count	Use this spin box to enter the maximum number of precursors to be multiplexed in an scan event. In spectra multiplexing, multiple preselected precursors are collected in the C-Trap for simultaneous detection in the Orbitrap analyzer. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 (no spectral multiplexing) to 10 fillings.
Isolation window	Use this list box to enter the isolation width (in mass-to-charge ratio units) for the parent ion of interest. The mass range for the peak is centered at the parent mass and ranges one-half of the isolation width to either side of the parent mass. You can set the isolation window to any value from *m/z* 0.4 to 50 times the first mass of the Data Dependent scan. The default value is *m/z* 4.0. Too low a value results in loss of sensitivity because not all of the ions in a mass peak or in neighboring isotopic peaks are isolated. Too high a value causes interference from neighboring peaks.

Parameter	Description
Isolation offset	Use this spin box to define an absolute offset by which the isolation window is shifted. The resulting window is non-symmetric with regard to the precursor *m/z*. Thus, more isotopes of a cluster might be covered, without the need of expanding the isolation window. Example: Precursor = *m/z* 524.265; Isolation window = 2.0 *m/z*; Isolation offset = +0.5 *m/z*; Resulting isolation window = *m/z* 523.765–525.765 To change the isolation offset, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set the isolation offset to any value from *m/z* -50 to +50. Default is *m/z* 0.
Spectrum data type	Use this list box to toggle between Profile data format and Centroid data format.

❖ **To show this view**

Click the Targeted-SIM symbol in the Graph pane.

Properties of PRM

The properties of a PRM experiment include the following parameters:

Parameter	Description
General	
Runtime	Use this box to specify the duration in minutes of the active scan event. Click into the field to display the spin boxes for the start time (minimum) and the end time (maximum) of the scan event. To change a value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range for the start time is from 0 to (end time minus 0.01) minutes. The valid range for the end time is from (start time plus 0.01) to 10000 minutes. Alternatively, drag the left and right edges of the corresponding time bar to the desired positions on the time line in the Scan Groups pane.
Polarity	Use this list box to toggle between positive ion and negative ion polarity.
Inclusion	Because this experiment requires inclusion masses, this field is always set to **on** (inclusion list enabled).
MS^2	
Microscans	Use this spin box to enter the number of microscans to be performed. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 to 10 microscans.

Parameter	Description
Resolution	Use this list box to select the mass resolution used during the selected scan event. The mass resolution of the Orbitrap analyzer is proportional to 1/sqrt(*m/z*). The scan time increases with increasing resolution and detect time. Available options are 15000, 30000, 60000, or 120000. Resolution is calculated according to the IUPAC peak width definition. The available settings represent m/Δm or the *m/z* value of an ion at *m/z* 200 divided by the full width at half its maximum height (FWHM).
AGC target	Use this list box to select the AGC target value for the selected scan event. The AGC target value controls the number of ions that are injected into the Orbitrap analyzer. Available options are 2e4, 5e4, 1e5, 2e5, 5e5, 1e6, 3e6, or 5e6.
Maximum IT	Use this list box to type or click a maximum allowed injection time to reach the AGC target value. For example, type **160**. The valid range is from 1 to 3000 milliseconds.
MSX count	Use this spin box to enter the maximum number of precursors to be multiplexed in an scan event. In spectra multiplexing, multiple preselected precursors are collected in the C-Trap for simultaneous detection in the Orbitrap analyzer. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 (no spectral multiplexing) to 10 fillings.
MSX isochronous ITs	This field shows **on** to indicate that for multiplexing scans, all isolated windows are injected into the HCD cell with the same injection time.
Isolation window	Use this list box to enter the isolation width (in mass-to-charge ratio units) for the parent ion of interest. The mass range for the peak is centered at the parent mass and ranges one-half of the isolation width to either side of the parent mass. You can set the isolation window to any value from *m/z* 0.4 to 50 times the first mass of the Data Dependent scan. The default value is *m/z* 4.0. Too low a value results in loss of sensitivity because not all of the ions in a mass peak or in neighboring isotopic peaks are isolated. Too high a value causes interference from neighboring peaks.
Isolation offset	Use this spin box to define an absolute offset by which the isolation window is shifted. The resulting window is non-symmetric with regard to the precursor *m/z*. Thus, more isotopes of a cluster might be covered, without the need of expanding the isolation window. Example: Precursor = *m/z* 524.265; Isolation window = 2.0 *m/z*; Isolation offset = +0.5 *m/z*; Resulting isolation window = *m/z* 523.765–525.765 To change the isolation offset, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set the isolation offset to any value from *m/z* -50 to +50. Default is *m/z* 0.

Parameter	Description
Fixed first mass	Use this box to specify whether to use a first mass of the Data Dependent scan. After an automated determination of the detection mass range, the first mass will be set to the defined fixed first mass. Because only masses up to 15 times the first mass can be trapped in the C-Trap, loss of ions in the upper detection range may occur. To change the Fixed first mass, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set Fixed first mass either to – (no Fixed first mass) or to any value from *m/z* 30 to 750. Default is –.
CE / Stepped CE	Use this box to specify the collision energy (CE). This dimensionless number is approximately equivalent to the HCD collision energy (in eV) for a reference ion of mass 500 and charge 1. The actual HCD energy is calculated on basis of mass and charge of the selected precursor ion. If more than one CE value is entered, the Q Exactive GC mass spectrometer will perform a stepwise fragmentation on the precursor ion. All fragments created in the steps are collected and sent to the Orbitrap analyzer for one scan detection. Enter up to three CE values manually separated by blanks or the locally defined “list separator” (defined for Microsoft™ Windows™ via the “Region and Language” settings). Alternatively, click the down arrow to display a dialog box and select the corresponding number of check boxes. To change an CE value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set CE to any value from 10 to 200. Default is 30.
Spectrum data type	Use this list box to toggle between Profile data format and Centroid data format.

❖ **To show this view**

Click the PRM symbol in the Graph pane.

Properties of Targeted-SIM / dd-MS^2

The properties of a Targeted-SIM / dd-MS^2 experiment include the following parameters:

Parameter	Description
General	
Runtime	Use this box to specify the duration in minutes of the active scan event. Click into the field to display the spin boxes for the start time (minimum) and the end time (maximum) of the scan event. To change a value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range for the start time is from 0 to (end time minus 0.01) minutes. The valid range for the end time is from (start time plus 0.01) to 10000 minutes. Alternatively, drag the left and right edges of the corresponding time bar to the desired positions on the time line in the Scan Groups pane.
Polarity	Use this list box to toggle between positive ion and negative ion polarity.
Inclusion	Because this experiment requires inclusion masses, this field is always set to **on** (inclusion list enabled).
SIM	
Microscans	Use this spin box to enter the number of microscans to be performed. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 to 10 microscans.
Resolution	Use this list box to select the mass resolution used during the selected scan event. The mass resolution of the Orbitrap analyzer is proportional to 1/sqrt(*m/z*). The scan time increases with increasing resolution and detect time. Available options are 15000, 30000, 60000, or 120000. Resolution is calculated according to the IUPAC peak width definition. The available settings represent m/Δm or the *m/z* value of an ion at *m/z* 200 divided by the full width at half its maximum height (FWHM).
AGC target	Use this list box to select the AGC target value for the selected scan event. The AGC target value controls the number of ions that are injected into the Orbitrap analyzer. Available options are 2e4, 5e4, 1e5, 2e5, 5e5, 1e6, 3e6, or 5e6.
Maximum IT	Use this list box to type or click a maximum allowed injection time to reach the AGC target value. For example, type **160**. The valid range is from 1 to 3000 milliseconds.
Loop count	Use this spin box to enter the number of repetitions of the corresponding scan event before continuing with the next scan event or experiment cycle. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 (no repetition) to 100 repetitions.

Parameter	Description
MSX count	Use this spin box to enter the maximum number of precursors to be multiplexed in an scan event. In spectra multiplexing, multiple preselected precursors are collected in the C-Trap for simultaneous detection in the Orbitrap analyzer. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 (no spectral multiplexing) to 10 fillings.
Isolation window	Use this spin box to enter the isolation width (in mass-to-charge ratio units) for the parent ion of interest. The mass range for the peak is centered at the parent mass and ranges one-half of the isolation width to either side of the parent mass. You can set the isolation window to any value from *m/z* 0.4 to 50 times the first mass of the Data Dependent scan. The default value is *m/z* 4.0. Too low a value results in loss of sensitivity because not all of the ions in a mass peak or in neighboring isotopic peaks are isolated. Too high a value causes interference from neighboring peaks.
Isolation offset	Use this spin box to define an absolute offset by which the isolation window is shifted. The resulting window is non-symmetric with regard to the precursor *m/z*. Thus, more isotopes of a cluster might be covered, without the need of expanding the isolation window. Example: Precursor = *m/z* 524.265; Isolation window = 2.0 *m/z*; Isolation offset = +0.5 *m/z*; Resulting isolation window = *m/z* 523.765–525.765 To change the isolation offset, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set the isolation offset to any value from *m/z* -50 to +50. Default is *m/z* 0.
Spectrum data type	Use this list box to toggle between Profile data format and Centroid data format.
dd-MS^2	
Microscans	Use this spin box to enter the number of microscans to be performed. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 to 10 microscans.
Resolution	Use this list box to select the mass resolution used during the selected scan event. The mass resolution of the Orbitrap analyzer is proportional to 1/sqrt(*m/z*). The scan time increases with increasing resolution and detect time. Available options are 15000, 30000, 60000, or 120000. Resolution is calculated according to the IUPAC peak width definition. The available settings represent m/Δm or the *m/z* value of an ion at *m/z* 200 divided by the full width at half its maximum height (FWHM).
AGC target	Use this list box to select the AGC target value for the selected scan event. The AGC target value controls the number of ions that are injected into the Orbitrap analyzer. Available options are 2e4, 5e4, 1e5, 2e5, 5e5, 1e6, 3e6, or 5e6.

Parameter	Description
Maximum IT	Use this list box to type or click a maximum allowed injection time to reach the AGC target value. For example, type **160**. The valid range is from 1 to 3000 milliseconds.
Loop count	Use this spin box to enter the number of repetitions of the corresponding scan event before continuing with the next scan event or experiment cycle. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 (no repetition) to 100 repetitions.
MSX count	Use this spin box to enter the maximum number of precursors to be multiplexed in an scan event. In spectra multiplexing, multiple preselected precursors are collected in the C-Trap for simultaneous detection in the Orbitrap analyzer. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 (no spectral multiplexing) to 10 fillings.
TopN	This field displays the product of the selected values for Loop count and MSX count as maximum number of the most abundant precursors that will be selected.
Isolation window	Use this spin box to enter the isolation width (in mass-to-charge ratio units) for the parent ion of interest. The mass range for the peak is centered at the parent mass and ranges one-half of the isolation width to either side of the parent mass. You can set the isolation window to any value from *m/z* 0.4 to 50 times the first mass of the Data Dependent scan. The default value is *m/z* 4.0. Too low a value results in loss of sensitivity because not all of the ions in a mass peak or in neighboring isotopic peaks are isolated. Too high a value causes interference from neighboring peaks.
Isolation offset	Use this spin box to define an absolute offset by which the isolation window is shifted. The resulting window is non-symmetric with regard to the precursor *m/z*. Thus, more isotopes of a cluster might be covered, without the need of expanding the isolation window. Example: Precursor = *m/z* 524.265; Isolation window = 2.0 *m/z*; Isolation offset = +0.5 *m/z*; Resulting isolation window = *m/z* 523.765–525.765 To change the isolation offset, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set the isolation offset to any value from *m/z* -50 to +50. Default is *m/z* 0.
Fixed first mass	Use this box to specify whether to use a first mass of the Data Dependent scan. After an automated determination of the detection mass range, the first mass will be set to the defined fixed first mass. Because only masses up to 15 times the first mass can be trapped in the C-Trap, loss of ions in the upper detection range may occur. To change the Fixed first mass, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set Fixed first mass either to – (no Fixed first mass) or to any value from *m/z* 30 to 750. Default is –.

Parameter	Description
CE / Stepped CE	Use this box to specify the collision energy (CE). This dimensionless number is approximately equivalent to the HCD collision energy (in eV) for a reference ion of mass 500 and charge 1. The actual HCD energy is calculated on basis of mass and charge of the selected precursor ion. If more than one CE value is entered, the Q Exactive GC mass spectrometer will perform a stepwise fragmentation on the precursor ion. All fragments created in the steps are collected and sent to the Orbitrap analyzer for one scan detection. Enter up to three CE values manually separated by blanks or the locally defined "list separator" (defined for Microsoft™ Windows™ via the "Region and Language" settings). Alternatively, click the down arrow to display a dialog box and select the corresponding number of check boxes. To change an CE value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can set CE to any value from 10 to 200. Default is 30.
Spectrum data type	Use this list box to toggle between Profile data format and Centroid data format.
dd Settings	
Underfill ratio	Use this box to specify a minimum percentage of the AGC Target value. If the mass peak of interest reaches this intensity within the Maximum IT, the Q Exactive GC mass spectrometer initiates a Data Dependent scan. The valid range is from 0 to 100%. Example: AGC Target = 1e5, Maximum IT = 250 ms, Underfill ratio = 1%: trigger threshold is 1000 charges in 250 ms.
Intensity threshold	This field displays the minimum intensity that a mass peak requires to initiate a Data Dependent scan.

Parameter	Description
Apex trigger	To obtain the highest quality data in the shortest period of time, it is best to defer data acquisition until near the apex of a chromatographic peak. Using the chromatographic peak apex detection provides numerous benefits for Data Dependent GC/MS/MS, including: • Improved MS/MS quality and signal levels • Shorter ion injection times • Reduced MS/MS acquisition of chemical background • Less dependence on static or dynamic exclusion lists • Reduced retriggering of scans on tailing chromatographic peaks Use this box to enter start (Minimum) and end (Maximum) of a retention time window in seconds relative to the occurrence of a precursor. If the Q Exactive GC mass spectrometer finds an apex within the window, it triggers a Data Dependent acquisition for the corresponding *m/z* or range of *m/z*. The mass spectrometer also triggers a Data Dependent scan when the end of the time window is reached without detecting an apex. The apex trigger always depends on other selection criteria (for example, intensity threshold). • Minimum Enter the minimum value for the time window (in seconds). You can enter any value from – (0) to 360. The default value is –. • Maximum Enter the maximum value for the time window (in seconds). You can enter any value from – (0) to 360. The default value is –.
Exclude isotopes	Use this field to define a window around a reference Data Dependent mass (including dynamic exclusion list masses) inside which other peaks in the window will not trigger a subsequent data-dependent scan. Use this feature, for example, to exclude isotope clusters. Available options are on (isotopes exclusion enabled) and – (isotopes exclusion disabled). Default setting is **on**.
Dynamic exclusion	Use this box to enter a time (in seconds) to prevent an ion from triggering a subsequent Data Dependent scan after it has already triggered a Data Dependent scan. The following inputs are possible: • – (no dynamic exclusion) • 0.1 to 50000 Dynamic exclusion is valuable if the Q Exactive GC mass spectrometer has obtained enough data on an ion or if the base peak corresponds to a contaminant or an ion of non interest. If you enable dynamic exclusion, and if the mass spectrometer performs a Data Dependent scan on an ion, that ion is placed on the exclusion list (a temporary reject mass list) for a user-specified period of time. Then, when the mass spectrometer performs another Data Dependent scan on another ion, that ion is also placed on the exclusion list, and so on.

❖ **To show this view**

Click the Targeted-SIM / dd-MS^2 symbol in the Graph pane.

Summary Page

The Summary page displays the parameters for mass spectrometer setup and contact closure that you specified on the Experiment Setup page.

❖ **To zoom in or out on the Summary page**

- Position the mouse pointer within the pane, press the <Ctrl> key, and roll the mouse wheel forward to zoom in.
- Position the mouse pointer within the pane, press the <Ctrl> key, and roll the wheel backward to zoom out.

NOTICE The content of this page will be printed together with the information for other instruments. ▲

❖ **To display this page**

In the Instrument Setup window, click the Summary tab.

Dialog Boxes of the Experiment Setup Page

This section provides a reference to the dialog boxes in the Experiment Setup page:

- Use the dialog boxes for global lists to define properties of masses that are used for setting up the experiments in the Experiment Setup page:
  - Lock Masses Dialog Box
  - Inclusion List Dialog Box
  - Exclusion List Dialog Box
  - Tag Masses Dialog Box
- Mass Calculator

Lock Masses Dialog Box

Use the Lock Masses dialog box to select a lock mass list for the active instrument method and to edit it. See Figure 3-12. Lock masses are peaks in the spectrum that the Q Exactive GC mass spectrometer uses for internal mass calibration. Using lock masses improves the mass accuracy of the mass analyzer. If you do not specify any lock masses, the mass spectrometer uses the external mass calibration.

The dialog box allows defining a time window of activity for each lock mass (=timed lock masses). If Start or End is left blank, the lock mass is active from the beginning or until the end of the method, respectively. In the latter case, the End field displays “end.” The usage of lock masses during the active instrument method is specified on the Properties pane of the method.

The table shows the properties of the masses contained in the active lock mass list (*.lock-masses). To change the sort order, click the respective table column header. To invert the sort order, click again.

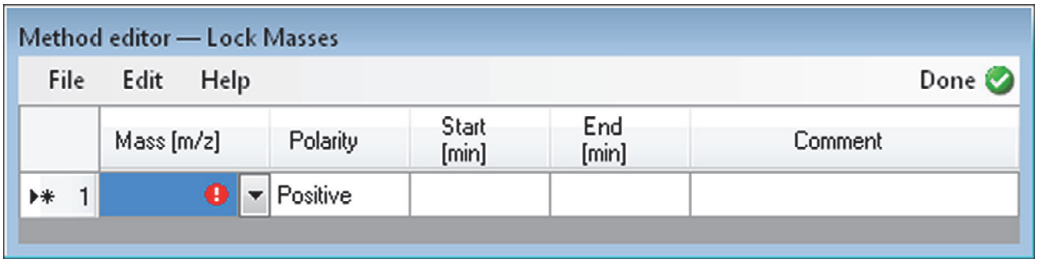


Figure 3-12. Method Editor—Lock Masses dialog box

The Lock Masses dialog box has the following parameters:

Parameter	Description
Mass [m/z]	Enter the mass-to-charge-ratio of the lock mass into the field (with a maximum of five decimals). To change this value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, enter a value in the spin box text field.
Polarity	Use this list box to toggle between positive ion and negative ion polarity. During a scan, the Q Exactive GC mass spectrometer uses only the lock masses with a polarity that matches the active ion mode.
Start [min]	Enter the start time in minutes. Alternatively, you can enter a time in seconds (by appending the letter *s*) or in hours (by appending the letter *h*). The software automatically converts your input into minutes.
End [min]	Enter the end time in minutes. Alternatively, you can enter a time in seconds (by appending the letter *s*) or in hours (by appending the letter *h*). The software automatically converts your input into minutes.
Comment	Enter a comment for the lock mass into the field. This field is optional.

❖ **To display this dialog box**

1. In the Method Editor, expand the Global Lists pane.
2. Click the  Lock Masses button.

Title Bar

The title bar of a global list displays the name of the dialog box. An asterisk (*) indicates a file with unsaved changes.

File Menu

The File menu provides commands for file operations. It has the following commands:

Command	Description
Import <Ctrl> + <I>	Opens a dialog box where you can select an existing mass collection. In addition to the program-specific data files, you can select *.csv or *.txt files that meet the requirements of the program. All available masses are replaced by the masses contained in the imported file. Click **Open** to import the file.
Export <Ctrl> + <S>	Opens a dialog box that you can use to save the active mass collection. In addition to using the program-specific data files, you can save the mass collection as tabulator-separated file (*.txt, for example) or comma-separated file (*.csv, for example). Enter a new file name, select the file type, and select the location (disk and directory) where you want to save it. Alternatively, you can select an existing file. Click **Save** to save the file.

Command	Description
Export selected <Ctrl> + <Shift> + <S>	Opens a dialog box that you can use to save the selected masses. In addition to using the program-specific data files, you can save the mass collection as tabulator-separated file (*.txt, for example) or comma-separated file (*.csv, for example). Enter a new file name, select the file type, and select the location (disk and directory) where you want to save it. Alternatively, you can select an existing file. Click **Save** to save the file. This command is available only when at least one line in the dialog box is selected.
Minimize <Ctrl> + <F4>	Closes the dialog box. Your edits will be retained. Alternatively, click the **Done** button at the right side of the menu bar.

Edit Menu

The Edit menu provides commands for editing the active file. It has the following commands:

Command	Description
Undo until last save <Ctrl> + <Z>	Discards all changes to the active file that have not been saved.
Copy <Ctrl> + <C>	Copies the data of the selected mass to the clipboard.
Replace by clipboard <Ctrl> + <V>	Replaces the available masses by the content of the clipboard. **NOTICE** The clipboard stores lock mass data as tabulator-separated text files. When the data in the clipboard are not in this format, the command is not available. ▲

Help Menu

Displays the Help for this dialog box.

Inclusion List Dialog Box

Use the Inclusion List dialog box to edit a list of specific masses (inclusion masses) that can activate a Data Dependent action. See Figure 3-13. The dialog box allows defining a time window of activity for each inclusion mass. If Start or End is left blank, the inclusion mass is active from the beginning or until the end of the method, respectively. In the latter case, the End field displays “end.”

To use an inclusion list in a experiment, set the Inclusion parameter on the Parameter pane to **on**. When a inclusion mass list is used in a Data Dependent experiment (Full MS / dd-MS^2 and Targeted-SIM / dd-MS^2), the ions will be selected for the Data Dependent scan event, *only* if they are:

1. appearing in the referring master scan (mostly the full scan) and
2. fulfill all of the other defined selection criteria, such as Intensity Threshold.

Because target-based experiments or experiment parts (Targeted-SIM, PRM, Targeted-SIM / dd-MS^2) are based on an inclusion list, the Inclusion parameter is always set to **on** for these experiments.

The table shows the properties of the masses contained in the active inclusion masses list (*.include-masses). To change the sort order, click the respective table column header. To invert the sort order, click again.

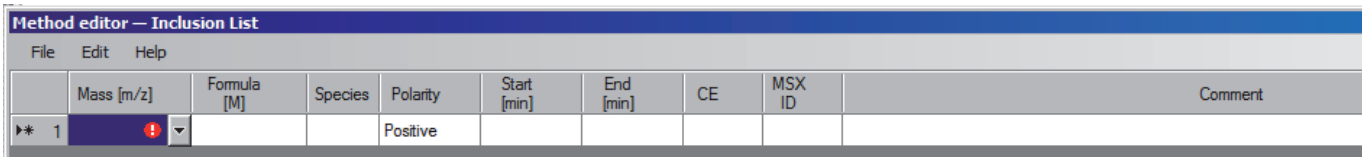


Figure 3-13. Method Editor—Inclusion List dialog box

The Inclusion List dialog box has the following parameters:

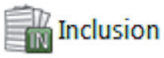
Parameter	Description
Mass [m/z]	Enter the mass-to-charge-ratio of the inclusion mass into the field (with a maximum of five decimals). **NOTICE** If you use the Mass Calculator to edit the Formula field, this field is automatically updated when you exit the editor. ▲

Parameter	Description
Formula [M]	Use this field to describe the structure of the active compound. The different ways of definition are called formula types. You can either enter the formula directly or use the Mass Calculator. To display this editor, click the down arrow in this field. Pay attention to the following restrictions: • When you enter a valid formula, the Method Editor will set the default values for charge state (1) and Species (+H/-H). Then it will display an automatically calculated value in the Mass [m/z] field. • When you enter a formula that the Method Editor cannot interpret, the Mass [m/z] field will stay empty. When you save the method, the Method Editor will warn you that the method is not in the shape to be executed (due to the missing *m/z* value). When you reopen the method, the Method Editor will display exactly the formula that was entered. • When you enter a formula, The Method Editor requires a value for the charge state to calculate an *m/z* value. You can, however, save the method without specifying a charge state for the entry. When you save the method, the Method Editor will warn you that the method is not in the shape to be executed (due to the missing charge state). When you reopen the method, the Method Editor will display exactly the formula that was entered. • Entries of compounds with calculated *m/z* values outside the measurement specifications of the mass spectrometer (*m/z*=30–3000) can be saved in an instrument method. The instrument software, however, will ignore these entries when it executes the method. • When you change the *m/z* value that was calculated for the given set of formula, species and charge state, the Method Editor will clear the formula and species definition. The charge state will be preserved. • When you change the polarity for a set of calculated *m/z*, formula, species and charge state, the Method Editor will clear the Mass [m/z] field if the species definition is not valid for this polarity. You then need to change the species definition.

Parameter	Description
Species	Use this text field to define adducts or modifications of the active compound that are expected to be formed. The default value is +H for positive polarity and –H for negative polarity. The Method Editor will then recalculate the value in the Mass [m/z] field, if possible. The Species field can be used in two ways: • Click the down arrow to display a list of predefined adducts for each polarity: - +H, +NH4, +C2H5, +C3H5, +C4H9 for positive polarity - -H for negative polarity - an empty entry to express adducts (for example, radical cations) for both polarities. The corresponding set will be displayed depending on the selected polarity. Selecting an adduct (A) will result in the strict behavior of applying one unit A to the compound and using "+H" or "-H" adduct depending on the charge state and active polarity. • Enter the modifications of the compound by using squared brackets and at least M as representation of the basic compound. The predefined adducts can be entered, too. This definition is used without additional auto dependencies, like adding protons. **NOTICE** If you use the Mass Calculator to edit the Formula field, the Species field is automatically filled when you exit the editor. ▲
Polarity	Use this list box to toggle between positive ion and negative ion polarity. During a scan, the Q Exactive GC mass spectrometer uses only the inclusion masses with a polarity that matches the active ion mode. **NOTICE** If you use the Mass Calculator to edit the Formula field, this field is automatically updated when you exit the editor. ▲
Start [min]	Enter the start of the time window (in minutes) that corresponds to the mass in this row. Alternatively, you can enter a time in seconds (by appending the letter *s*) or in hours (by appending the letter *h*). Q Exactive GC Tune automatically converts your input into minutes.
End [min]	Enter the end of the time window in minutes. Alternatively, you can enter a time in seconds (by appending the letter *s*) or in hours (by appending the letter *h*). Q Exactive GC Tune automatically converts your input into minutes.
CE	Use the spin box to enter the RF amplitude (as a percentage) used to fragment ions. The valid range is 10 to 200%. The default value is 35%. In the list, the software adds the prefix "ce:" to collision energy values (for example, ce:35). A high collision energy value results in more energy deposition (which generally leads to more fragmentation). A low collision energy value results in less energy deposition (which generally leads to less fragmentation).

Parameter	Description
MSX ID	Enter the number of the multiplexed scan event in which the inclusion mass was analyzed.
Comment	Enter a comment for the inclusion mass into the field. This field is optional.

❖ **To display this dialog box**

1. In the Method Editor, expand the Global Lists pane.
2. Click the 

button.

Title Bar

The title bar of a global list displays the name of the dialog box. An asterisk (*) indicates a file with unsaved changes.

File Menu

The File menu provides commands for file operations. It has the following commands:

Command	Description
Import <Ctrl> + <I>	Opens a dialog box where you can select an existing mass collection. In addition to the program-specific data files, you can select *.csv or *.txt files that meet the requirements of the program. All available masses are replaced by the masses contained in the imported file. Click **Open** to import the file. When you want to import *.csv or *.txt files with entries that contain formulas, an extra third column “Formula type,” which contains the prefix, must specify the formula type. Valid values for the fields that can be (re)interpreted are: • for Peptide type: “Peptide”, “p”, and “p:” (all case insensitive), • for Amino acid type: “Amino acid”, “aa”, “aa:” (all case insensitive), • for Chemical formula type: “Chemical formula”, “c”, “c:” (all case insensitive), and “” (empty).
Export <Ctrl> + <S>	Opens a dialog box that you can use to save the active mass collection. In addition to using the program-specific data files, you can save the mass collection as tabulator-separated file (*.txt, for example) or comma-separated file (*.csv, for example). Enter a new file name, select the file type, and select the location (disk and directory) where you want to save it. Alternatively, you can select an existing file. Click **Save** to save the file. When you export *.csv or *.txt files with entries that contain formulas, an extra third column “Formula type” specifies the formula type. Values for the fields that are: • for Peptide type: “Peptide”, • for Amino acid type: “Amino acid”, • for Chemical formula type: “” (empty).

Command	Description
Export selected <Ctrl> + <Shift> + <S>	Opens a dialog box that you can use to save the selected masses. In addition to using the program-specific data files, you can save the mass collection as tabulator-separated file (*.txt, for example) or comma-separated file (*.csv, for example). Enter a new file name, select the file type, and select the location (disk and directory) where you want to save it. Alternatively, you can select an existing file. Click **Save** to save the file. This command is available only when at least one line in the dialog box is selected.
Minimize <Ctrl> + <F4>	Closes the dialog box. Your edits will be retained. Alternatively, click the **Done** button at the right side of the menu bar.

Edit Menu

The Edit menu provides commands for editing the active file. It has the following commands:

Command	Description
Undo until last save <Ctrl> + <Z>	Discards all changes to the active file that have not been saved.
Copy <Ctrl> + <C>	Copies the data of the selected mass to the clipboard.
Replace by clipboard <Ctrl> + <V>	Replaces the available masses by the content of the clipboard. **NOTICE** The clipboard stores lock mass data as tabulator-separated text files. When the data in the clipboard are not in this format, the command is not available. ▲

Help Menu

Displays the Help for this dialog box.

Exclusion List Dialog Box

Use the Exclusion List dialog box to edit a list of specific masses (exclusion masses) that will not trigger a subsequent Data Dependent scan even if they are present. See Figure 3-14. The dialog box allows defining a time window of activity for each exclusion mass. If Start or End is left blank, the exclusion mass is active from the beginning or until the end of the method, respectively. In the latter case, the End field displays “end.”

To use an exclusion list in a experiment, set the Exclusion parameter on the Parameter pane to **on**.

The table shows the properties of the masses contained in the active exclusion masses list (*.exclude-masses). To change the sort order, click the respective table column header. To invert the sort order, click again.

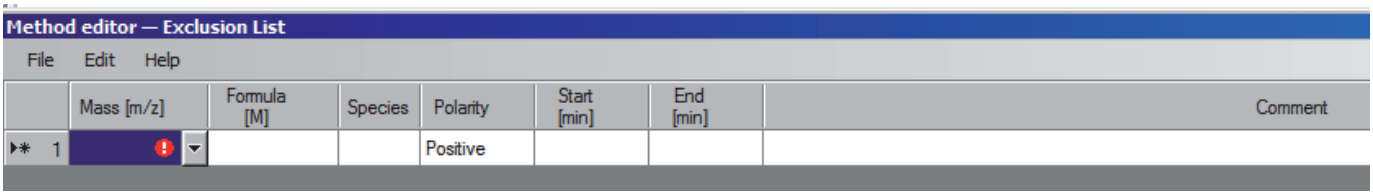


Figure 3-14. Method Editor—Exclusion List dialog box

The Exclusion List dialog box has the following parameters:

Parameter	Description
Mass [*m/z*]	Enter the mass-to-charge-ratio of the exclusion mass into the field (with a maximum of five decimals). **NOTICE** If you use the Mass Calculator to edit the Formula field, this field is automatically updated when you exit the editor. ▲
Formula [M]	Use this field to describe the structure of the active compound. You can either enter the formula directly or use the Mass Calculator. To display this editor, click the down arrow in this field. Pay attention to the following restrictions: • When you enter a valid formula, the Method Editor will set the default values for charge state (1) and Species (+H/–H). Then it will display an automatically calculated value in the Mass [m/z] field. • When you enter a formula that the Method Editor cannot interpret, the Mass [m/z] field will stay empty. When you save the method, the Method Editor will warn you that the method is not in the shape to be executed (due to the missing *m/z* value). When you reopen the method, the Method Editor will display exactly the formula that was entered. • When you enter a formula, The Method Editor requires a value for the charge state to calculate an *m/z* value. You can, however, save the method without specifying a charge state for the entry. When you save the method, the Method Editor will warn you that the method is not in the shape to be executed (due to the missing charge state). When you reopen the method, the Method Editor will display exactly the formula that was entered. • Entries of compounds with calculated *m/z* values outside the measurement specifications of the mass spectrometer (*m/z*=30–3000) can be saved in an instrument method. The instrument software, however, will ignore these entries when it executes the method. • When you change the *m/z* value that was calculated for the given set of formula, species and charge state, the Method Editor will clear the formula and species definition. The charge state will be preserved. • When you change the polarity for a set of calculated *m/z*, formula, species and charge state, the Method Editor will clear the Mass [*m/z*] field if the species definition is not valid for this polarity. You then need to change the species definition.

Parameter	Description
Species	Use this field to define adducts or modifications of the active compound that are expected to be formed. The default value is +H for positive polarity and –H for negative polarity. The Method Editor will then recalculate the value in the Mass [m/z] field, if possible. The Species field can be used in two ways: • Click the down arrow to display a list of predefined adducts for each polarity: - +H, +NH4, +C2H5, +C3H5+, +C4H9 for positive polarity - -H for negative polarity - an empty entry to express adducts (for example, radical cations) for both polarities. The corresponding set will be displayed depending on the selected polarity. Selecting an adduct (A) will result in the strict behavior of applying one unit A to the compound and using "+H" or "-H" adduct depending on charge state and active polarity. • Enter the modifications of the compound by using squared brackets and at least M as representation of the basic compound. The predefined adducts can be entered, too. This definition is used without additional auto dependencies, like adding protons. If you select or define a species and the Formula field is empty, the species definition will stay grayed out. **NOTICE** If you use the Mass Calculator to edit the Formula field, this field is automatically updated when you exit the editor. ▲
Polarity	Use this list box to toggle between positive ion and negative ion polarity. During a scan, the Q Exactive GC mass spectrometer uses only the exclusion masses with a polarity that matches the active ion mode. **NOTICE** If you use the Mass Calculator to edit the Formula field, this field is automatically updated when you exit the editor. ▲
Start [min]	Enter the start of the time window (in minutes) that corresponds to the mass in this row. Alternatively, you can enter a time in seconds (by appending the letter *s*) or in hours (by appending the letter *h*). The software automatically converts your input into minutes.
End [min]	Enter the end of the time window in minutes. Alternatively, you can enter a time in seconds (by appending the letter *s*) or in hours (by appending the letter *h*). The software automatically converts your input into minutes.
Comment	Enter a comment for the exclusion mass into the field. This field is optional.

❖ **To display this dialog box**

1. In the Method Editor, expand the Global Lists pane.
2. Click the  button.

Title Bar

The title bar of a global list displays the name of the dialog box. An asterisk (*)indicates a file with unsaved changes.

File Menu

The File menu provides commands for file operations. It has the following commands:

Command	Description
Import <Ctrl> + <I>	Opens a dialog box where you can select an existing mass collection. In addition to the program-specific data files, you can select *.csv or *.txt files that meet the requirements of the program. All available masses are replaced by the masses contained in the imported file. Click **Open** to import the file. When you want to import *.csv or *.txt files with entries that contain formulas, an extra third column “Formula type,” which contains the prefix, must specify the formula type. Valid values for the field that can be (re)interpreted are: • for Chemical formula type: “Chemical formula”, “c”, “c:” (all case insensitive), and “” (empty).
Export <Ctrl> + <S>	Opens a dialog box that you can use to save the active mass collection. In addition to using the program-specific data files, you can save the mass collection as tabulator-separated file (*.txt, for example) or comma-separated file (*.csv, for example). Enter a new file name, select the file type, and select the location (disk and directory) where you want to save it. Alternatively, you can select an existing file. Click **Save** to save the file. When you export *.csv or *.txt files with entries that contain formulas, an extra third column “Formula type” specifies the formula type. The value for the field is: • for Chemical formula type: “” (empty).
Export selected <Ctrl> + <Shift> + <S>	Opens a dialog box that you can use to save the selected masses. In addition to using the program-specific data files, you can save the mass collection as tabulator-separated file (*.txt, for example) or comma-separated file (*.csv, for example). Enter a new file name, select the file type, and select the location (disk and directory) where you want to save it. Alternatively, you can select an existing file. Click **Save** to save the file. This command is available only when at least one line in the dialog box is selected.
Minimize <Ctrl> + <F4>	Closes the dialog box. Your edits will be retained. Alternatively, click the **Done** button at the right side of the menu bar.

Edit Menu

The Edit menu provides commands for editing the active file. It has the following commands:

Command	Description
Undo until last save <Ctrl> + <Z>	Discards all changes to the active file that have not been saved.
Copy <Ctrl> + <C>	Copies the data of the selected mass to the clipboard.
Replace by clipboard <Ctrl> + <V>	Replaces the available masses by the content of the clipboard. **NOTICE** The clipboard stores lock mass data as tabulator-separated text files. When the data in the clipboard are not in this format, the command is not available. ▲

Help Menu

Displays the Help for this dialog box.

Tag Masses Dialog Box

An MS/MS scan can be triggered by a mass that has a 'partner' mass a user-defined delta away. Use the Tag Masses dialog box to edit a list of such mass deltas. See Figure 3-12. This is useful for applications where mass tags are used, or where mass pairs are known, and can greatly reduce the number of MS/MS scans on non-tagged species.

Tag masses can be used only in the Full MS / dd-MS^2 experiment. To use tag masses, set the Tags parameter on the Parameter pane to **on**.

The table displays the properties of the available tag masses. To change the sort order, click the respective table column header. To invert the sort order, click again.

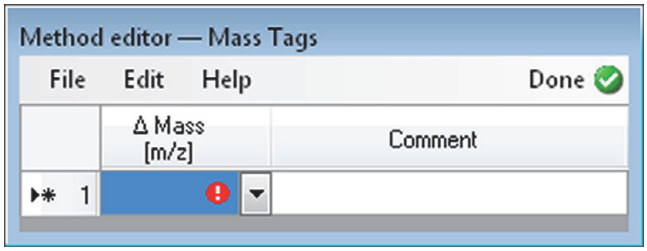
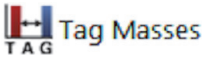


Figure 3-15. Method Editor—Tag Masses dialog box

The Tag Masses dialog box has the following parameters:

Parameter	Description
Δ Mass [m/z]	Use this spin box to enter the mass difference (with a maximum of five decimals). Depending on the sign, one precursor of the pair will be triggered. If you want to trigger on the heavy precursor (with a tag or isotope label), you need to enter a negative sign (for example, –6.02). It will be checked if an *m/z* distance towards lower *m/z* will be found. If so, the precursor at higher *m/z* will be selected. A positive *m/z* distance leads to the selection of the precursor with a lower *m/z*. If both precursors should be selected, two entries are needed, one with positive and one with negative sign. The valid range is 0.00001 to ±4000.00000.
Comment	Enter a comment into the field. This field is optional.

❖ **To display this dialog box**

1. In the Method Editor, expand the Global Lists pane.
2. Click the 

button.

Title Bar

The title bar of a global list displays the name of the dialog box. An asterisk (*) indicates a file with unsaved changes.

File Menu

The File menu provides commands for file operations. It has the following commands:

Command	Description
Import <Ctrl> + <I>	Opens a dialog box where you can select an existing mass collection. In addition to the program-specific data files, you can select *.csv or *.txt files that meet the requirements of the program. All available masses are replaced by the masses contained in the imported file. Click **Open** to import the file.
Export <Ctrl> + <S>	Opens a dialog box that you can use to save the active mass collection. In addition to using the program-specific data files, you can save the mass collection as tabulator-separated file (*.txt, for example) or comma-separated file (*.csv, for example). Enter a new file name, select the file type, and select the location (disk and directory) where you want to save it. Alternatively, you can select an existing file. Click **Save** to save the file.

Command	Description
Export selected <Ctrl> + <Shift> + <S>	Opens a dialog box that you can use to save the selected masses. In addition to using the program-specific data files, you can save the mass collection as tabulator-separated file (*.txt, for example) or comma-separated file (*.csv, for example). Enter a new file name, select the file type, and select the location (disk and directory) where you want to save it. Alternatively, you can select an existing file. Click **Save** to save the file. This command is available only when at least one line in the dialog box is selected.
Minimize <Ctrl> + <F4>	Closes the dialog box. Your edits will be retained. Alternatively, click the **Done** button at the right side of the menu bar.

Edit Menu

The Edit menu provides commands for editing the active file. It has the following commands:

Command	Description
Undo until last save <Ctrl> + <Z>	Discards all changes to the active file that have not been saved.
Copy <Ctrl> + <C>	Copies the data of the selected mass to the clipboard.
Replace by clipboard <Ctrl> + <V>	Replaces the available masses by the content of the clipboard. **NOTICE** The clipboard stores lock mass data as tabulator-separated text files. When the data in the clipboard are not in this format, the command is not available. ▲

Help Menu

Displays the Help for this dialog box.

Mass Calculator

Use the Mass Calculator to specify the properties of compounds that are used in the Inclusion List dialog box and the Exclusion List dialog box.

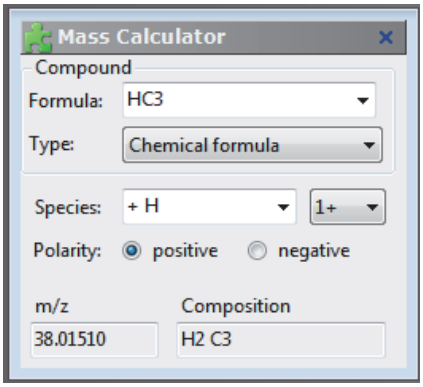


Figure 3-16. Mass Calculator (Method Editor)

When you display the Mass Calculator, its fields are filled with the values of the active list fields, if available. Otherwise, its fields are filled with the default values. The Method Editor applies the set and calculated values of the Mass Calculator to the active list fields, when you exit the editor by doing one of the following:

- Press **Enter** at any position.
- Click at any position outside of the editor.
- Click the down arrow in the Formula field again.

The Mass Calculator has the following parameters:

Parameter	Description
Compound	
Formula	Use the text field to specify the basic composition of the compound. Click the down arrow to display a list of the last entered compounds. The syntax rules and interpretation accept the kind of formula that is also accepted on the Spectrum Simulation page (Isotope Simulation area) of the Qual Browser.
Type	Use the Type list to specify the formula type used in the Formula field: • **Chemical formula** Formula is defined as the resulting elemental composition of the entered formula. This is the default selection. Independent of the entry in the Species field, the elemental composition and the mass of the compound, Formula is completely determined by the given formula and selected formula type.

Parameter	Description
Species	Use this text field to define adducts or modifications of the active compound that are expected to be formed. The default value is +H for positive polarity and –H for negative polarity. The Species field can be used in two ways: • Click the down arrow to display a list of predefined adducts for each polarity: - +H, +NH4, +C2H5, +C3H5, +C4H9 for positive polarity - -H for negative polarity - an empty entry to express adducts (for example, radical cations) for both polarities. The corresponding set will be displayed depending on the selected polarity. Selecting an adduct (A) will result in the strict behavior of applying one unit A to the compound and using "+H" or "-H" adduct depending on the charge state and active polarity. • Enter the modifications of the compound by using squared brackets and at least M as representation of the basic compound. The predefined adducts can be entered, too. This definition is used without additional auto dependencies, like adding protons.
Charge state	Use the list box to select the resulting charge state. Available options are 1+ to 25+ for positive polarity and 1– to 25– for negative polarity. The default charge state is 1+ for positive polarity and 1– for negative polarity.
Polarity	Use the option buttons to select the applied polarity. The default polarity is **positive**.
m/z	This text field displays the calculated mass-to-charge ratio, with five decimal places (for example, 524.26496).
Composition	This text field displays the determined total formula (for example, C23 H38 N7 O5 S).

❖ **To display the Mass Calculator**

Click the down arrow in a Formula cell of the Inclusion List dialog box or the Exclusion List dialog box.

Chapter 4 Explore Q Exactive GC Tune

This chapter provides information about Q Exactive GC Tune, its views, functions, and features.

Contents

Q Exactive GC Tune Overview

The Q Exactive GC Tune program is used to operate the Q Exactive GC mass spectrometer. Figure 4-1 shows Q Exactive GC Tune. To access information about Q Exactive GC Tune, use the title bar, the toolbar, menu commands, display views, and Help.

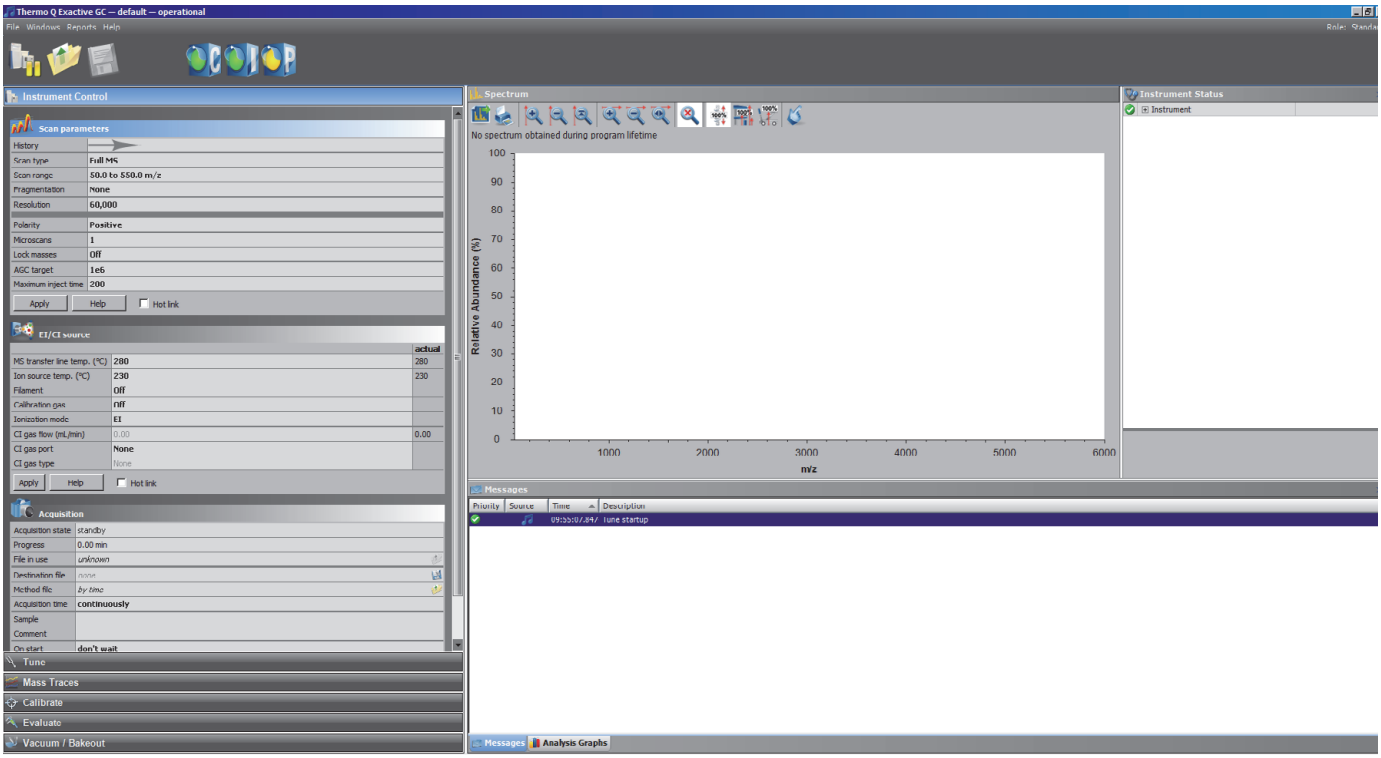


Figure 4-1. Q Exactive GC Tune

❖ **To display this window**

- Choose **Start > Programs > Thermo Exactive Series > Tune**, or
- Click  on the desktop.

NOTICE When you are running Q Exactive GC Tune, an Q Exactive GC Tune icon ( ) is displayed in the Microsoft™ Windows™ system tray. Double-click the icon to display the Q Exactive GC Tune window when it is minimized. Right-click the icon to display a shortcut menu that provides commands for restoring or terminating Q Exactive GC Tune. ▲

Title Bar

The Q Exactive GC Tune title bar displays the name of the program, the name of the current Tune Method, and the instrument status. An asterisk indicates a Tune Method with unsaved changes.

Menus

The Q Exactive GC Tune menu bar displays the available menus. The Q Exactive GC Tune menus are as follows:

- File Menu
- Windows Menu
- Reports Menu
- Help Menu

On the far right side of the menu bar, the active user role is displayed.

Toolbar

The Toolbar provides symbol shortcuts for frequently used commands. It is located below the title bar of the Q Exactive GC Tune window.

Tasks Panel

The tasks panel on the left side of the Q Exactive GC Tune window comprises five windows:

- The Instrument Control Window on the left side of the Q Exactive GC Tune window comprises three windows:
  - Scan Parameters Window for defining a scan depending on the scan mode and scan type combination.
  - EI/CI Source Window for displaying and editing parameters of the EI or CI ion source.
  - Click the EI/CI source window title bar in the Instrument Control window.Acquisition Window for acquiring and storing measurement data.
- Mass Traces Window for selecting mass traces that are displayed in the Analysis Graphs Window.
- Calibrate Window for performing an automatic optimization of the calibration parameters.
- Evaluate Window for performing an automatic check of the instrument precision.
- Vacuum / Bakeout Window for displaying the pressure values at the vacuum gauges and performing an instrument bakeout.

Display Panel

The display panel on the right side of the Q Exactive GC Tune window comprises up to five windows:

- Spectrum Window for displaying a real-time data plot. The spectrum window is always visible.
- Instrument Status Window for displaying instrument parameters.
- Messages Window for displaying current status information about the instrument, the control service, or other programs.
- Analysis Graphs Window for displaying a real-time graph.
- Debug Messages Window for displaying messages that can be used during software development. The debug messages window is not available for standard users.

In addition to these five windows, the display panel may show additional windows for controlling other installed instruments.

Q Exactive GC Tune Menus

The Q Exactive GC Tune window has the following menus:

- File Menu
- Windows Menu
- Reports Menu
- Help Menu

User Role

On the far right side of the menu bar, the active user role is displayed. The user role specifies the number of parameters that are available for the active experiment. Standard User is the user role with the lowest number of available parameters.

Click the entry (Advanced, for example) to display a list that displays the available user roles. To change the active user role, click the respective entry. The number of available user roles depends on the installed licenses. See "License Dialog Box" on page 4-49.

File Menu

The File menu provides commands for file and program operations. It has the following commands:

Command	Description
Load Tune File	Displays a dialog box that you can use to find and open a tune file (*.mstune) that already exists.
Save Tune File	Saves the active tune file with the current settings.
Save Tune File as	Opens a dialog box that you can use to enter a new file name and to select the location (disk and directory) where you want to save it. Click **Save** to save the tune file with the current settings. **NOTICE** An * appears next to the tune file name displayed in the title bar of tune if the current tune file has been modified and not saved. Any changes made to the tune file must be saved before attempting to use the new setting from an acquisition that loads the tune file in a method.

Command	Description
Export System Settings	Opens a dialog box that you can use to save the current system settings (*.ini) to be imported on another computer that has no instrument attached and is intended for method editing. This computer has to have installed all needed software including Thermo Platform, Thermo Xcalibur and all instrument drivers installed. **NOTICE** Imported values will be overwritten if an instrument will be attached later. ▲
Terminate	Closes the active window. If the active tune file contains unsaved changes, a dialog box warns about losing data when you terminate the program.

Windows Menu

The Windows menu provides commands for customizing the information displayed in the Q Exactive GC Tune window. The system highlights the icon to the left of a command if the window is active. Deselect the command to hide the window.

The Windows menu has the following commands:

	Command	Description
	View > Show all	Displays all windows of Q Exactive GC Tune.
	Tooltips	Displays a short description of an item on the Q Exactive GC Tune window when you rest the mouse pointer over it.
	Spectrum	Displays the Spectrum window. The Spectrum Window is always visible.
	Instrument Status	Displays/Hides the Instrument Status window.
	Messages	Displays/Hides the Messages window.
	Analysis Graphs	Displays/Hides the Analysis Graphs window.
	Debug Messages	Displays/Hides the Debug Messages window. The debug messages window is not available for standard users.

	Command	Description
	Mass Calculator	Displays the Mass Calculator, which helps you to calculate the mass of an compound of interest.
	Installed instruments	Depending on the individual configuration of your system, additional commands may allow displaying windows for controlling other installed instruments (TriPlus RSH autosampler, for example.)

NOTICE Q Exactive GC Tune may open some windows (the analysis graphs window, for example) without user interaction if new important information is available. ▲

Reports Menu

The Reports menu provides access to various calibration reports. The individual commands are available only when the respective reports have been created. If no reports exist (in a new system, for example), the Reports menu is shown grayed out and has no commands.

The Reports menu has the following commands:

Command	Description
Calibration > Latest	Opens the latest complete calibration report in PDF format.
Calibration > List	Displays the list of up to five (5) complete calibration reports. To open a report, double-click any of the calibration from the list.
Spectral Mass Calibration (neg) > Latest	Opens the latest negative mode calibration report in PDF format.
Spectral Mass Calibration (neg) > List	Displays the list of up to five (5) negative mode calibration reports. To open a report, double-click any of the spectral mass calibration from the list.
Spectral Mass Calibration (pos) > Latest	Opens the latest positive mode calibration report in PDF format.

Command	Description
EI Tune Report	The choices are Latest and List. Latest displays the most recent EI Tune report. List displays the list of up to five (5) reports. To open a report, double-click any of the tune reports from the list.
Spectral Mass Calibration (pos) > List	Displays the list of up to five (5) positive mode calibration reports. To open a report, double-click any of the spectral mass calibration from the list.
All Reports	Opens a file manager window with a list of all previously acquired spectral mass calibration reports.

Help Menu

The Help menu groups commands that provide information about Q Exactive GC Tune. It has the following commands:

Command	Description
Help Overview	Displays Q Exactive GC Tune Help.
Help Content	Displays the table of contents for Q Exactive GC Tune Help.
Help Index	Displays the index for Q Exactive GC Tune Help.
About	Displays the About dialog box with information about the instrument and the current Q Exactive GC Tune version.

Toolbar

The Q Exactive GC Tune toolbar provides symbol shortcuts for frequently used commands. It is located below the title bar of the Q Exactive GC Tune window. To activate a toolbar function, click the corresponding toolbar button.

Some symbols provide information about the current statuses of respective instrument components either by their color or by displaying a tooltip when you rest the mouse pointer on them.

The following functions are available:

Button		Description
	General instrument state On / Standby / Off Run by Xcalibur	Click to toggle between the instrument operating statuses On, Standby, and Off. The button reflects the instrument status: • When the mass spectrometer is On, Q Exactive GC Tune displays  . • When the mass spectrometer is in Standby, Q Exactive GC Tune displays  . • When the mass spectrometer is Off, Q Exactive GC Tune displays  . If the mass spectrometer is controlled by an Xcalibur sequence, Q Exactive GC Tune displays  . The acquisition can only be stopped from Xcalibur Sequence Setup View.
	Open	Click to display a dialog box that you can use to find and open a *.mstune file that already exists. Alternatively, choose **File > Load Tune File**.
	Save	Click to open a dialog box that you can use to select the location (disk and directory) where you want to save the tune file. Alternatively, choose **File > Save Tune File as**. This button is not available when the settings of the active tune file are not changed.

Button		Description
	Communication Status	Shows the actual communication status of the system: • Green: communication with instrument is OK. • Yellow: only service is accessible (no instrument). • Red: communication is broken (no instrument, no service).
	Hardware Status	Shows the actual hardware status of the system (top instrument tree state): • Green: all readbacks are in specifications (green hooks). • Red: one or more readbacks are out of range.
	Performance Status	Shows the actual performance status of the system: • Green: the last evaluation/calibration was successful. • Yellow: the last evaluation/calibration was successful, but is out of date. • Red: the evaluation/calibration was not successful. By default, the performance status icon turns yellow 25 hours after the last successful mass calibration or check. With exception of standard users, users can change this value in the System node of the instrument status window according to their mass accuracy requirements.
	Procedure active	If a procedure is active (for example, system bakeout or tune) Q Exactive GC Tune displays an animated icon next to the performance status button.
	Acquisition indicator	If the mass spectrometer is acquiring data, Q Exactive GC Tune displays an animated icon next to the performance status button.

Tasks Panel

Use the windows of the tasks panel to perform procedures that maintain the quality of measurements with the Q Exactive GC mass spectrometer.

The tasks panel is always visible. The windows in the tasks panel can be minimized or maximized. Click the title bar of an individual window to display it.

- Instrument Control Window
- Mass Traces Window
- Calibrate Window
- Evaluate Window
- Vacuum / Bakeout Window

Instrument Control Window

Use the windows of the Instrument Control window to enter individual physical settings and to acquire scans. The following windows are available:

- Scan Parameters Window
- EI/CI Source Window
- Click the EI/CI source window title bar in the Instrument Control window.Acquisition Window

The Instrument Control window is always visible. Click the title bar of an individual window to display it. Click again to hide it. To change the order of windows within the Instrument Control window, drag individual windows by their title bars to the new places.

Scan Parameters Window

Use the Scan Parameters window to define a scan depending on the selected scan mode and scan type combination. See Figure 4-2. The Q Exactive GC mass spectrometer updates the scan parameters only after you click **Apply** or select the Hot link check box.

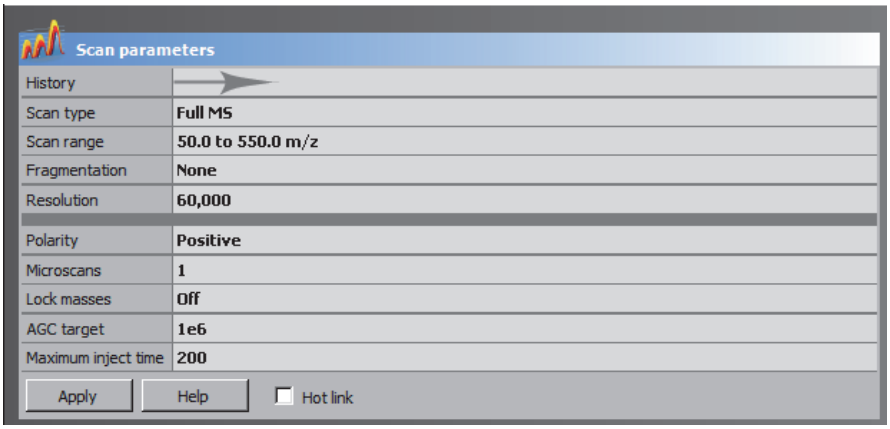


Figure 4-2. Scan Parameters window

The Scan Parameters window has the following parameters:

Parameter	Description
History	Click to display the Scan Parameters History dialog box. Here, select from a list of recent scans based on short scan descriptions. The maximum number of list items is 100. After a change of parameters, the history is updated when you click **Apply** or when you change the status of the Hot link check box.
Scan Type	Displays the current scan type. To change the settings, click into the field to display the Scan Type dialog box.
Scan Range	Displays the current scan range. To change the settings, click into the field to display the Scan Range dialog box.
Fragmentation	Displays the current settings for HCD fragmentation. If no fragmentation type is selected, the text field shows *None*. To change the settings, click into the field to display the Fragmentation dialog box.
Resolution	Use this list box to select the mass resolution for FTMS analysis. The mass resolution of the Orbitrap analyzer is proportional to 1/sqrt(*m/z*). The scan time increases with increasing resolution and detect time. Available options are 15000, 30000, 60000, or 120000. Resolution is calculated according to the IUPAC peak width definition. The available settings represent m/Δm or the *m/z* value of an ion at *m/z* 200 divided by the full width at half its maximum height (FWHM).
Polarity	Use this list box to toggle between positive ion and negative ion polarity.

Parameter	Description
Microscans	Use this spin box to select the number of microscans to be performed. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, you can enter a value in the spin box text field. The valid range is from 1 to 10 microscans.
Lock masses	Displays the active lock masses. During a scan, the Q Exactive GC mass spectrometer uses only the lock masses with a polarity that matches the ion mode. If no lock masses are selected, the text field shows *Off*. To change the settings, click into the field to display the Lock Masses dialog box. **NOTICE** Q Exactive GC Tune allows using active lock masses in user-defined calibration mass lists. ▲
AGC target	Use this list box to select the AGC target value. The AGC target value controls the number of ions that are injected into the Orbitrap analyzer. Available options are 2e4, 5e4, 1e5, 2e5, 5e5, 1e6, 3e6, and 5e6.
Maximum inject time	Use this list box to type or click a maximum injection time for AGC. For example, type **160**. The valid range is from 1 to 3000 milliseconds.
Buttons	
Apply	Sends all changes to the instrument.
Help	Displays the Help for this window.
Hot Link	Select the Hot link check box to allow Q Exactive GC Tune to send any changes immediately to the instrument. A green frame around the parameter box indicates an active hot link.

❖ **To display this window**

Click the Scan Parameters window title bar in the Instrument Control window.

EI/CI Source Window

Use the EI/CI source window to specify electrical ionization (EI) or chemical ionization (CI) source parameters. The Q Exactive GC mass spectrometer updates the EI/CI source parameters only after you click **Apply** or with the Hot link check box being selected. See Figure 4-3.

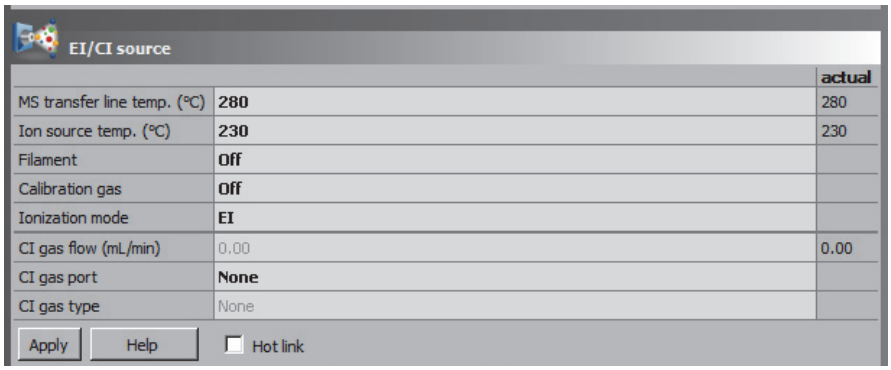


Figure 4-3. EI/CI source window

The EI/CI source window has the following parameters:

Parameter	Description
MS Transfer Line Temp	Displays the temperature (in °C) for the part of the transfer line that's inside the vacuum manifold. To change the MS transfer line temperature, enter a new temperature into the box. Temperatures from 200 – 280 °C are recommend for most applications.The mass spectrometer changes the MS transfer line temperature when you click **Apply** or with the Hot link check box selected. The MS Transfer Line Temp readback is to the right of the spin box. This readback displays the actual flow rate (in °C).
Ion Source Temp	Displays the ion source temperature. Temperatures from 200 – 350 °C are recommend for most applications. To change the ion source temperature, enter a new temperature into the box.The mass spectrometer changes the ion source temperature when you click **Apply** or with the Hot link check box selected. The Ion Source Temp readback is to the right of the spin box. This readback displays the actual flow rate (in °C).
Filament	Displays whether the filament is turned on or off. The choices are **On** or **Off** and can be set in the drop-down menu. source. To preserved the filament lifetime, set to off when not using the mass spectrometer. The mass spectrometer changes the Filament setting when you click **Apply** or with the Hot link check box selected.
Calibration Gas	Displays whether the calibration gas is turned on. The choices are **On** or **Off** and can be set in the drop-down menu. source. To preserved the calibration gas, set to off when not running an analysis or checking for leaks on the mass spectrometer. The mass spectrometer changes the Calibration Gas setting when you click **Apply** or with the Hot link check box selected.

Parameter	Description
Ionization Mode	Displays either EI or CI ionization mode depending on the ion source installed in the Q Exactive GC mass spectrometer. The CI or EI/CI ion volume must be installed in the ion source cartridge to use CI ionization mode. Also, the mode must be set to CI to turn on CI gas. The mass spectrometer changes the ionization mode when you click **Apply** or with the Hot link check box selected.
CI Gas Flow	Displays the CI gas flow rate (in mL/min). To change the CI gas flow rate, enter a value in the spin box text field. The mass spectrometer changes CI gas flow rate when you click **Apply** or with the Hot link check box selected. The CI Gas Flow readback is to the right of the spin box. This readback displays the actual flow rate (in mL/min) of the CI gas.
CI Gas Port	Displays the port connected to the CI gas. The choices are **Port A** and **Port B** if CI is selected as the ionization mode. If EI is the selected ionization mode the field is automatically set to **None**. To change the CI gas port, select Port A or Port B depending on which port is attached to the CI gas. The mass spectrometer changes the CI gas port when you click **Apply** or with the Hot link check box selected.
CI Gas Type	Displays the type of CI gas to be used for the analysis. Selection options are **Methane**, **Isobutane**, **Ammonia**, **Carbon Dioxide**, or **Other**. Use the spin box to select one of the choices. The mass spectrometer changes the CI gas type when you click **Apply** or with the Hot link check box selected.
Buttons	
Apply	Sends all changes to the instrument.
Help	Displays the Help for this window.
Hot Link	Select the Hot link check box to allow Q Exactive GC Tune to send any changes immediately to the instrument. A green frame around the parameter box indicates an active hot link.

❖ **To display this window**

Click the EI/CI source window title bar in the Instrument Control window.**Acquisition Window**

Use the Acquisition window to enter parameters for acquiring and storing scan data as well as to monitor the progress of the active acquisition.

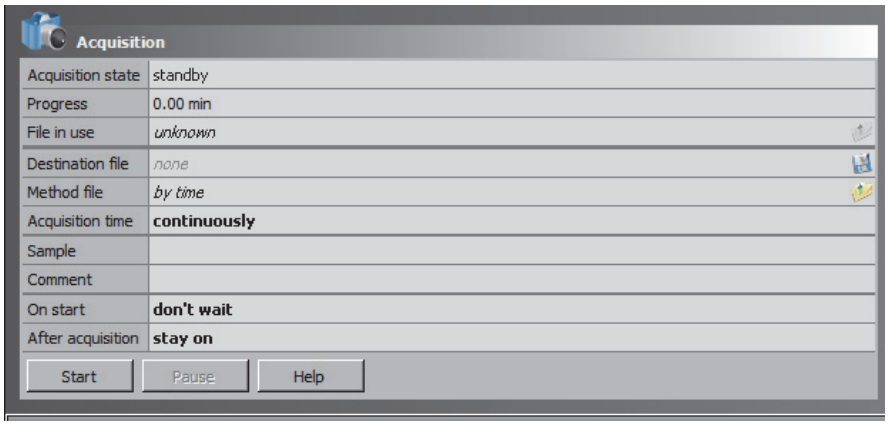


Figure 4-4. Acquisition window

The Acquisition window has the following parameters:

Parameter	Description
Acquisition state	Displays the status of the current acquisition.
Progress	Displays the elapsed time of the current acquisition.
File in use	Displays the name of the raw file to be acquired to disk for the current sample. To display the file in Qual Browser, click the button. This value is up to date even when Q Exactive GC Tune is acquiring data under control of Xcalibur.
Destination file	Displays the full path of the folder where your *.raw files will be saved. To change the path, type the full path (Drive:\path) in the text box or click the button to the right of the text box to browse your directories and select the folder where your *.raw files should be saved. **NOTICE** Saving raw files on network drives typically causes problems. Preferably select a local path (for example, C:\Xcalibur\data). ▲

Parameter	Description
Method file	Displays the name of the instrument method (*.meth) to be used during the current acquisition. The tune file used in that instrument method will be used for your scan but other devices configured in that instrument method will not be included. Type in the name of the instrument method, or click the  button to the right of the text box to browse your computer for an instrument method file (typically saved in C:\Xcalibur\methods). When the active acquisition is controlled by the settings of the parameter Acquisitions time, this parameter is not available. Instead, the text field shows *by time*.
Sample	Displays the name of the current sample. To change the sample name, type the new name in the text field.
Comment	Displays a comment on the current sample, if available. To change the comment, type the new name in the text field.
Acquisition time	Displays the duration of the acquisition time. Use the option buttons to select one of the following settings: • Continuously The acquisition, once it is started, will continue until you stop (**Stop** button) or pause (**Pause** button) it. • Scans Specify the number of scans for the current acquisition. You can set the number of scans to any value from 1 to 10000. To change this value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, enter a value in the spin box text field. • Minutes Specify the time in minutes for the current acquisition. You can set the acquisition time from 0.01 to 15000.00 minutes. To change this value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, enter a value in the spin box text field. When the active acquisition is controlled by a method file, this parameter is not available. Instead, the text field shows *by method*.
On start	Use the list box to set the start mode of the data acquisition. The options are as follows: • don't wait Initiates acquisition of the raw file immediately when you click **Start** in the Acquisition window. • wait for contact closure When you click **Start** in the Acquisition window, the acquisition pauses. Q Exactive GC Tune initiates acquisition of the raw file either when it receives a contact closure signal (for example, from an analog autosampler) or when you click **Resume**.

Parameter	Description
After acquisition	Use the list box to set the instrument mode after the data acquisition has stopped. The options are as follows: • stay on The instrument remains On when the current acquisition is completed. • enter standby mode Sets the instrument to Standby when the current acquisition is completed. • switch off Switches Off the instrument when the current acquisition is completed.
Buttons	
Start / Stop	Allows starting and stopping data acquisition. When no acquisition is in progress, Q Exactive GC Tune displays **Start**. When an acquisition is in progress, Q Exactive GC Tune displays **Stop**. To start an acquisition, click **Start**. To stop an acquisition, click **Stop**. To pause an acquisition, click the **Pause / Resume** button. **NOTICE** No Q Exactive GC Tune controlled data acquisition is possible if the instrument is controlled by Xcalibur or TraceFinder. ▲
Pause / Resume	Allows pausing and resuming an acquisition in progress. When no acquisition is in progress, the button is inactive (gray). When an acquisition is in progress, Q Exactive GC Tune displays **Pause**. When an acquisition is paused, Q Exactive GC Tune displays **Resume**. To pause an acquisition in progress, click **Pause**. To resume an acquisition that is paused, click **Resume**. To stop an acquisition in progress, click the **Start / Stop** button. **NOTICE** The acquisition can be started by pressing **Resume** if the mass spectrometer is waiting for contact closure. ▲
Help	Displays the Help for this window.

❖ **To display this window**

Click the Acquisition window title bar in the Instrument Control window.

Tune Window

Use the Tune window to d automatically tune the Q Exactive GC mass spectrometer window. A daily tune and calibration are suggested for the Q Exactive GC instrument. Select AutoTune to use the last tuned values. See Figure 4-6.

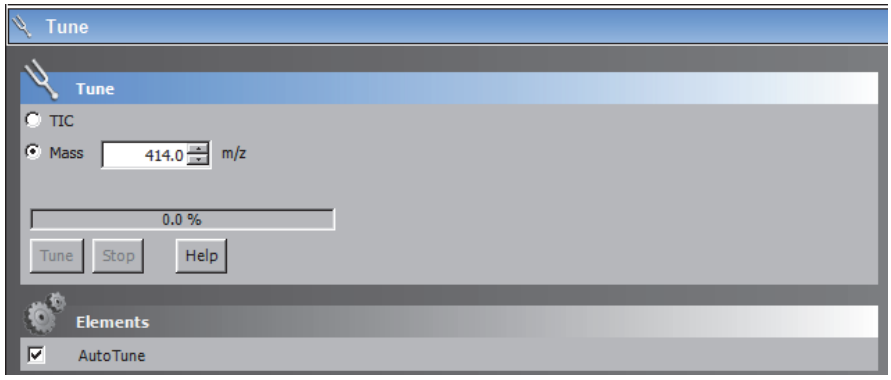


Figure 4-5. Tune window

NOTICE The calibration gas and filament must be turned on before tuning the Q Exactive GC instrument. ▲

The Tune window has the following parameters:

Parameter	Description
Tune	
TIC	Select this check box to tune on the total ion current.
Mass	Select an *m/z* value that you want to tune on. You can set the mass-to-charge ratio to any value from *m/z* 30 to *m/z* 3000. To change this value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, enter a value in the spin box text field.
Elements	Check **AutoTune** to use the last tuned values. In advanced user mode, you can leave AutoTune unchecked and change individual tune values in **Instrument Status > Tune Values**. If a cleaned source is inserted in the instrument, check **Preset Defaults** to tune to the original Tune default values.
Buttons	
Start	Click **Start** to start tuning the instrument. The instrument needs to be in On status to start a plotting procedure. When tuning is in progress, the Q Exactive GC Tune displays **Stop**.
Stop	Click **Stop** to stop tuning the Q Exactive GC.
Help	Displays the Help for this window.

Mass Traces Window

Use the Mass Traces window to display mass traces for the total ion current and up to five *m/z* values in the Analysis Graphs window. See Figure 4-6. You can plot mass traces when manually adjusting source parameters such as gas flows. You can also optimize the collision energy.

NOTICE Make sure to tune your instrument for both ion modes if you want to use a switching method! Settings for both polarity modes are saved in the same tune file! ▲

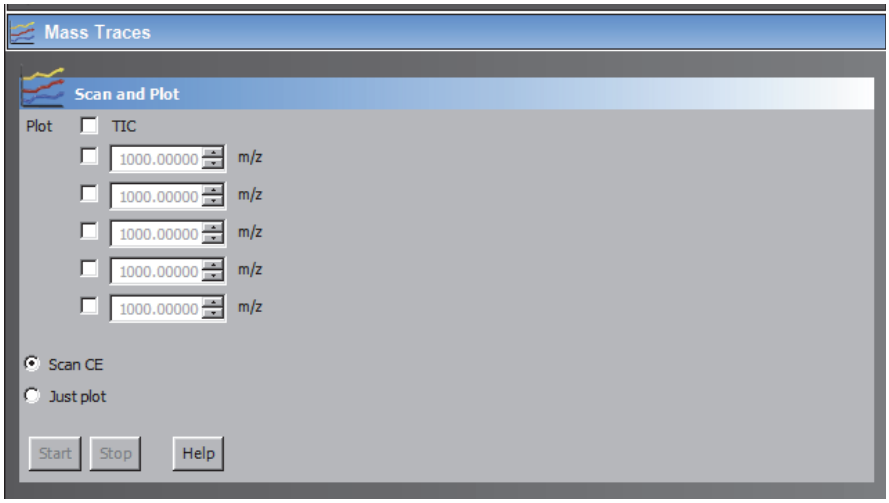


Figure 4-6. Mass Traces window

The Mass Traces window has the following parameters:

Parameter	Description
Plot	
TIC	Select this check box to plot the total ion current.
Masses to plot	Select a check box for each *m/z* value that you want to plot (up to five). When you select a check box, the spin box becomes active. The spin box displays the mass-to-charge ratio to plot. You can set the mass-to-charge ratio to any value from *m/z* 30 to *m/z* 3000. To change this value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, enter a value in the spin box text field.
Scan CE	Select this option button to optimize the collision energy on the total ion current.

Parameter	Description
Just plot	Select this option button to plot the mass trace without optimization.
Buttons	
Start	Click **Start** to start plotting the mass traces. The instrument needs to be in On status to start a plotting procedure. When plotting is in progress, the Q Exactive GC Tune displays **Stop**.
Stop	Click **Stop** to stop plotting the mass traces.
Help	Displays the Help for this window.

Calibrate Window

Use the Calibrate window of the tasks panel to perform an automatic optimization of the calibration parameters. See Figure 4-7. Calibration parameters are instrument parameters whose values do not vary with the type of experiment.

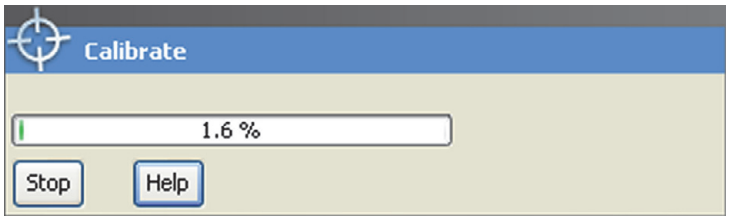


Figure 4-7. Calibrate window

Standard users can choose the ion mode they want to calibrate in the Calmix Calibration window. Advanced users can use the Calmix Calibration window to calibrate the instrument with the standard calibration gas. Alternatively, they can use the Customized Calibration window to enter parameters for a calibration with ions of their choosing.

NOTICE If the system was in Off mode before, it is necessary to put the instrument into On mode for at least 90 minutes before a mass calibration is performed. ▲

The calibrating procedure requires that you introduce calibration gas into the mass spectrometer at a steady rate.

The Calibrate window has the following parameters:

Parameter	Description
Progress bar	Displays the elapsed percentage of the current calibration procedure.
Buttons	

Parameter	Description
Calibrate	Click **Calibrate** to start an automatic calibration of the mass spectrometer. The instrument needs to be in On status to start a calibration. The duration of the automatic calibration depends on the selected check boxes; a complete automatic calibration requires about four minutes. When calibration is in progress, Q Exactive GC Tune displays **Stop**.
Stop	Click **Stop** to stop a calibration in progress.
Help	Displays the Help for this window.

❖ **To display this window**

Click  in the tasks panel.

Calmix Calibration Window

Use the Calmix Calibration window to select parameters and ion modes when performing an automatic calibration. See Figure 4-8 and Figure 4-9. The window displays the current statuses of the individual calibration items on the right side.

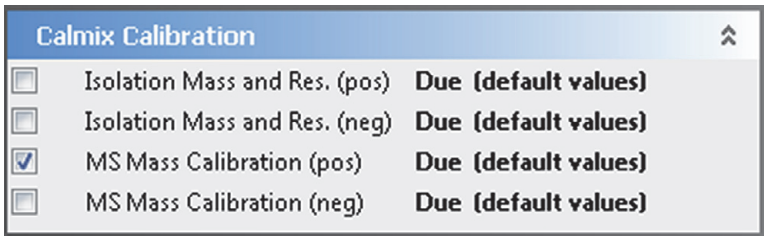


Figure 4-8. Calmix Calibration window (for standard users)

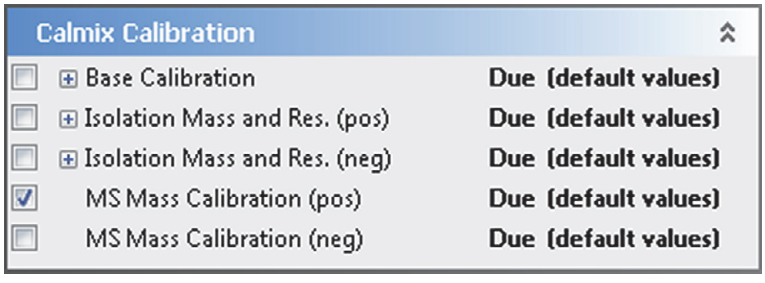


Figure 4-9. Calmix Calibration window (for advanced users)

NOTICE Select **MS Mass Calibration (pos)** for daily calibration in EI mode.

The Calmix Calibration window has the following parameters:

Parameter	Description
Base Calibration	The Base Calibration item displays a hierarchical list of items in a tree view. Here, you can select individual parameters that you want to calibrate. If a check box is not selected, no subentry is selected. If the check box is selected, all subentries are selected. If the check box is filled, some subentries are selected. This parameter is not available for standard users.
Isolation Mass and Res. (pos) Isolation Mass and Res. (neg)	Select this check box to perform an automatic calibration for isolation mass and resolution of the quadrupole in the respective ion mode. In a hierarchical list of items in a tree view, advanced users can select individual parameters that they want to calibrate. If the check box is not selected, no subentry is selected. If the check box is selected, all subentries are selected. If the check box is filled, some subentries are selected.
MS Mass Calibration (pos)	Select this check box to perform an automatic calibration for all calibration parameters in the positive ion mode. Use methane reagent gas for positive ion calibration in CI mode.. Refer to the *Q Exactive GC Operating Manual* for information.
MS Mass Calibration (neg)	Select this check box to perform an automatic calibration for all calibration parameters in the negative ion mode. Use negative ion chemical ionization for calibrations in negative mode. Methane is recommended as the optimal reagent gas for automated calibrations and evaluations. Refer to the *Q Exactive GC Operating Manual* for information.

Customized Calibration Window

Use the Customized Calibration window to perform a mass calibration with user-defined ions. Enter the *m/z* values on which to calibrate. See Figure 4-10. This window is not available for standard users.

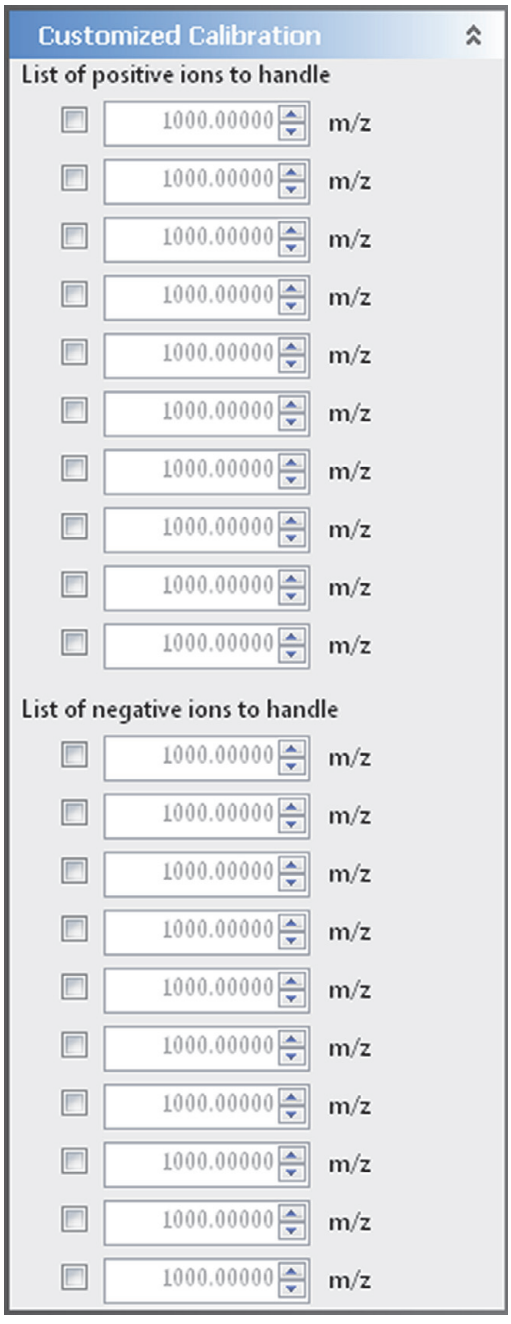


Figure 4-10. Customized Calibration window

NOTICE Use this window for (positive and negative) mass calibration only! For all other calibration procedures, use the standard calibration ions! ▲

The Customized Calibration window has the following parameters:

Parameter	Description
Masses to calibrate	Select a check box for each *m/z* value that you want to plot (up to ten for each ion mode). When you select a check box, the spin box becomes active. The spin box displays the mass-to-charge ratio to plot. You can set the mass-to-charge ratio to any value from *m/z* 30 to *m/z* 3000. To change this value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, enter a value in the spin box text field.

Managing Mass Calibrations

If you want to disable a calibration from the Tune software, do the following.

First, go to **Instrument > Control > Mass Calibration Data > Higher Order Shape**, right click the **Spec. shape[x]** cell with the values you no longer want to use and select **Disable This Set**. See Figure 4-11.

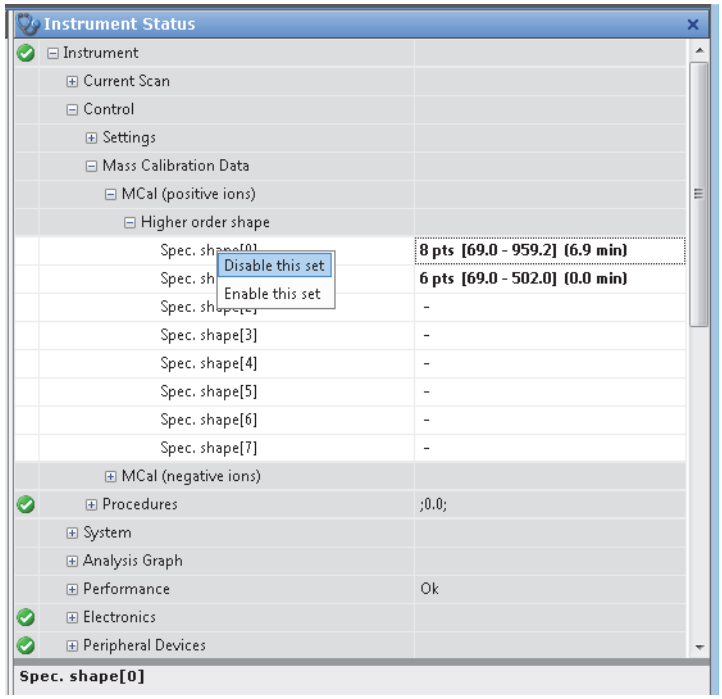


Figure 4-11. Disabling Spectral Values

The calibration is now disabled in the software. The disabled set of calibration points is no longer used to assign mass to measured ion frequencies. See Figure 4-12.

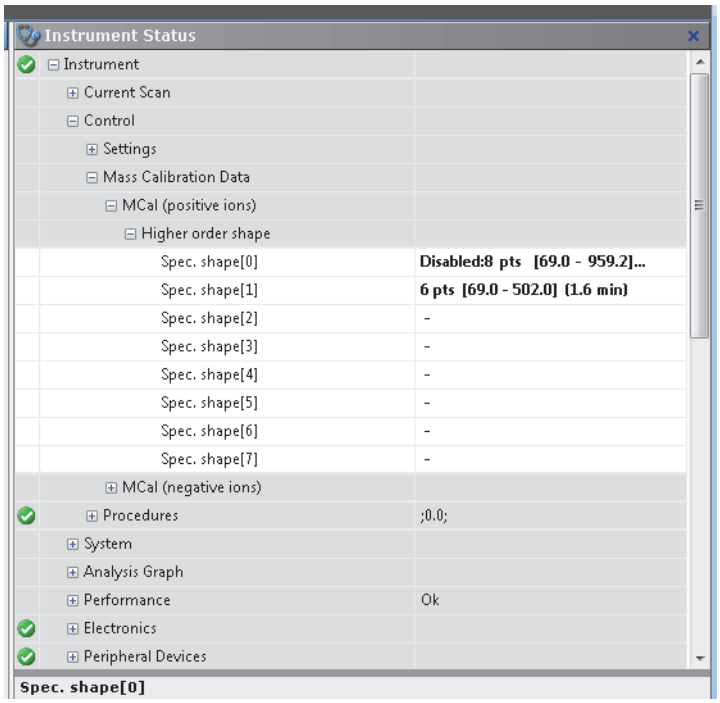


Figure 4-12. Disabling Spectral Values

To remove older calibrations from the software, right click on the disabled **Spec. shape[x]** cell, and choose **Remove All but Last Calibration**. See Figure 4-13.

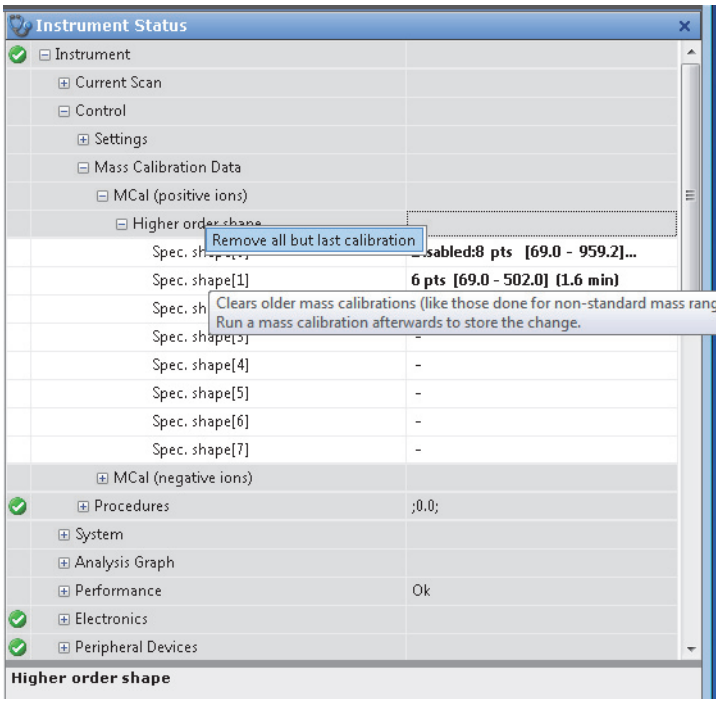


Figure 4-13. Removing Older Calibrations

The older calibrations no longer appear in the tune software. See Figure 4-14.

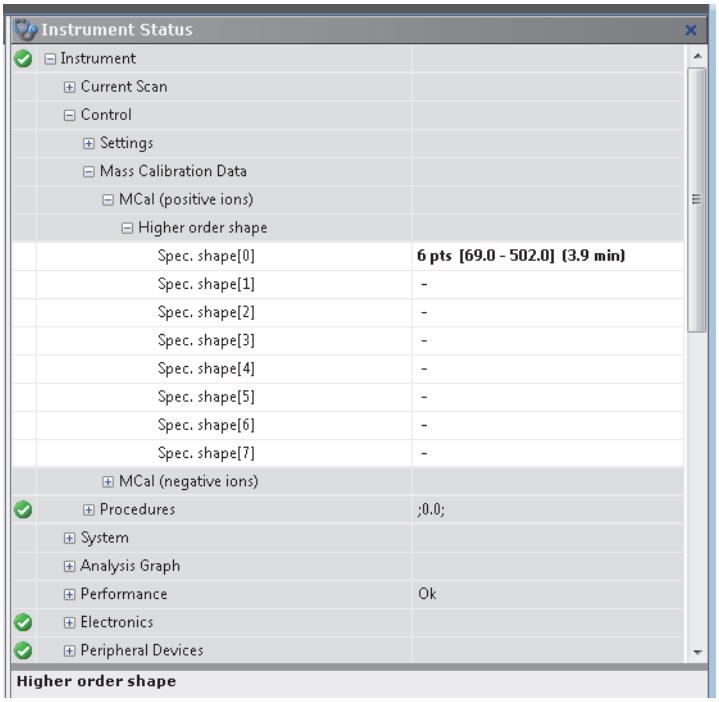


Figure 4-14. Tune Software with Older Calibrations Removed

Run a mass calibration with older values removed to store the change in the software.

High Mass Calibration

To manually add high mass points to your Calibration in Tune, first tun an injection with a compound or compounds containing the high mass points you want add to your calibration.

Next, go to **Advanced** user setting in Tune and set **Instrument > Settings > EI/CI Ion Source > Manual High Mass Calibration** and set **Include with Mass Calibration** to **Yes**. See Figure 4-15.

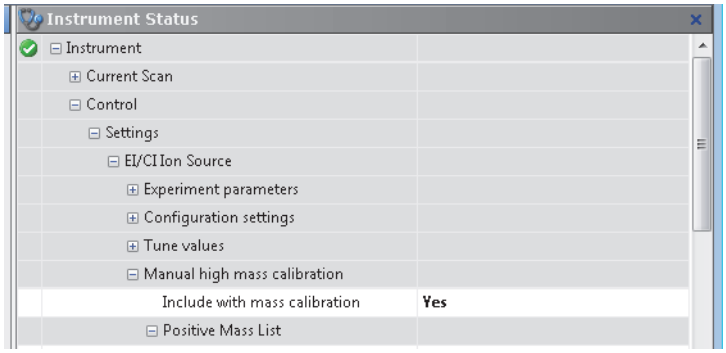


Figure 4-15. Enabling Manual High Mass Calibration

In the **Instrument Status** panel, go to **Instrument > Settings > EI/CI Ion Source > Positive** (or **Negative**) **Mass List** and enter the theoretical and observed masses for each acquired compound. See Figure 4-16.

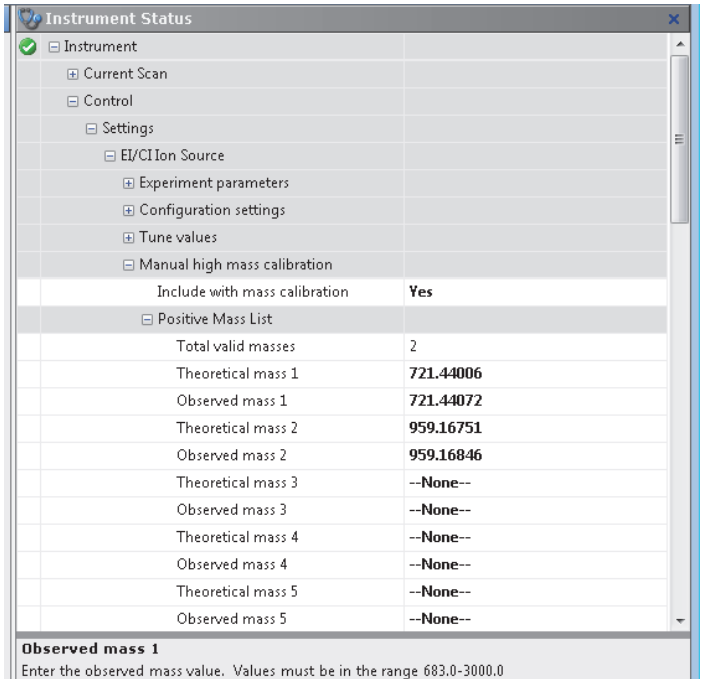


Figure 4-16. Entering Theoretical and Observed Masses

Run the mass calibration as usual. Check the calibration report to ensure that the high masses were added. See Figure 4-17.

Manual High Mass List Added:
Theo mass: 721.44006 observed mass: 721.44072 frequency: 541.7813
Theo mass: 959.16751 observed mass: 959.16846 frequency: 469.870468

Procedure result: passed (rms = 0.05/0.66 ppm)

Calibration Reagent: _____

Lot Number: _____

Expiration Date: _____

Figure 4-17. Calibration Report with High Masses Added

If you want to run a calibration without the high mass values, but want to save the values in the software, go to **Instrument > Settings > EI/CI Ion Source > Manual High Mass Calibration** and set **Include with Mass Calibration** to **No**. See Figure 4-18.

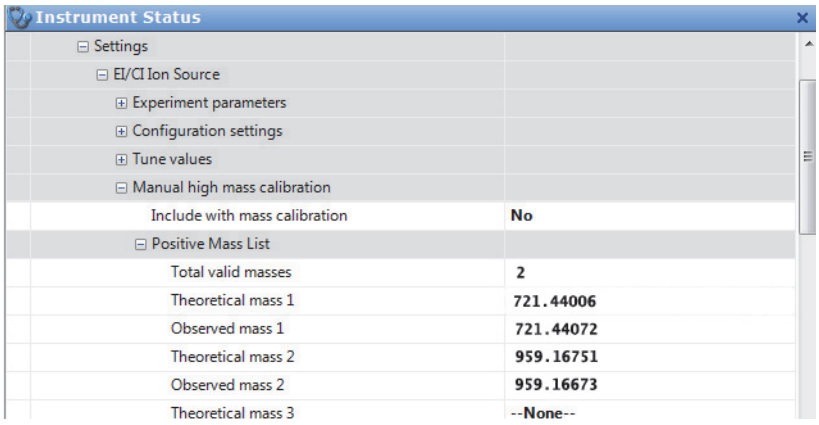


Figure 4-18. Calibration Report with High Masses Added

Evaluate Window

Use the Evaluate window of the tasks panel to perform an automatic check of the instrument calibration. See Figure 4-19.

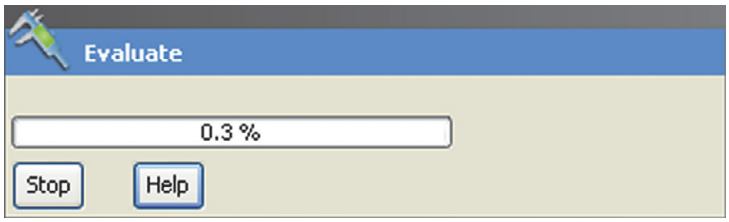


Figure 4-19. Evaluate window

To enable an evaluation, standard users must select at least one check box in the Calmix Evaluation window. Advanced users must select at least one check box either in the Calmix Evaluation window or in the Customized Evaluation window.

The evaluation procedure requires that you introduce calibration gas into the mass spectrometer for at least 30 seconds. The filament must be on before starting the evaluation procedure.

The Evaluate window has the following parameters:

Parameter	Description
Progress bar	Displays the elapsed percentage of the current evaluation procedure.
Buttons	
Evaluate	Click **Evaluate** to start an automatic evaluation of calibration parameters. The instrument needs to be in On status to start an evaluation. When evaluation is in progress, Q Exactive GC Tune displays **Stop**.
Stop	Click **Stop** to stop an evaluation in progress.
Help	Displays the Help for this window.

Calmix Evaluation Window

Use the Calmix Evaluation window to enable the evaluation with the standard calibration gas ions.

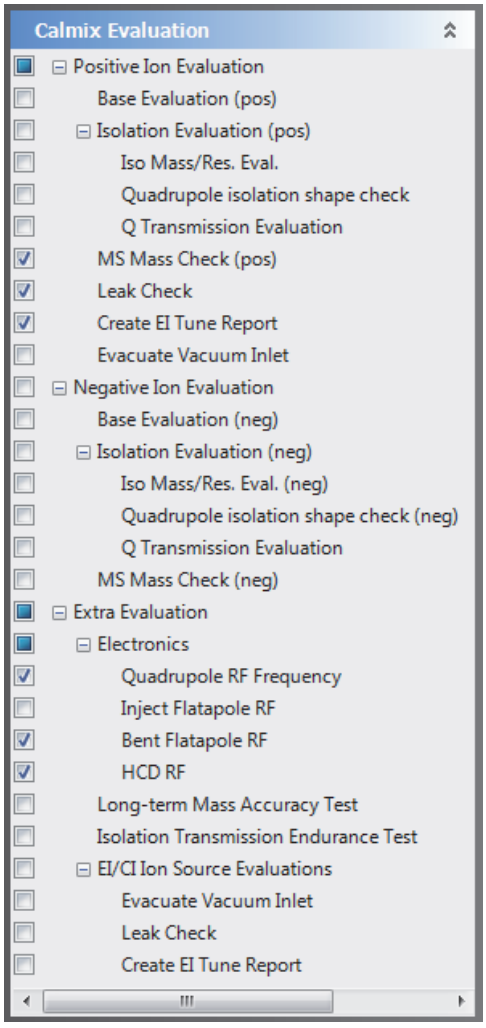


Figure 4-20. Calmix Evaluation window (for advanced users)

The Calmix Evaluation window has the following parameters:

Parameter	Description
In a hierarchical list of items in a tree view, you can select individual parameters that you want to evaluate. If the check box is not selected, no subentry is selected. If the check box is selected, all subentries are selected. If the check box is filled, some subentries are selected.	
Positive Ion Evaluation / Negative Ion Evaluation	
Select the check boxes to evaluate various parameters for the respective ion mode.	
-Base Evaluation	Evaluates the basic performance of the instrument.
-Isolation Evaluation	Evaluates the performance of the quadrupole.

Parameter	Description
--Iso Mass/Res. Eval.	Evaluates the instrument performance for quadrupole isolation and resolution. This parameter is not available for standard users.
--Q Transmission Evaluation	Evaluates the quadrupole transmission against minimum requirements. This evaluation requires a calibrated quadrupole and a stable calibration gas signal. This parameter is not available for standard users.
--Quadrupole Isolations Shape Check	Evaluates the quadrupole isolation window for different isolation widths. This parameter is not available for standard users.
-MS Mass Check	Evaluates the *m/z* accuracy of the Orbitrap analyzer.
-Leak Check	Evaluates the oxygen detected in background relative to oxygen introduced with the calibration gas reference leak.
-EI Tune Report	Scans positive EI calibrant ions using the current tune parameters and makes a report. The EI Tune Report may be opened and viewed from the Reports menu.
-Evacuate Vacuum Inlet	Evacuates the vacuum interlock for 30 seconds.
Extra Evaluation	
Select the check boxes to evaluate various parameters that are not contained in the standard evaluations.	
Electronics	Evaluates the performance of electronic components.
-Quadrupole RF Frequency	Checks the quadrupole RF voltage. This parameter is not available for standard users.
-Inject Flatapole RF	Checks the inject flatapole RF voltage. This parameter is not available for standard users.
-Bent Flatapole RF	Checks the bent flatapole RF voltage. This parameter is not available for standard users.
-HCD RF	Checks the RF voltage of the collision cell. This parameter is not available for standard users.
Long-term Mass Accuracy Test	Performs a mass accuracy test over an extended period. This parameter is not available for standard users.
Isolation Transmission Endurance Test	Tests the behavior of the mass isolation optic elements under permanent ion load. The procedure requires a stable calibration gas signal with TIC > 5.0E+07; it will run for about 16 minutes. This parameter is not available for standard users.

Customized Evaluation Window

Use the Customized Evaluation window to enter the *m/z* values on which to evaluate by using a user-defined calibrant. This window is not available for standard users.

NOTICE Not all calibrations or evaluations will use this customized list. ▲

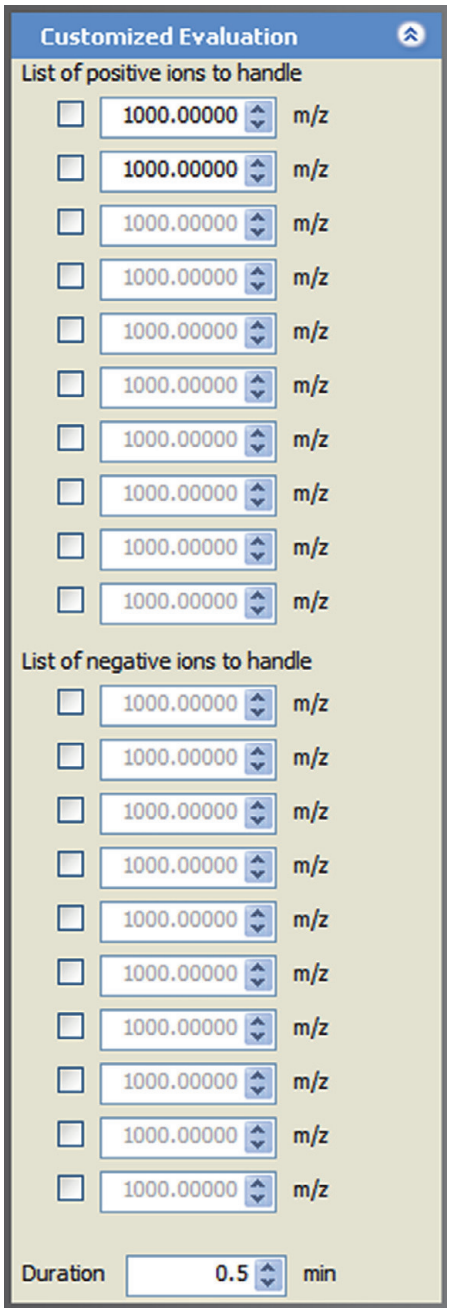


Figure 4-21. Customized Evaluation window

The Customized Evaluation window has the following parameters:

Parameter	Description
Masses to evaluate	Select a check box for each *m/z* value that you want to plot (up to ten for each ion mode). When you select a check box, the spin box becomes active. The spin box displays the mass-to-charge ratio to plot. You can set the mass-to-charge ratio to any value from *m/z* 30 to *m/z* 3000. To change this value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, enter a value in the spin box text field.
Duration	The spin box displays the duration (in minutes) of the evaluation procedure. You can set the duration to any value from 0.2 to 2160 minutes. The default duration is 0.5 minutes. To change this value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. Alternatively, enter a value in the spin box text field.

❖ **To display this window**

Click 

in the Tasks window.

Vacuum / Bakeout Window

The Vacuum / Bakeout window of the tasks panel reads back the pressure values at the vacuum gauges and allows performing an instrument bakeout.

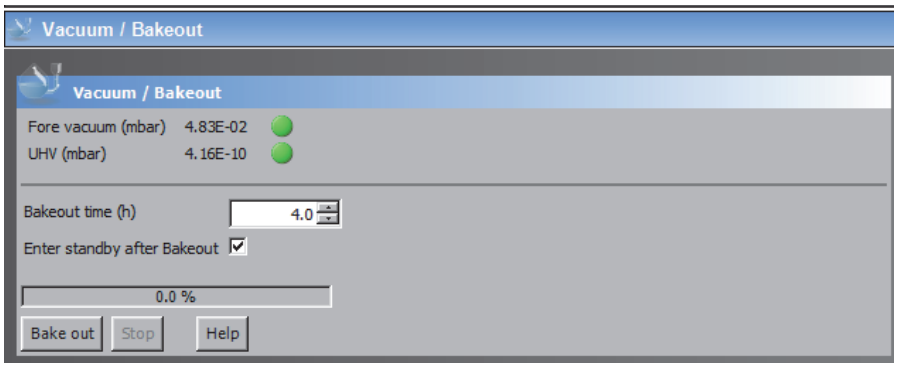


Figure 4-22. Vacuum / Bakeout window

NOTICE After the bakeout time has expired, the instrument requires a cooling and stabilization time of about three hours. ▲

The Vacuum / Bakeout window has the following parameters:

Parameter	Description
Vacuum	
Green LEDs indicate that the readback values are sufficient for operating the Q Exactive GC mass spectrometer. If an LED indicates a vacuum problem, use the instrument status window for error diagnosis.	
Fore vacuum	This readback displays the actual pressure (in mbar) in the forevacuum line and ion source region as read by the Pirani gauge.
UHV	This readback displays the actual pressure (in mbar) in the Orbitrap chamber as read by the ion gauge.
Bakeout	
Bakeout time	Enter the desired baking duration (in hours) into the spin box. The range is 4 to 100 hours. When you click **Bake out**, the mass spectrometer starts the baking routine. The baking script is stopped after the preset duration. Click **Stop** to abort the baking routine. **NOTICE** The instrument has to cool down for another three hours before it can be used. ▲
Enter standby after Bakeout	Select the check box to set the instrument to standby mode after the bakeout procedure is finished.
Progress bar	Displays the elapsed percentage of the current baking procedure.

Parameter	Description
Bake out	Click **Bake out** to start the bakeout routine. To stop a bakeout in progress, click the **Stop** button. **NOTICE** Bakeout is possible only when the instrument status is Off. ▲
Stop	Click **Stop** to abort the bakeout routine. **NOTICE** The instrument has to cool down for another three hours before it can be used. ▲
Help	Displays the Help for this window.

❖ **To display this window**

Click 

in the tasks panel.

Display Panel

The windows in the display panel provide real-time information about the instrument status, the Q Exactive GC Tune software, or other programs.

The following windows are available:

- Spectrum Window
- Instrument Status Window
- Messages Window
- Analysis Graphs Window
- Debug Messages Window

The spectrum window is always visible. The debug messages window is not available for standard users.

Depending on the individual configuration of your system, additional windows may allow controlling other installed instruments.

Q Exactive GC Tune offers various ways to arrange windows in the display panel:

❖ **To display a window in the display panel**

Choose the respective command in the Windows menu.

❖ **To hide a window in the display panel**

- Choose the respective command in the Windows menu.
- Right-click the title bar of a window to display the shortcut menu. Choose **Hide**.

❖ **To change the position of a window in the display panel**

- Use the mouse to drag the window by its title bar to the new location, which can be even outside the Q Exactive GC Tune window.
- Within the display panel, you can dock the window to any one of the four sides. While you drag the window, Q Exactive GC Tune displays icons to indicate the available docking positions.
- Right-click the title bar of a *docked* window to display the shortcut menu. Choose **Floating** to undock the window.

  Alternatively, double-click on the title bar of the window.

- Right-click the title bar of an *undocked* window to display the shortcut menu. Choose **Floating** to dock the window to its last docking position.

  Alternatively, double-click the title bar of the window.

Spectrum Window

The spectrum window displays real-time data generated during calibration, tuning, and diagnostic tests. See Figure 4-23. The spectrum window is always visible.

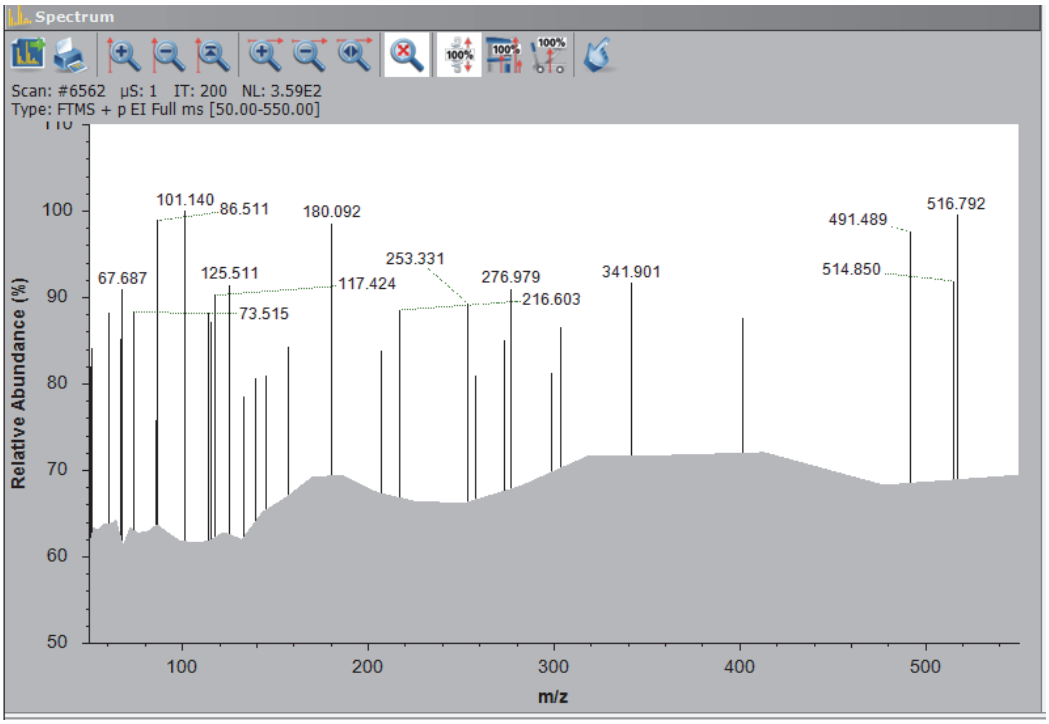


Figure 4-23. Spectrum window

The spectrum window allows using the mouse for zooming.

❖ **To zoom in or out on a spectrum**

- Click and drag with the left mouse button (hand cursor) from the beginning to the end of the portion you want to see enlarged.
- Alternatively, use the mouse wheel for zooming. Position the mouse pointer within the spectrum and roll the wheel forward to zoom in on the spectrum area.
- To zoom in on the spectrum with respect to one axis only, position the mouse pointer within the axis area and roll the mouse wheel forward. Roll the wheel backward to zoom out.

- To increase the zooming factor by two, keep the **<Shift>** key pressed while using the mouse wheel.

To return to a display of the full spectrum, click the  button in the toolbar or choose **Unzoom** in the shortcut menu.

Press the **<Shift>** key to enable mouse panning. When mouse panning is active (mouse pointer changes to hand cursor), you can shift the spectrum along the X-axis while keeping the left mouse button pressed.

The spectrum window has the following parameters:

Parameter	Description
Header	
Scan #	The number of scans since the beginning of the last acquisition (or since the last time the mass spectrometer was rebooted)
☒S	Number of microscans
IT	Inject time (in milliseconds)
NL	Normalization level
Type	Scan type The scan type information comprises ion polarity, source type, fragmentation type and energy (if active), and scan range. When the acquisition is controlled by an instrument method, the information includes the current scan segment and the current scan event.

❖ **To display this window**

Choose **Windows > Spectrum**.

Toolbar

Use the buttons in the toolbar to manipulate the spectrum display.

The toolbar of the spectrum window has the following parameters:

Parameter	Description
	Click to copy the current spectrum into the clipboard.
	Click to plot the current spectrum. You can view a print preview before the spectrum is printed. In a Page Setup dialog box, you can set up printer's parameters (paper size and orientation, for example).
	Click to zoom in on the Y-axis (factor 2).
	Click to zoom out on the Y-axis (factor 2).
	Click to normalize the Y scale: Q Exactive GC Tune always displays the largest peak in the spectrum at full scale (vertical scale = largest peak in spectrum).
	Click to zoom in on the X-axis (factor 2).
	Click to zoom out on the X-axis (factor 2).
	Click to display the entire mass range.
	Click to normalize the display (X-axis, Y-axis).
	Click to set to fixed mode: the height of the Y-axis is set equal to the height of the largest peak in the current spectrum and remains fixed even if larger peaks occur.
	Click to set the Y-axis to the height of the highest peak so that the largest peak in the spectrum is always displayed at full scale.
	Click to set to creep mode: the Y scale of the mass spectrum automatically increases if the peak intensity increases, but does not decrease if the peak intensity decreases.
	Click to toggle between mouse panning and mouse zooming: • When mouse panning is active, you can shift the spectrum along the X-axis while keeping the left mouse button pressed (hand cursor). • When mouse zooming is active, you can zoom in the spectrum by using the mouse.

Shortcut Menu

Right-click the spectrum window to display the shortcut menu. It has the following commands:

Command	Description
Save Image As	Saves the current graph as image file. A Save As dialog opens and you can enter the file name, file type, and the directory.
Unzoom	Reverts the zoom action.
Display options	Opens the Display Options dialog box.

Instrument Status Window

The instrument status window displays real-time status information for the instrument components. All parameters are arranged in a tree view. See Figure 4-24. The Control node is not available for standard users.

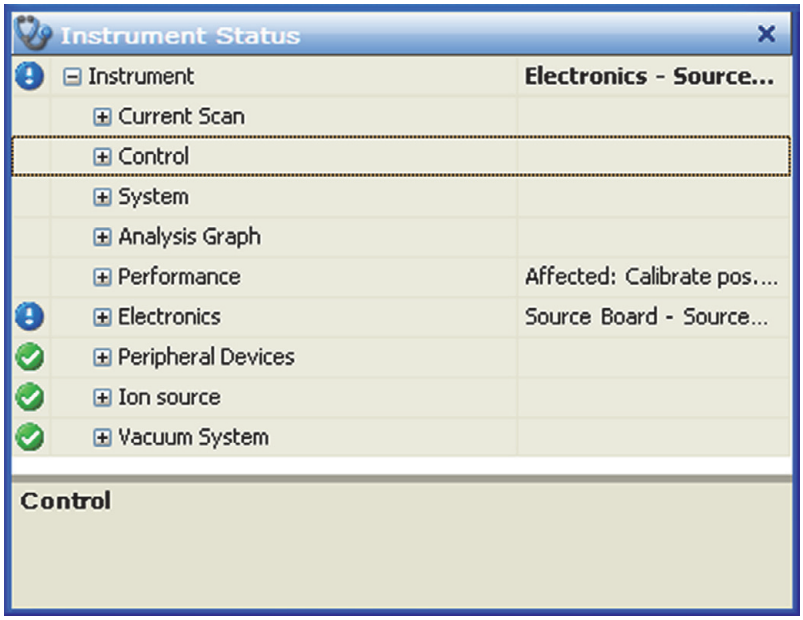


Figure 4-24. Instrument status window

In addition to showing numerical values of parameters, the instrument status window uses icons to indicate the statuses of system components. Thus, you can use the instrument status window for a quick error diagnosis.

When expanded, some nodes provide shortcut menus that are displayed when you right-click an item. The available commands depend on the selected node and the user privileges. Generally, the shortcut menus allow changing the current settings.

NOTICE For normal operation, it is not necessary to change the settings in the instrument status window. It just allows monitoring the system in more detail (more readbacks, etc.) than the other windows. ▲

❖ **To display this window**

Choose **Windows > Instrument Status**.

Messages Window

The messages window displays real-time information about the statuses of the instrument, the control service, or other programs. For further analysis, you can copy the content of the messages window to a text editor.

❖ **To copy content from the messages window to a text editor**

1. Select part of the content with the mouse, or press **<Ctrl>** + **<A>** to select the complete content of the messages window.
2. Press **<Ctrl>** + **<C>** to copy the text to the clipboard.
3. Open a document in the text editor.
4. Press **<Ctrl>** + **<V>** to insert the copied text into the document.

The messages window has the following parameters:

Parameter	Description
The messages are sorted with respect to time. To change the sort order, click the respective table column header. To invert the sort order, click again.	
Priority	The message priority is indicated by symbols:  Info; status is normal / command successful  Warning; no user action required  Error; user action required  Fatal error; program cannot proceed
Source	The message source is indicated by symbols:  Message from Q Exactive GC Tune program  Message from instrument  Message from service

Parameter	Description
Time	Time and date of the message
Description	Displays the message text.

❖ **To display this window**

Choose **Windows > Messages**.

Analysis Graphs Window

The analysis graphs window displays real-time data generated during calibration, tuning, and diagnostic tests. See Figure 4-25. To save data displayed in the window, choose **Save Image As** in the shortcut menu of the window.

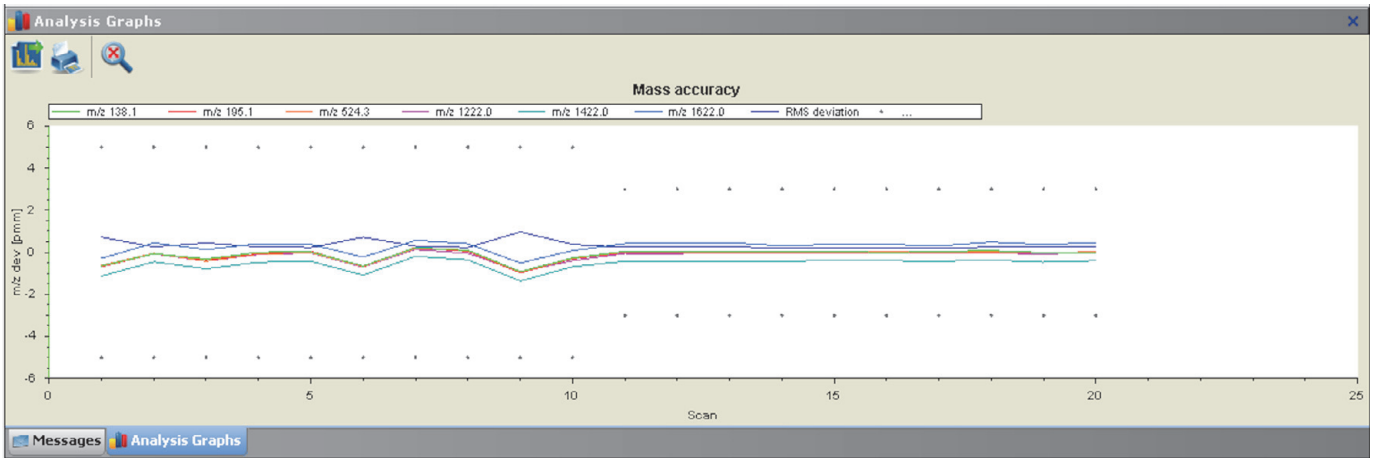


Figure 4-25. Analysis Graphs window

The analysis graphs window allows using the mouse for zooming.

❖ **To zoom in or out on an analysis graph**

- Click and drag with the left mouse button (hand cursor) from the beginning to the end of the portion you want to see enlarged.
- Alternatively, use the mouse wheel for zooming. Position the mouse pointer within the graph and roll the wheel forward to zoom in on the graph area.
- To zoom in on the graph with respect to one axis only, position the mouse pointer within the axis area and roll the mouse wheel forward. Roll the wheel backward to zoom out.
- To increase the zooming factor by two, keep the **<Shift>** key pressed while using the mouse wheel.

To return to a display of the full spectrum, click the  button in the toolbar or choose **Unzoom** in the shortcut menu.

Press the **<Shift>** key to enable mouse panning. When mouse panning is active (mouse pointer changes to hand cursor), you can shift the graph along both axes while keeping the left mouse button pressed.

❖ **To display this window**

Choose **Windows > Analysis Graphs**.

Toolbar

The toolbar of the analysis graphs window has the following parameters:

Parameter	Description
	Click to copy the current graph into the clipboard.
	Click to plot the current graph. You can view a print preview before the graph is printed. In a Page Setup dialog box, you can set up printer's parameters (paper size and orientation, for example).
	Click to normalize the display (X-axis, Y-axis).

Shortcut Menu

Right-clicking on the analysis graphs window displays the shortcut menu. It has the following commands:

Command	Description
Save Image As	Saves the current graph as image file. A Save As dialog opens and you can enter the file name, file type, and the directory.
Unzoom	Reverts the zoom action.

Debug Messages Window

The debug messages window displays messages that can be used during method development. The debug messages window is not available for standard users. For further analysis, you can copy the content of the debug messages window to a text editor.

❖ **To copy content from the debug messages window to a text editor**

1. Select part of the content with the mouse, or
   press **<Ctrl>** + **<A>** to select the complete content of the messages window.
2. Press **<Ctrl>** + **<C>** to copy the text to the clipboard.
3. Open a document in the text editor.
4. Press **<Ctrl>** + **<V>** to insert the copied text into the document.

The debug messages window has the following parameters:

Parameter	Description
The messages are sorted with respect to time. To change the sort order, click the respective table column header. To invert the sort order, click again.	
Priority	The message priority is indicated by symbols:  Debugging message  Info; status is normal / command successful  Warning; no user action required  Error; user action required  Fatal error; program cannot proceed
Source	The message source is indicated by symbols:  Message from Q Exactive GC Tune program.  Message from instrument.  Message from service.
Time	Time and date of the message
Description	Displays the message text.

❖ **To display this window**

Choose **Windows > Debug Messages**.

Shortcut Menu of the Debug Messages Window

The debug messages window has a shortcut menu that is displayed when you right-click into the window. Use the commands of the shortcut menu to configure the scope of the messages displayed in the window. Click a command to activate it; click it again to deactivate it.

The following message priorities can be displayed:

- Debug

- Log
- Info
- Warning
- Error
- Fatal

For each message priority, the following message sources are available:

- Instrument
- Service
- Application

A  icon in front of a message source shows that it is selected. A  icon in front of a message priority shows that it is shown for all three message sources. A  icon in front of a message priority shows that it is shown for one or two message sources.

In addition to the commands for configuring the content of the debug messages window, the **Clear list** command allows removing the complete content of the window.

Dialog Boxes

This section provides a reference to the dialog boxes in Q Exactive GC Tune.

Dialog Boxes Displayed from the Menu Bar

- About Dialog Box
- License Dialog Box
- Mass Calculator

Dialog Boxes Displayed from the Toolbar

- Scan Parameters History Dialog Box

Dialog Boxes Displayed from the Scan Parameters Window

- Fragmentation Dialog Box
- Scan Parameters History Dialog Box
- Scan Range Dialog Box
- Scan Type Dialog Box

Dialog Boxes for Editing Lock Masses

- Collection Modification Dialog Box
- Delete Lock Mass Collection Dialog Box
- Lock Masses Dialog Box
- Lock Mass Removal Dialog Box
- Lock Mass Replacement Dialog Box
- Name Change Dialog Box
- New Lock Mass Collection Dialog Box

Dialog Boxes Displayed from the Spectrum Window

- Display Options Dialog Box

About Dialog Box

Use the About dialog box to display information about the instrument, the current Q Exactive GC Tune version, and the active licenses. See Figure 4-26. To copy the instrument identification to the clipboard, click the button.

Figure 4-26. About dialog box

The About dialog box has the following parameters:

Parameter	Description
Buttons	
OK	Saves your changes and closes the dialog box.
Add license	Displays the License dialog box.
Back / Next	Display various dialog boxes with legal information and information about third party licenses used by Q Exactive GC Tune.
Help	Displays the Help for this dialog box.

❖ **To display this dialog box**

Choose **Help > About**.

License Dialog Box

Use the License dialog box to enter licenses that activate additional features. See Figure 4-27.

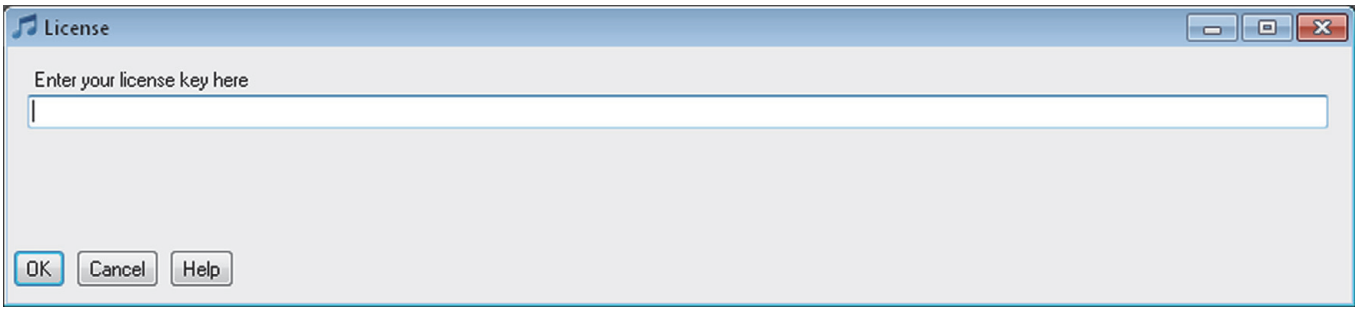


Figure 4-27. License dialog box

NOTICE This feature is used by a qualified Thermo Fisher Scientific engineer for service. ▲

The License dialog box has the following parameters:

Parameter	Description
License key	Use the text box to enter the license key.
Buttons	
OK	Saves your changes and closes the dialog box.
Cancel	Discards your changes and closes the dialog box.
Help	Displays the Help for this dialog box.

❖ **To display this dialog box**

1. Choose **Help > About** to display the About dialog box.
2. In the About dialog box, click **Add license**.

Mass Calculator

Use the Mass Calculator to calculate the exact mass of an compound of interest.

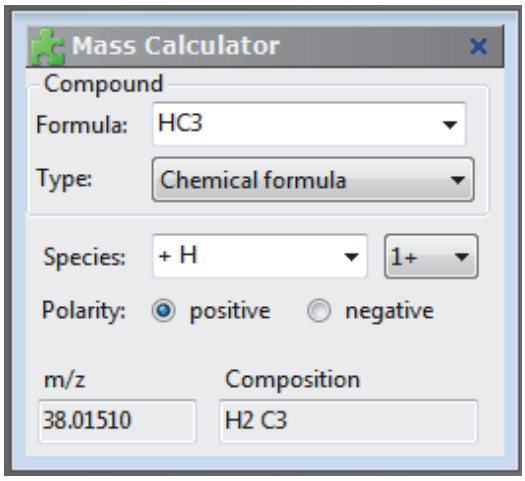


Figure 4-28. Mass Calculator (Tune)

The Mass Calculator has the following parameters:

Parameter	Description
Compound	
Formula	Use the text field to specify the basic composition of the compound. Click the down arrow to display a list of the last entered compounds. The syntax rules and interpretation accept the kind of formula that is also accepted on the Spectrum Simulation page (Isotope Simulation area) of the Qual Browser.
Type	Use the Type list to specify the formula type used in the Formula field: • **Chemical formula** Formula is defined as the resulting elemental composition of the entered formula. This is the default selection. Independent of the entry in the Species field, the elemental composition and the mass of the compound, Formula is completely determined by the given formula and selected formula type.

Parameter	Description
Species	Use this text field to define adducts or modifications of the active compound that are expected to be formed. The Species field can be used in two ways: • Click the down arrow to display a list of predefined adducts for each polarity: - +H, +NH4, +C2H5, +C3H5, +C4H9 for positive polarity - -H for negative polarity - an empty entry to express adducts (for example, radical cations) for both polarities. The corresponding set will be displayed depending on the selected polarity. Selecting an adduct (A) will result in the strict behavior of applying one unit A to the compound and using "+H" or "-H" adduct depending on the charge state and active polarity. • Enter the modifications of the compound by using squared brackets and at least M as representation of the basic compound. The predefined adducts can be entered, too. This definition is used without additional auto dependencies, like adding protons.
Charge state	Use the list box to select the resulting charge state. Available options are 1+ to 25+ for positive polarity and 1– to 25– for negative polarity. The default charge state is 1+ for positive polarity and 1– for negative polarity.
Polarity	Use the option buttons to select the applied polarity. The default polarity is **positive**.
m/z	This text field displays the calculated mass-to-charge ratio, with five decimal places (for example, 524.26496).
Composition	This text field displays the determined total formula (for example, C23 H38 N7 O5 S).

❖ **To display the Mass Calculator**

Choose **Windows > Mass Calculator**.

Scan Parameters History Dialog Box

Use the Scan Parameters History dialog box to select from a list of recent scans based on short scan descriptions. See Figure 4-29. Select a scan from the list to populate all scan parameters in the Scan Parameters window with the parameters from that scan.

If the Hot link check box is selected in the Scan Parameters window, selecting another list item immediately changes the instrument parameters.

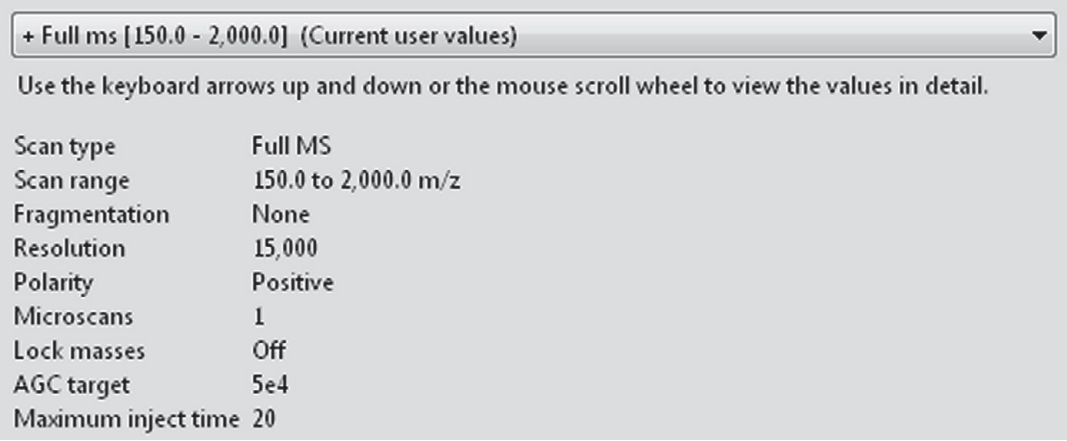


Figure 4-29. Scan Parameters History dialog box

❖ **To display this dialog box**

In the Scan Parameters window, click the History field.

Scan Type Dialog Box

Use the Scan Type dialog box to select the scan type to be used during the currently selected scan event. See Figure 4-30.

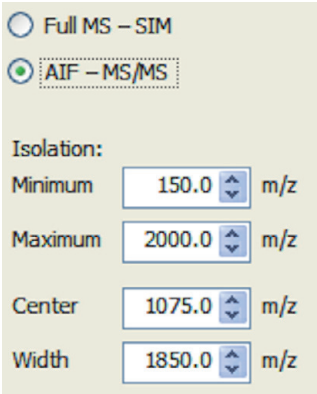


Figure 4-30. Tune—Scan Type dialog box

The Scan Type dialog box has the following parameters:

Parameter	Description
AIF - MS/MS	Select this option button to enable a Full MS scan (isolation width > 10 amu) or a selected ion monitoring (isolation width ≤10 amu) scan type. **NOTICE** For Full MS – SIM experiments, set the scan options in the Scan Range dialog box.
Isolation	
Use the Isolation area to set the scan range of the selected scan type.	
Minimum	Use this spin box to select the minimum value for the scan range (in mass-to-charge ratio units) used during the currently selected scan event. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can enter any value from 30.0 to 2500.
Maximum	Use this spin box to select the maximum value for the scan range (in mass-to-charge ratio units) used during the currently selected scan event. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can enter any value from 30.4 to 3000.0.
Center	Use this spin box to select the center mass (in mass-to-charge ratio units) of the scan range. When the isolation width is < 50, the software considers Center = Precursor. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can enter any value from 30.2 to 2750.0.
Precursor	Use this spin box to select the mass of a precursor ion of interest. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can enter any value from *m/z* 30.2 to 4250.
Width	Use this spin box to select either the width of the scan range (in mass-to-charge ratio units) used for a SIM scan or the isolation width for the precursor ion of interest during an MS/MS scan. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. For Full MS scans, you can enter any value from *m/z* 0.4 to 2800.0. For SIM and MS/MS scans, you can enter any value from *m/z* 0.4 to 10.

❖ **To display this dialog box**

In the Scan Parameters window, click the Scan type field.

Scan Range Dialog Box

Use the Scan range dialog box to specify the scan range of the data acquisition. See Figure 4-31. The scan range affects the amount of stored and transmitted data.

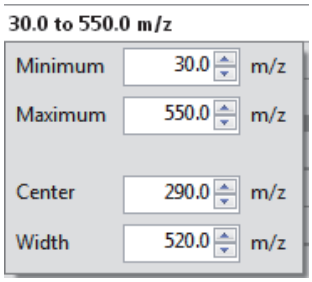


Figure 4-31. Scan range dialog box

The Scan Range dialog box allows setting either minimum and maximum or center and width of the scan range. If you change one pair of values, the other pair is changed accordingly.

NOTICE The ratio for maximum-to-minimum should not exceed 15. If necessary, Q Exactive GC Tune will correct the settings automatically. ▲

The Scan range dialog box has the following parameters:

Parameter	Description
Minimum	Use this spin box to select the minimum value for the scan range (in mass-to-charge ratio units) used during the currently selected scan event. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can enter any value from 30.0 to 2500.0. **NOTICE** Scanning below 50 *m/z* might reduce instrument performance. ▲
Maximum	Use this spin box to select the maximum value for the scan range (in mass-to-charge ratio units) used during the currently selected scan event. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can enter any value from 30.4 to 3000.0.

Parameter	Description
Center	Use this spin box to select the center mass (in mass-to-charge ratio units) of the scan range. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can enter any value from 30.2 to 2750.0.
Width	Use this spin box to select the width of the scan range (in mass-to-charge ratio units) used during the currently selected scan event. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can enter any value from 0.4 to 2800.0.

❖ **To display this dialog box**

In the Scan Parameters window, click the Scan range field.

Fragmentation Dialog Box

Use the Fragmentation dialog box to activate a fragmentation with the HCD collision cell. You can specify the fragmentation voltages to be used. See Figure 4-32.

Figure 4-32. Tune—Fragmentation dialog box

The Fragmentation dialog box has the following parameters:

Parameter	Description
HCD	
Activates the HCD collision cell. When you select this check box, Q Exactive GC Tune automatically changes the scan type, if necessary. **NOTICE** When HCD fragmentation is on, automatic tuning comprises only HCD relevant parameters. ▲	
CE	Use this box to select the collision energy for the HCD fragmentation. To change the value, click the arrows in the spin box to increment [up arrow] or decrement [down arrow] the value. You can enter any value from 10 to 200.

❖ **To display this dialog box**

In the Scan Parameters window, click into the Fragmentation field.

Collection Modification Dialog Box

Use this dialog box to modify a lock mass collection by importing lock masses from an xml file See Figure 4-35. This dialog box appears only when the xml file contains information about lock mass collections that differs from the available lock mass collections.

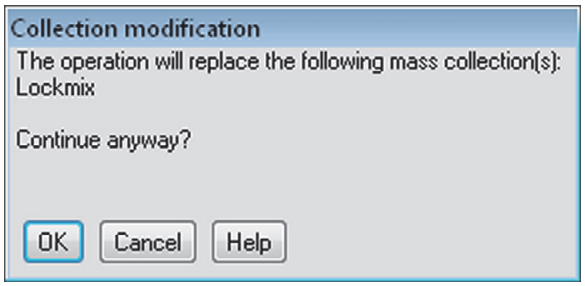


Figure 4-33. Collection Modification dialog box

The Collection Modification dialog box has the following parameters:

Parameter	Description
OK	Saves your changes and closes the dialog box. The lock mass collection is updated with the data contained in the xml file.
Cancel	Discards your changes and closes the dialog box.
Help	Displays the Help for this dialog box.

❖ **To display this dialog box**

1. Choose **Import > Merge with file content** in the shortcut menu of the Lock Masses dialog box.
2. In the file selection dialog box, select an xml file to import from.

Lock Masses Dialog Box

Use the Lock Masses dialog box to edit lock mass lists. See Figure 4-34. The table shows all available lock masses. Lock mass collections store information about lock mass usage.

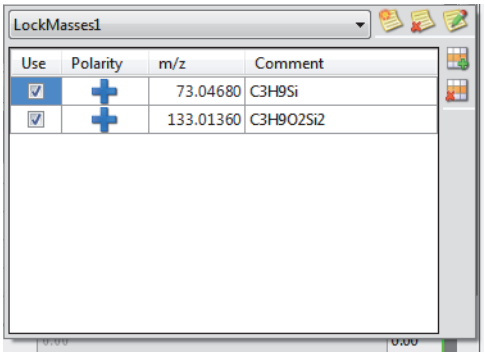


Figure 4-34. Tune—Lock Masses dialog box

The Lock Masses dialog box has the following parameters:

Parameter	Description
Lock mass lists	
Lock mass lists	Use the list box to select an existing lock mass collection.
	Displays the New Lock Mass Collection dialog box where you can enter the name for a new lock mass collection.
	Deletes the active lock mass collection. The lock masses themselves are still available. This button is not available when no lock mass collection is selected.
	Displays the Name Change dialog box where you can edit the name of the active lock mass collection. This button is not available when no lock mass collection is selected.

Parameter	Description
Available lock masses	
The table displays the properties of the available lock masses and whether they are used in the selected lock mass collection. To change the sort order, click the respective table column header. To invert the sort order, click again.	
Lock mass table	• Use Select the check box to use the lock mass. A ☑ indicates that the lock mass is used in the active lock mass collection. The lock mass is displayed in the Lock masses field of the Scan Parameters window. • Polarity Click the symbol to change the polarity of the lock mass. **NOTICE** During a scan, the Q Exactive GC mass spectrometer uses only the lock masses with a polarity that matches the active ion mode. ▲ • m/z Enter the mass of the lock mass into the field (with a maximum of five decimals). • Comment Enter a comment for the lock mass into the field. This field is optional.
	Creates a new lock mass.
	Deletes the selected lock masses.

❖ **To display this dialog box**

In the Scan Parameters window, click into the Lock Masses field.

Shortcut Menu of the Lock Masses dialog box

The Lock Masses dialog box has a shortcut menu that is displayed when you right-click into the dialog box.

It has the following commands:

Command	Description
Select all masses in use	Selects all masses used in the active lock mass collection. This command is not available when no lock mass is used.
Import > Merge with clipboard	Appends the content of the clipboard to the available lock masses. **NOTICE** The clipboard stores lock mass data as tabulator-separated text files. When the data in the clipboard are not in this format, the command is not available. ▲

Command	Description
Import > Replace by clipboard	Replaces the available lock masses by the content of the clipboard. **NOTICE** The clipboard stores lock mass data as tabulator-separated text files. When the data in the clipboard are not in this format, the command is not available. ▲
Import > Merge with file content	Opens a dialog box so that you can select a .csv, .txt, or .xml file that contains a list of lock masses. The content of the file is appended to the available lock masses. See Exporting and Importing Lock Masses for information about the format of lock mass files.
Import > Replace by file content	Opens a dialog box so that you can select a .csv, .txt, or .xml file that contains a list of lock masses. All available lock masses are replaced by the masses contained in the imported file. The present lock masses are removed from all existing lock mass lists. You have to confirm your action in the Lock mass replacement dialog box. See Exporting and Importing Lock Masses for information about the format of lock mass files.
Export > Copy selected to clipboard	Copies the data of the selected lock mass to the clipboard.
Export > Copy all to clipboard	Copies the data of all available lock masses to the clipboard.
Export > Copy selected to file	Opens the dialog box, where you can save the data of the selected lock mass(es) as a .csv, .txt, or .xml file. See Exporting and Importing Lock Masses for information about the format of lock mass files.
Export > Copy all to file	Opens the dialog box, where you can save the data of all available lock masses as a .csv, .txt, or .xml file. See Exporting and Importing Lock Masses for information about the format of lock mass files.

Lock Mass Removal Dialog Box

Use this dialog box to delete one or more lock masses that are selected in the Lock Masses dialog box. See Figure 4-35.

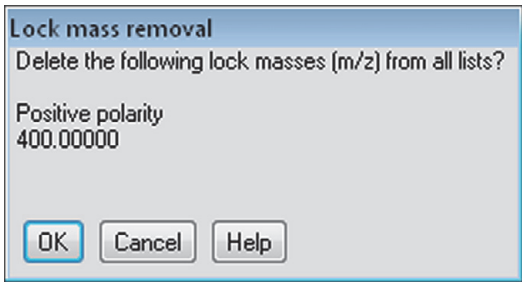


Figure 4-35. Lock Mass Removal dialog box

The Lock Mass Removal dialog box has the following parameters:

Parameter	Description
OK	Saves your changes and closes the dialog box. The lock masses are now removed from the table in the Lock Masses dialog box.
Cancel	Discards your changes and closes the dialog box.
Help	Displays the Help for this dialog box.

❖ **To display this dialog box**

In the Lock Masses dialog box, click the  button.

Lock Mass Replacement Dialog Box

Use this dialog box to replace all available lock masses by masses contained either in a csv, txt, or xml file or in the clipboard. See Figure 4-35.

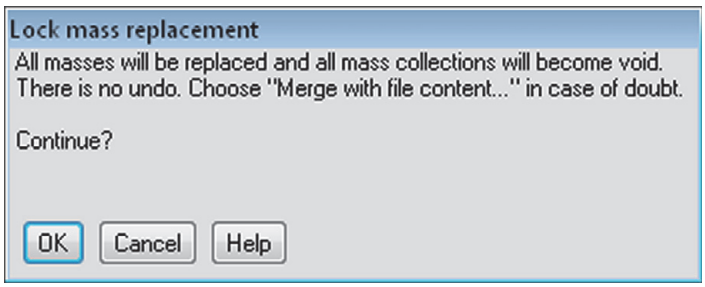


Figure 4-36. Lock Mass Replacement dialog box

The Lock Mass Replacement dialog box has the following parameters:

Parameter	Description
OK	Removes the lock masses and replaces them by the imported lock masses. The present lock mass collections become void.
Cancel	Closes the dialog box.
Help	Displays the Help for this dialog box.

❖ **To display this dialog box**

Choose **Import > Replace by file content** or **Import > Replace by clipboard** in the shortcut menu of the Lock Masses dialog box.

New Lock Mass Collection Dialog Box

Use this dialog box to enter a name for the lock mass collection that is displayed in the Lock Masses dialog box. See Figure 4-37.

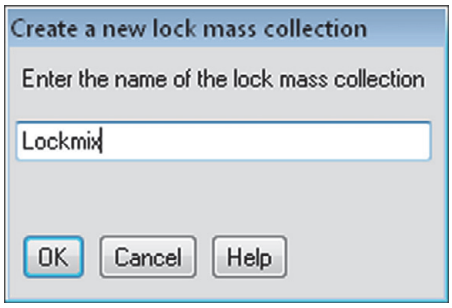


Figure 4-37. New Lock Mass Collection dialog box

The New Lock Mass Collection dialog box has the following parameters:

Parameter	Description
Name of lock mass collection	Use this text field to enter the name for the new lock mass collection.
Buttons	
OK	Saves your changes and closes the dialog box. The name of the lock mass collection is now displayed in the list box of the Lock Masses dialog box.
Cancel	Discards your changes and closes the dialog box.
Help	Displays the Help for this dialog box.

❖ **To display this dialog box**

In the Lock Masses dialog box, click the button.

Delete Lock Mass Collection Dialog Box

Use this dialog box to delete an existing lock mass collection. See Figure 4-38. The lock masses themselves are not deleted.

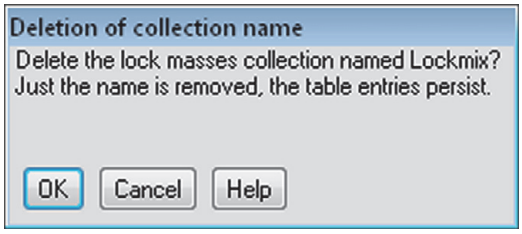


Figure 4-38. Delete Lock Mass Collection dialog box

The Delete Lock Mass Collection dialog box has the following parameters:

Parameter	Description
OK	Saves your changes and closes the dialog box. The name of the lock mass collection is now removed from the list box of the Lock Masses dialog box.
Cancel	Discards your changes and closes the dialog box.
Help	Displays the Help for this dialog box.

❖ **To display this dialog box**

In the Lock Masses dialog box, click the button.

Name Change Dialog Box

Use this dialog box to change the name of an existing lock mass collection. See Figure 4-39.

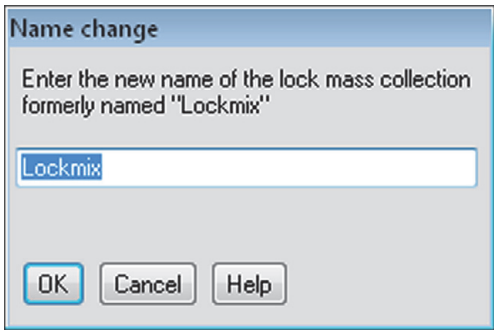


Figure 4-39. Name Change Dialog Box

The Name Change dialog box has the following parameters:

Parameter	Description
Name of lock mass collection	Use this text field to enter the new name for the lock mass collection.
Buttons	
OK	Saves your changes and closes the dialog box. The name of the lock mass collection is now displayed in the list box of the Lock Masses dialog box.
Cancel	Discards your changes and closes the dialog box.
Help	Displays the Help for this dialog box.

❖ **To display this dialog box**

In the Lock Masses dialog box, click the  button.

Display Options Dialog Box

Use this dialog box to modify the appearance of the displayed mass spectrum in the spectrum window. See Figure 4-40.

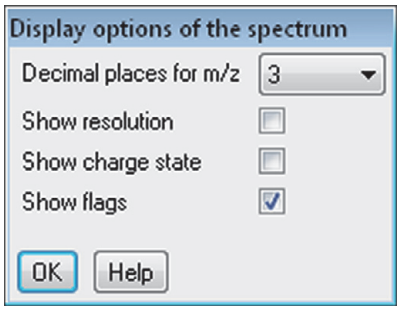


Figure 4-40. Display Options Dialog Box

The Display Options dialog box has the following parameters:

Parameter	Description
Decimal places for m/z	Use this list box to set how many decimal places of precision are shown when mass positions are annotated on mass spectra. The valid range is 0 to 5.
Show resolution	Select this check box to have Q Exactive GC Tune annotate mass peaks with the mass resolution.
Show charge state	Select this check box to have Q Exactive GC Tune annotate mass peaks with the charge state.
Show flags	Select this check box to have Q Exactive GC Tune indicate peaks that are contained in the active lock mass collection. **NOTICE** The scan parameters window must show entries for lock masses. ▲
Buttons	
OK	Saves your changes and closes the dialog box.
Help	Displays the Help for this dialog box.

❖ **To display this dialog box**

In the shortcut menu of the spectrum window, choose **Display options**.

Tune Reports

To generate a tune report, select the **EI Tune Report** box in the **Calmix Evaluation Window** (see "Calmix Evaluation Window" on page 4-31). To view an EI tune report, in the top menu, go to **Reports > EI Tune Report**. See Figure 4-41.

Q Exactive GC Tune Report

Thermo SCIENTIFIC

Date of Tune Report: 2015-05-08 16:31:09
Instrument Identification: Exactive Series slot #MP-5 (001999C91DFD)
Software Version: 2.6-262803/2.6.0.2629
Data System User: USER\username
Tune File: C:\Xcalibur\methods\EI_Positive.mstune

Ion Source Tune Settings

Parameter	Set value	Read value
Emission current (µA)	50.0	49.0
Electron lens voltage (V)	15.0	14.6
Filament voltage (V)	-64.7	-64.8
Source offset voltage (V)	5.3	5.1
Repeller voltage (V)	7.7	7.5
Lens 1 voltage (V)	-49.5	-48.9
Lens 2 voltage (V)	-1.0	-1.4
Lens 3 voltage (V)	-20.7	-20.5

Ion Transfer and Analyzer Injection Tune Settings

Parameter	Set value	Read value
Inter Flatapole Lens (V)	2.5	2.3
Injection Flatapole DC (V)	1.8	1.8
C-Trap Energy Offset (V)	0.0	N/A

Name:_____ Signature:_____ Date: _____

Figure 4-41. EI Tune Report

NOTICE The accuracy and precision of the read values are not carefully controlled. The read values should be within approximately 1.3 V of the set value. If the read values are much different, have a qualified person check for a short in the wires supplying the ion source, such as wires touching the manifold or magnet yoke. ▲

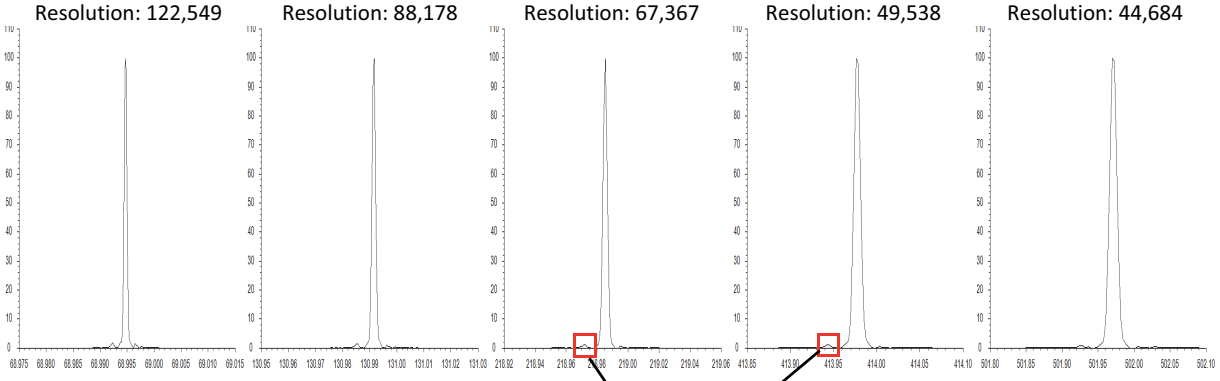
NOTICE Shoulder peaks are part of the frequency signal measured from the Orbitrap and are labeled as exception peaks so they do not get processed as real peaks. If these shoulder peaks become too intense relative to the main peak, run the eFT parameter and MS mass calibrations. If this does not bring the shoulder peaks below 5%, the lenses admitting ions into the Orbitrap may need to be tuned by a qualified service person. ▲

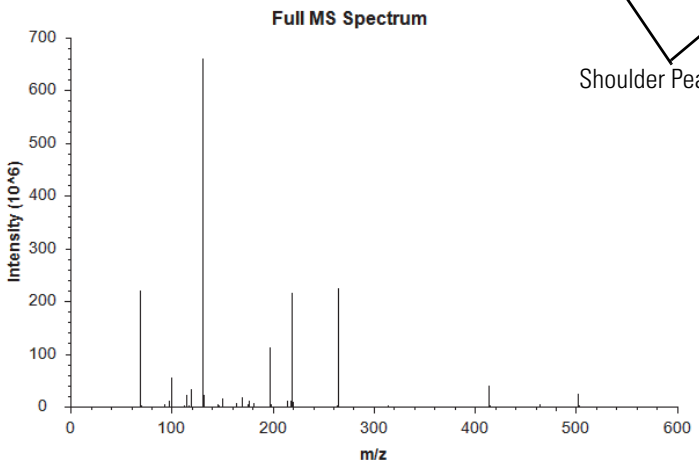
Q Exactive GC Tune Report

Thermo SCIENTIFIC

Date of Tune Report:	2015-05-08 16:31:09
Last Mass Calibration:	2015-05-08 16:25:12
Instrument Identification:	Exactive Series slot #MP-5 (001999C91DFD)
Software Version:	2.6-262803/2.6.0.2629
Data System User:	USER\username
Tune File:	C:\Xcalibur\methods\EI_Positive.mstune

m/z 68.99471	m/z 130.99155	m/z 218.98513	m/z 413.97727	m/z 501.97099
Intensity: 2.208e+08	Intensity: 6.607e+08	Intensity: 2.159e+08	Intensity: 4.148e+07	Intensity: 2.544e+07
Accuracy: 0.64 ppm	Accuracy: 0.61 ppm	Accuracy: 0.24 ppm	Accuracy: 0.70 ppm	Accuracy: 0.79 ppm
Resolution: 122,549	Resolution: 88,178	Resolution: 67,367	Resolution: 49,538	Resolution: 44,684





Shoulder Peaks

Scan Type	Full MS
Scan range	50 to 550 m/z
Resolution @ m/z 200	60,000
AGC target	1,000,000
Microscans	10
Ionization mode	EI
Polarity	Positive
Electron energy	70 eV
Emission current	50 µA
Filament selection	2
Ion source temperature	200.0 °C
MS transfer line temperature	249.9 °C
Fore vacuum	3.69e-02 mbar
Ultra-high vacuum	5.36e-10 mbar

Theoretical Monoisotopic Peak (m/z)	Mass Accuracy (ppm)	Relative Abundance (%)	Theoretical Isotope Peak (m/z)	Mass Accuracy (ppm)	Isotope Ratio (%)
68.99466	0.64	33.42	69.99802	0.74	1.22
130.99147	0.61	100.00	131.99482	0.11	3.32
218.98508	0.24	32.67	219.98844	0.09	4.38
413.97698	0.70	6.28	414.98033	1.10	8.62
501.97059	0.79	3.85	502.97394	0.65	10.17

Last Leak Check: 2015-05-08 16:26:40, Leak check completed: 0.81%

Name:_____ Signature:_____ Date: _____

The relative intensities of ions displayed in the EI tune report vary according to the ions specified in the tune as well as the ion source temperature.

Parameters of interest in the tune report are:

Parameter	Description
m/z 219	The resolution should be above the value set in the tune file. The peak at m/z 219 will be low in intensity after venting the instrument or admitting moist atmospheric air into the nitrogen supply. Its intensity can indicate the trapping gas purity.
Accuracy	If the instrument has been recently calibrated, this value should be < 1 ppm. I f accuracy is not < 3 ppm, a perform a mass calibration.
Ultra-high vacuum	This value should be < 8e-10 mbar
Last leak check	This parameter displays the results from the last date and time a leak check was run. Values should be less than 10%. If the result is over 10%, the software reports that the leak check failed. See the *Q Exactive GC Operating Manual* for troubleshooting information. If you cannot find a leak and the leak check remains higher than 10%, contact customer support.

Chapter 5 Procedures in Q Exactive GC Tune

This chapter describes procedures that you may need when using Q Exactive GC Tune.

Contents

Using Lock Masses and Lock Mass Collections

This section provides instructions for using lock masses and lock mass collections.

The following topics are available:

- Adding a Lock Mass
- Deleting Lock Masses
- Exporting and Importing Lock Masses
- Creating a new Lock Mass Collection
- Renaming a Lock Mass Collection
- Adding a Lock Mass to a Lock Mass Collection
- Removing a Lock Mass from a Lock Mass Collection
- Deleting a Lock Mass Collection

Adding a Lock Mass

❖ **To add a lock mass**

1. In the Lock Masses dialog box, click the  button. A new row appears at the end of the lock mass table. See Figure 5-1.

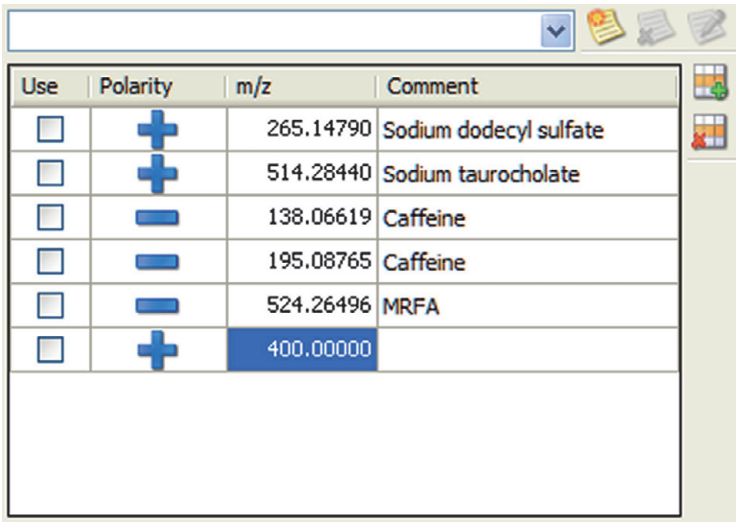


Figure 5-1. Adding a new lock mass

2. Enter the data for the new lock mass into the new table row:

   a. Select the Use check box if you want to use the new lock mass in the active lock mass collection.

b. Click the cell in the Polarity column until the correct sign appears:  for positive or  for negative.

c. Enter the m/z value for the new lock mass (with a maximum of five decimals).

d. Optionally, enter a comment for the new lock mass.

The new lock mass is now available to be used.

Deleting Lock Masses

❖ **To delete lock masses**

1. In the Lock Masses dialog box, select one lock mass or several lock masses:
   - To select a single lock mass, click into the respective table row.
   - To select adjacent lock masses, click the first table row. Then hold down the **<Shift>** key while you click the last row.
   - To select nonadjacent lock masses, click the first table row. Then hold down the **<Ctrl>** key while you click the other rows that you want to add to the selection.
2. Click the  button. The Lock Mass Removal dialog box appears and requests your confirmation for deleting the listed lock masses.
3. Click **OK** to confirm your input and to close the dialog box. All selected lock masses are removed from the list in the Lock Masses dialog box.

Exporting and Importing Lock Masses

Q Exactive GC Tune allows exporting lock mass data to the clipboard or to a file. Other applications can then use the data. You can also copy the export files to other computers where Q Exactive GC Tune is installed. So you can reuse the lock masses without having to recreate them. To reuse the lock masses, import the data either from the clipboard or from a lock mass file.

With a spreadsheet, a text editor, or an XML editor, you can create lock mass files even without using Q Exactive GC Tune.

Lock Mass Files

Q Exactive GC Tune uses the following file formats for exporting and importing lock mass data:

- Comma-separated value lists (.csv)

- Tabulator-separated text files (.txt)
- XML files (.xml)

Lock mass data are stored in the following order:

1. Usage status: True for used, False for unused
2. Ion polarity: positive mode or negative mode
3. *m/z*: up to five decimals
4. Comment: optional

The clipboard stores lock mass data as tabulator-separated text files. In contrast to . csv and .txt files, .xml files additionally store information about lock mass collections and lock mass usage.

Exporting Lock Masses

Q Exactive GC Tune allows exporting lock mass data to the clipboard or to a file.

❖ **To export selected lock masses to the clipboard**

1. In the Lock Masses dialog box, select the lock masses you want to export:
   - To select a single lock mass, click into the respective table row.
   - To select adjacent lock masses, click the first table row. Then hold down the **<Shift>** key while you click the last row.
   - To select nonadjacent lock masses, click the first table row. Then hold down the **<Ctrl>** key while you click the other rows that you want to add to the selection.
2. Right-click into the dialog box to display the shortcut menu.
3. Choose **Export > Copy selected to clipboard** to copy the selected lock mass to the clipboard.

❖ **To export all lock masses to the clipboard**

1. In the Lock Masses dialog box, right-click into the dialog box to display the shortcut menu.
2. Choose **Export > Copy all to clipboard** to copy all available lock mass to the clipboard.

❖ **To export some lock masses to a file**

1. In the Lock Masses dialog box, select the lock masses you want to export:

- To select a single lock mass, click into the respective table row.
- To select adjacent lock masses, click the first table row. Then hold down the **<Shift>** key while you click the last row.
- To select nonadjacent lock masses, click the first table row. Then hold down the **<Ctrl>** key while you click the other rows that you want to add to the selection.

2. Right-click into the dialog box to display the shortcut menu.

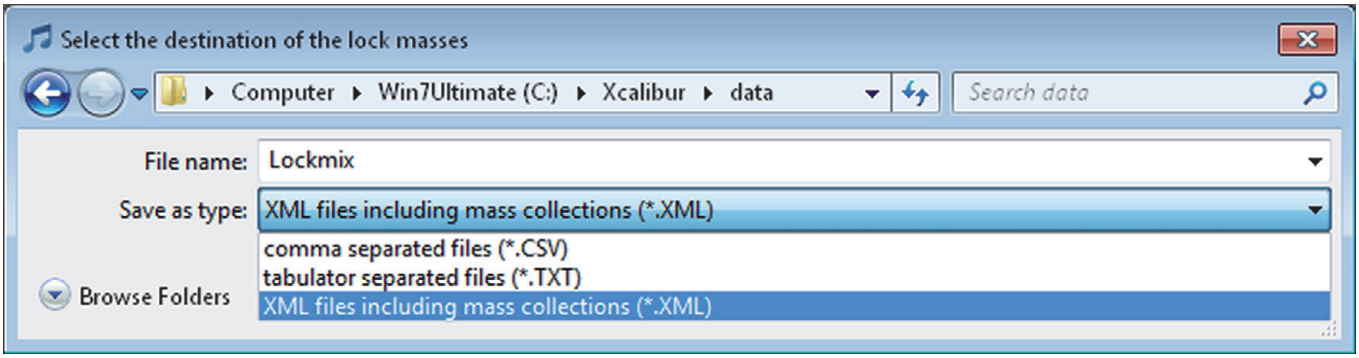


Figure 5-2. Selecting the lock mass export file

3. Choose **Export > Copy selected to file** to open the file selection dialog box. See Figure 5-2.
   a. Browse to the destination of the export file.
   b. Select the format of the export file.
   c. Enter a new name for the export file or select an existing file.
   d. Click **Save** to export the selected lock masses to the file. If you want to overwrite an existing file, you have to confirm your action in a dialog box. The file selection dialog box is closed.

❖ **To export all lock masses to a file**

1. In the Lock Masses dialog box, right-click into the dialog box to display the shortcut menu.

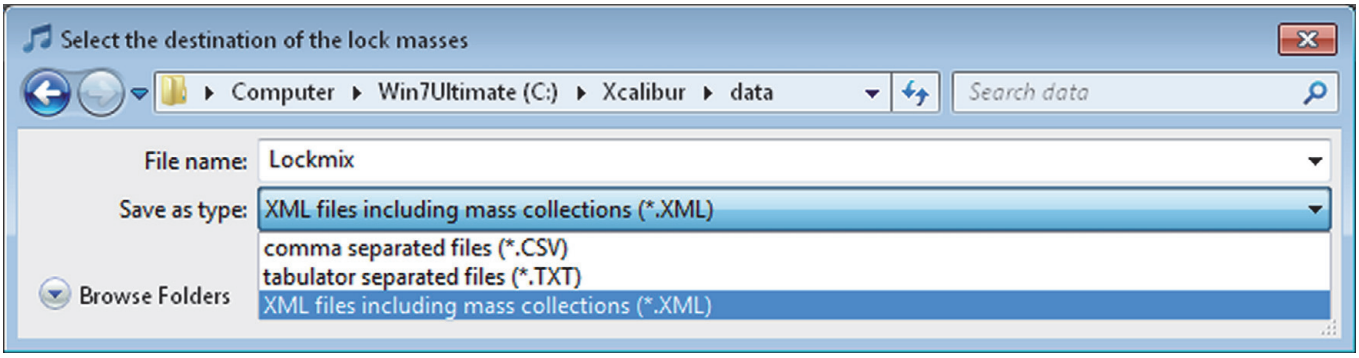


Figure 5-3. Selecting the lock mass export file

2. Choose **Export > Copy all to file** to open the file selection dialog box. See Figure 5-3.

   a. Browse to the destination of the export file.

   b. Select the format of the export file.

   c. Enter a new name for the export file or select an existing file.

   d. Click **Save** to export the lock masses to the file. If you want to overwrite an existing file, you have to confirm your action in a dialog box. The file selection dialog box is closed.

Importing Lock Masses

Q Exactive GC Tune allows importing lock mass data from the clipboard or from a file. The imported lock masses either are added to the present lock masses or they replace them.

❖ **To add lock masses from the clipboard**

1. Copy lock mass data in an appropriate format to the clipboard. See Lock Mass Files for information about the requirements.

2. In the Lock Masses dialog box, right-click into the dialog box to display the shortcut menu.

3. Choose **Import > Merge with clipboard** to import the lock mass data from the clipboard to the lock mass table.

NOTICE The clipboard stores lock mass data as tabulator-separated text files. When the data in the clipboard are not in this format, the command is not available. ▲

4. The lock mass list in the Lock Masses dialog box displays additional rows for the imported lock masses.

❖ **To add lock masses from a file**

1. In the Lock Masses dialog box, right-click into the dialog box to display the shortcut menu.
2. Choose **Import > Merge with file content** to open the file selection dialog box.
   a. Browse to the destination of the import file.
   b. Select the format of the import file.
   c. Select one of the displayed files.
   d. Click **Open** to import the lock masses to Q Exactive GC Tune. The dialog box is closed.
3. If you are importing an xml file that contains information about lock mass collections that differs from the present lock mass collections, the Collection modification dialog is displayed. Click **OK** to confirm the operation and to close the dialog box.The lock mass list in the Lock Masses dialog box displays additional rows for the imported lock masses.

❖ **To replace all lock masses by clipboard data**

1. Copy lock mass data in an appropriate format to the clipboard. See Lock Mass Files for information about the requirements.
2. In the Lock Masses dialog box, right-click into the dialog box to display the shortcut menu.
3. Choose **Import > Replace by clipboard** to import the lock mass data from the clipboard to the lock mass table.

NOTICE The clipboard stores lock mass data as tabulator-separated text files. When the data in the clipboard are not in this format, the command is not available. ▲

4. The Lock Mass Replacement dialog box is displayed. Click **OK** to confirm the import.
5. The Lock Mass Replacement dialog box is closed. The lock mass list in the Lock Masses dialog box displays only the imported lock masses.

❖ **To replace all lock masses from a file**

1. In the Lock Masses dialog box, right-click into the dialog box to display the shortcut menu.

2. Choose **Import > Replace by file content**.
3. The Lock Mass Replacement dialog box is displayed. Click **OK** to confirm the import.The Lock Mass Replacement dialog box is closed. The file selection dialog box is displayed.
   a. Browse to the destination of the import file.
   b. Select the format of the import file.
   c. Select one of the displayed files.
   d. Click **Open** to import the lock masses to Q Exactive GC Tune. The dialog box is closed.
4. The lock mass list in the Lock Masses dialog box displays only the imported lock masses.

   If you have imported lock mass data from an xml file that contained information about lock mass collections and lock mass usage, the dialog box displays the corresponding information.

Creating a new Lock Mass Collection

❖ **To create a lock mass collection**

1. In the Lock Masses dialog box, click the  button.
2. In the New Lock Mass Collection dialog box, enter a name for the new lock mass collection. See Figure 5-4.

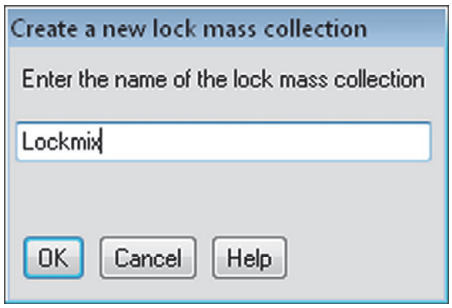


Figure 5-4. Creating a lock mass collection

3. Click **OK** to confirm your input and to close the dialog box.
4. The name of the new lock mass collection appears in the list box of the Lock Masses dialog box.

Renaming a Lock Mass Collection

❖ **To rename a lock mass collection**

1. In the Lock Masses dialog box, select an existing lock mass collection in the list box.
2. Click the  button. The Name Change dialog box is displayed. See Figure 5-5.

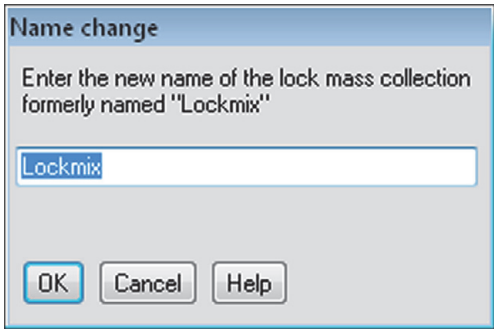


Figure 5-5. Changing the name of a lock mass collection

3. In the text field, replace the old name of the lock mass collection with the new name.
4. Click **OK** to confirm your input and to close the dialog box.
5. In the Lock Masses dialog box, the new lock mass collection is displayed in the list box.

Adding a Lock Mass to a Lock Mass Collection

❖ **To add a lock mass to a lock mass collection**

1. In the Lock Masses dialog box, select an existing lock mass collection in the list box.
2. Select the Use check box in the table row of the lock mass.
3. A ☑ indicates that the lock mass is used in the active lock mass collection.

Removing a Lock Mass from a Lock Mass Collection

❖ **To remove a lock mass from a lock mass collection**

1. In the Lock Masses dialog box, select an existing lock mass collection in the list box.
2. Clear the Use check box in the table row of the lock mass.

3. A ☐ indicates that the lock mass is not used in the active lock mass collection.

Deleting a Lock Mass Collection

❖ **To delete a lock mass collection**

1. In the Lock Masses dialog box, select an existing lock mass collection in the list box.
2. Click the  button.
3. In the Delete Lock Mass Collection dialog box, click **OK** to confirm your action and to close the dialog box.
4. The name of the lock mass collection is removed from the list box of the Lock Masses dialog box.

Performing a System Bakeout

The system bakeout of the mass spectrometer removes unwanted gases or molecules (collected or remaining) from the high vacuum region of the instrument. Ions can collide with those gases or molecules resulting in lower overall sensitivity. Therefore, Thermo Fisher Scientific recommends baking out the instrument if the high vacuum decreases noticeably during routine operation.

Bakeout is mandatory after maintenance or service work is performed in the analyzer region where the system is vented. You should bake out an instrument that has been vented for at least twelve hours (12 hours) before you can start using it again.

In case the system has been vented during a power failure, it is necessary to bake out the system to obtain the operating vacuum.

NOTICE Before you start the bakeout, ensure that the pumps are up and running at their operating speed. If you have just switched on the mass spectrometer, this will take about 10 minutes. To check the pump speed, open the Instruments Status window and expand the Vacuum System node of the Instrument tree. ▲

❖ **To perform a system bakeout**

1. In the Q Exactive GC Tune window, click on the **On/Standby** button to put the instrument in Off condition. (See image in margin.)

2. In the Tasks panel, click 

to display the Vacuum / Bakeout window.

3. Enter the desired baking duration (in hours) into the spin box. The range is 4 to 100 hours. See Figure 5-6.

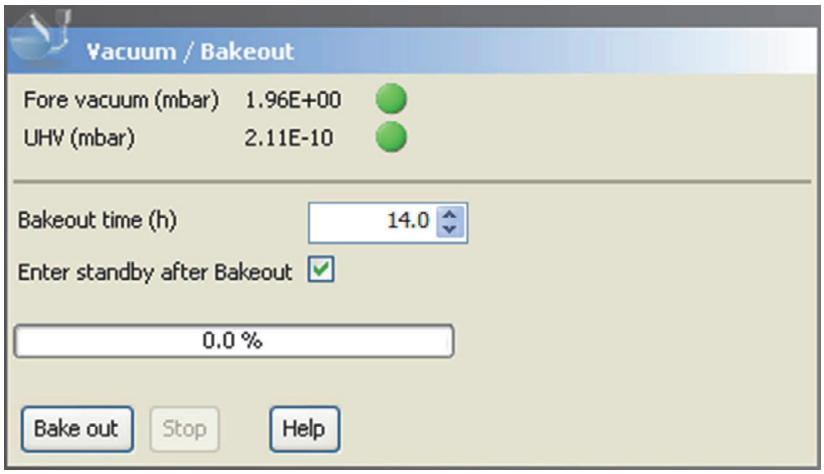


Figure 5-6. Vacuum / Bakeout window

4. Click **Bake out**. A dialog box shows the duration of the baking procedure. See Figure 5-7. Click **Yes** to confirm the message.

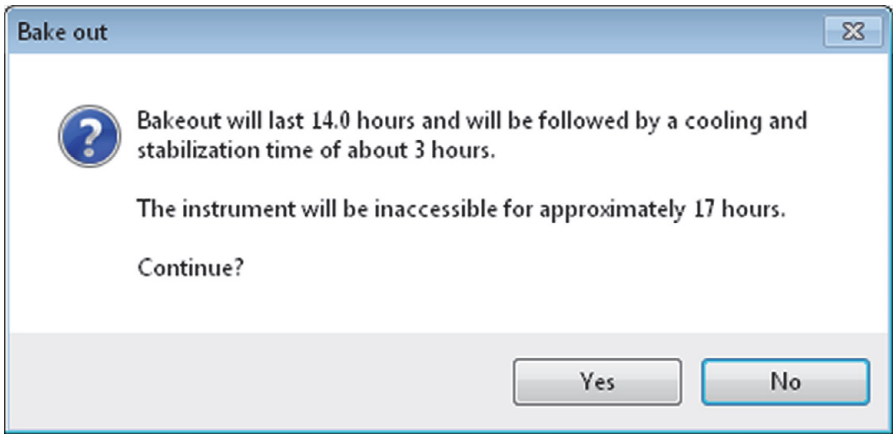


Figure 5-7. Bakeout message box

5. The message box disappears and the baking procedure starts. The instrument indicates the active bakeout procedure by a flashing Vacuum LED at the front side. Additionally, the Q Exactive GC Tune software displays a corresponding message box.

6. The baking of the instrument stops after the preset duration. The Vacuum LED keeps flashing until the cooling and stabilization time (of about 3 hours) is finished.

   Click **Stop** in the Vacuum / Bakeout window to abort the baking routine before the preset time.

Changing Default Settings of Q Exactive GC Tune

With exception of standard users, users can use the instrument status window to change default settings of Q Exactive GC Tune.

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Displaying the High Vacuum Readback

The High Vacuum readback in the Vacuum / Bakeout window is visible only when the ion gauge in the high vacuum chamber is switched on. With exception of standard users, users can switch on this gauge manually.

❖ **To switch on the High Vacuum readback in the Vacuum / Bakeout window**

1. Click 

in the tasks panel to display the Vacuum / Bakeout window.
2. If the instrument status window is not visible, choose **Windows > Instrument Status**.
3. In the instrument status window, click **Instrument > Vacuum System**.
4. Right-click the High Vacuum parameter to display the shortcut menu.
5. Choose **Turn on** to switch on the ion gauge in the high vacuum chamber.
6. The Vacuum / Bakeout window now displays the High Vacuum readback.

To extend the lifetime of the ion gauge in the high vacuum chamber, it is switched off again after a preset time of about thirty minutes.

Changing the Settings for the Performance Status Check

By default, the performance status icon on the toolbar turns yellow 25 hours after the last successful mass calibration or check. With exception of standard users, users can change this value in the System node of the instrument status window according to their mass accuracy requirements.

❖ **To change the settings for the performance status check**

1. If the instrument status window is not visible, choose **Windows > Instrument Status**.
2. In the instrument status window, click **Instrument > System > Configuration settings > Performance Check**. The Mass calibration due time (h) field displays the current validity period of the mass calibration (25 hours, for example). See Figure 5-8.

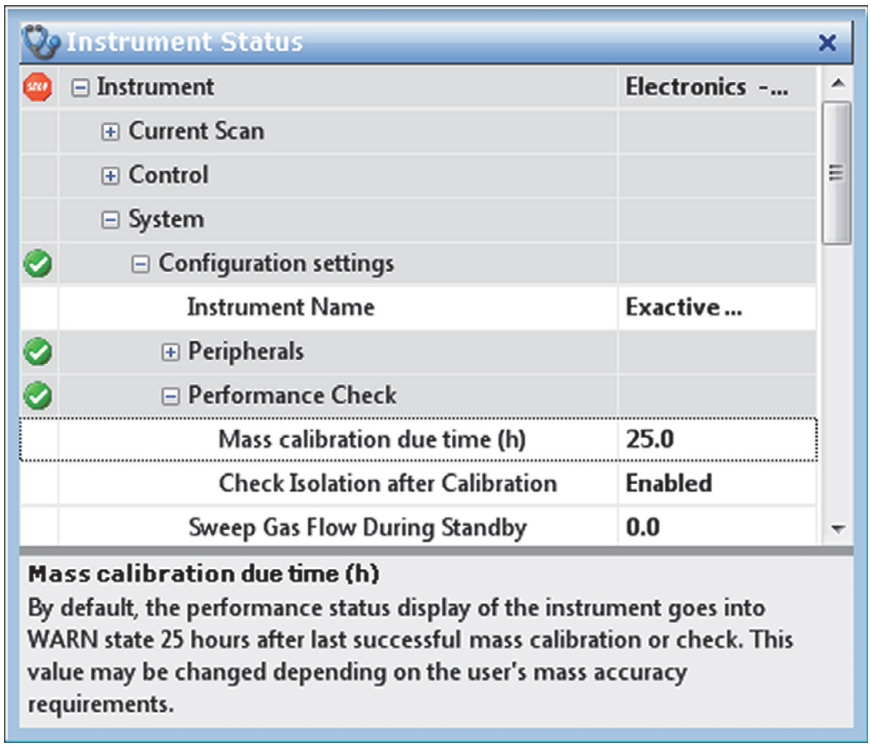

Figure 5-8. Instrument status window—Performance Check node

3. To change the value, right-click the number in the text field to display the shortcut menu.

4. In the Set text field, enter the new validity period (40, for example). See Figure 5-9. You can enter a value between 1 and 500 hours.

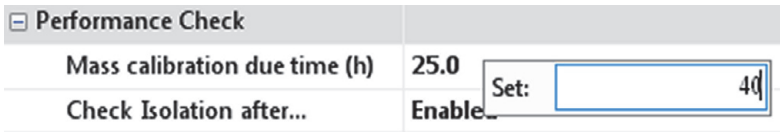

Figure 5-9. Changing the calibration validity period

5. Press the **<Enter>** key to confirm your input. The shortcut menu disappears and the new value is displayed in the instrument status window.

Chapter 6 Reference Information

This chapter provides reference information for the following:

Contents

Log Files

Log files (*.log) are created for a Q Exactive GC mass spectrometer. The default directory for the log files is C:\Xcalibur\system\Exactive\log. The Instrument Configuration window shows the actual path.

Additional log files are created every time the Exactive window service starts. The file name shows date and time of the service start, as shown in the following example:

Thermo Exactive--2015-10-05--15-17-05.log

NOTICE The system automatically deletes all log files that are older than 18 months. ▲

Each line in a log file is a message. A message has several properties that are listed at the beginning of the line, followed by the message body.

Properties are enclosed in brackets. A property is a property name followed by an equal sign followed by the property value. These properties exist:

Abbreviation	Meaning
Time	Time of the message. The current local time is displayed followed by the time offset to UTC.
Acc	Windows™ account name that caused this message; “(none)” is used for an instrument source. Other known values are “ExactiveUser” and “Xcalibur_System”.
User	User name description of the Windows account
Comp	Computer name on which this message was caused, “(none)” is used for an instrument source.
App	Application name that caused this message; “(none)” is used for an instrument source.
PID	Process identifier of the application process
Inst	Instrument affected
Conn	Connection in charge for the communication with the instrument
Type	Type of the message. Message types are “info”, “warning”, “error” and “FATAL error”

Access to the log files is regulated by the Microsoft™ Windows user account or group account. Full access to the log files is granted to Administrators. Members of the Power User group can read, delete, and modify these files. Standard users can read these files.

Tune Files and Calibration Files

Tune files and calibration files contain information for operating the instrument.

Tune Files

Tuning optimizes voltage settings to ensure highest sensitivity. The resulting tune file (*.mstune) is specific to a particular analyte and its analysis settings.

During installation of the instrument, the service engineer creates a tune file default.mstune in the folder C:\Xcalibur\methods\. Use this file as a starting point for optimizing the mass spectrometer for your specific measurement requirements. Use this folder to store your personal tune files.

Calibration File

After having tuned the Q Exactive GC mass spectrometer, calibrate the instrument to ensure the mass accuracy of the measurement results. Calibration parameters are instrument parameters whose values do not vary with the type of experiment. They are stored *automatically* in a calibration file (*.mscal) in the folder C:\Xcalibur\system\Exactive\instrument\msx_instrument_files\.

The file master_cal.mscal contained in this folder is the calibration file used for operating the instrument. It will be overwritten with new calibration values every time a calibration procedure is successful.

NOTICE Never save or change files in the folder C:\Xcalibur\system\Exactive\instrument\msx_instrument_files\! Files in this folder are automatically managed by the instrument software. ▲

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