

Orbitrap Tribrid Series 4.0 SP1 Release Notes

These release notes refer to the features of the Thermo Scientific™ Orbitrap™ Tribrid™ Series version 4.0 instrument control application and provide details of the resolved and known issues that exist in the Thermo Scientific Orbitrap Tribrid Series version 4.0 SP1 instrument control application.

The Orbitrap Tribrid Series (OTS) instrument control software is used to collect high-quality mass spectrometry data on Thermo Scientific Orbitrap Tribrid Series mass spectrometers (MSs). The MS instruments include the Orbitrap Ascend™, Orbitrap Eclipse™, Orbitrap Fusion Lumos™, Orbitrap Fusion™, Orbitrap IQ-X™, and Orbitrap ID-X™. The instrument control application consists of the Tune and Method Editor application packages that enable control of the instrument.

- The Tune application displays acquired mass spectra in a continuous loop and continuously reports the observed values of various instrument parameters that indicate instrument status. Tune is used not only to view spectra but to provide tools to tune and calibrate the instrument for maximum performance with a variety of scan types, scan modes, ion polarities, scan rates, and resolution settings.

The Tune application provides a host of diagnostic functions for easy troubleshooting. You can also use features to manage the USB-connected devices, for example, the external divert valve and the syringe pump. Finally, this application supports report generation so that you can document the outcome of various diagnostics, calibrations, and optimizations.

- In the Method Editor application, you can set up experiments by using the entire complement of scan types, advanced filters, and conditional logic, designing customized sequences of scans to interrogate complicated samples. For example, one method might have a full scan followed by one or more filters and then a data-dependent MSⁿ level scan on the reduced mass list. You have the choice of using your preferred fragmentation technique for MSⁿ scans.

Using the Method Editor application, you can also specify peripheral device controls as part of an experiment. Methods constructed in the Method Editor can be executed in high-level applications such as the Thermo Xcalibur™ data system.

Additionally, with the Thermo Foundation™ Instrument Configuration options, you can set up conditions specific to your instrument and the experiment run.

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For information about installing the Orbitrap Tribrid Series instrument control software, refer to the DVD insert or the installation instructions provided with the application. For information about configuring and using the Orbitrap Tribrid Series systems, refer to the manuals available as PDF files or the Help.

Features

The Orbitrap Tribrid Series 4.0 instrument control software incorporates the following new and improved features:

- Support for the Orbitrap Ascend MS.
- Support for the Auto-Ready ion source, enabling automated and remote calibration. Previously only available on the Orbitrap IQ-X MS; now includes the Orbitrap Ascend MS.
- Orbitrap Ascend MS with an optional HMRⁿ+ license. The option supports an extended mass range up to m/z 16,000.
- The Real-Time Library Search is now available for both peptide and small molecule application modes on the Orbitrap Ascend and Orbitrap Eclipse MSs—including additional functionalities enabling new data-driven acquisition schemes.
- New Real-Time Search functionalities available on the Orbitrap Ascend and Orbitrap Eclipse MSs.
- Updated accepted mass and isolation ranges across all Orbitrap Tribrid Series MS instruments.
- Support for the new AcquireX Ab workflow for peptide mapping through the Xcalibur data system (version 4.6).
- Support for the Thermo Scientific Chromeleon™ Chromatography Data System. The Chromeleon Driver Compatibility Matrix reflects the compatibility tested for combinations of Orbitrap Tribrid Series Instrument Control Software versions, Chromeleon software versions, and instrument models.

For more information and to view the regularly updated compatibility matrix, sign on to <https://support.thermoinformatics.com/downloads/default.aspx>, and select **Chromeleon > Chromeleon > Related Drivers > Driver Compatibility Matrix**.

Usability enhancements:

- Refined management of tables in DIA scans
- Expanded multiplexing to up to 20 ions in msx tSIM/tMS2/DIA scans
- New, improved, and updated templates (including CHIMERYS and AcquireX Ab)
- Enabled acquisition with Dynamic Retention Time functionality in combination with FAIMS

These are the minimum hardware and software configurations required for Orbitrap Tribrid Series 4.0 SP1 operation.

System	Requirements
Computer	<ul style="list-style-type: none">• 3.1 GHz Quad core processor with 16 GB RAM• Video card and monitor capable of 1920 × 1080 resolution• 1 TB hard drive• DVD drive <p>Note To expedite processing time for some AcquireX workflows, Thermo Fisher Scientific recommends 32 GB of memory, even though the minimum requirement for Instrument Control Software version 4.0 SP1 is 16 GB.</p>
Mass spectrometer	Orbitrap Tribrid Series
Software	<ul style="list-style-type: none">• Microsoft™ Windows™ 10 Enterprise Long-Term Servicing Channel (LTSC), 2019, 64-bit• MongoDB Community Version 4.0.28• Thermo Scientific software:<ul style="list-style-type: none">– Foundation 3.1 SP9, or later– Xcalibur 4.6 or later– FreeStyle™ 1.8 SP2 or later

Minimum requirements

General

If there are network, connectivity, or security issues, contact Thermo Fisher Scientific Technical Support to ensure that the security settings, firewalls, and antivirus software are compatible with Thermo Scientific specifications.

Installation

IMPORTANT Always reboot the data system computer when prompted during the installation.

❖ To install the Orbitrap Tribrid Series instrument control software

1. (For new a installation only) Perform the following steps:
 - a. Click **ThermoLauncher.exe** to launch the installation wizard.
 - b. Click **Adobe Reader 20.9** to install the Adobe Acrobat Reader DC application.
 - c. Click **Xcalibur 4.6**. Then follow the installation wizard to install Foundation 3.1 SP9, Xcalibur 4.6, Almanac 1.8, and FreeStyle 1.8 SP2.
2. (To upgrade from Xcalibur 4.4 or 4.5 to Xcalibur 4.6). Perform the following steps:
 - a. Before you upgrade the instrument control software, run the calibration tests in the Tune application. If any failures occur, run the applicable calibration procedure.

Note The installation wizard will install the .NET Framework 4.8, if required. The computer will restart and the Xcalibur installation will continue. If the Xcalibur installation does not automatically continue, relaunch the installation wizard.

d. Continue to [step 4](#).

Note Contact Thermo Fisher Scientific Technical Support if your system does not pass all calibrations.

- b. Launch the Thermo Foundation Instrument Configuration. Then remove all configured instruments from the Configured Devices list.
 - c. Uninstall SII for Xcalibur 1.7 (or earlier) and LC Devices 3.2 SP3 (or earlier), if applicable.
 - d. Click **ThermoLauncher.exe** to launch the installation wizard.
 - e. Click **Adobe Reader 20.9** to install the Adobe Acrobat Reader DC application.
 - f. Click **Xcalibur 4.6** and proceed as follows:
 - i. Follow the installation wizard to uninstall any older versions of FreeStyle, Almanac, Xcalibur, and Foundation.
 - ii. Follow the installation wizard to install Foundation 3.1 SP9, Xcalibur 4.6, Almanac 1.8, and FreeStyle 1.8 SP2.
 - g. Continue to [step 4](#).
3. (To upgrade from Xcalibur 4.3 and earlier to Xcalibur 4.6). Perform the following steps:

Note It is recommended that a Thermo Fisher Scientific field service engineer perform this upgrade.

- a. Before you upgrade the instrument control software, run the calibration tests in the Tune application. If any failures occur, run the applicable calibration procedure.

Note Contact Thermo Fisher Scientific Technical Support if your system does not pass all calibrations.

- b. Launch the Thermo Foundation Instrument Configuration. Then remove all configured instruments from the Configured Devices list.
- c. Uninstall any existing Orbitrap Tribrid Series Instrument Control Software.
- d. Uninstall SII for Xcalibur 1.7 (or earlier) and LC Devices 3.2 SP3 (or earlier), if applicable.
- e. Uninstall Xcalibur.
- f. Uninstall Foundation.
- g. Click **ThermoLauncher.exe** to launch the installation wizard.

- h. Click **Adobe Reader 20.9** to install the Adobe Acrobat Reader DC application.
- i. Click **Xcalibur 4.6**. Then follow the installation wizard to install Foundation 3.1 SP9, Xcalibur 4.6, Almanac 1.8, and FreeStyle 1.8 SP2.

Note The installation wizard will install the .NET Framework 4.8, if required. The computer will restart and the Xcalibur installation will continue. If the Xcalibur installation does not automatically continue, relaunch the installation wizard.

4. (Optional) Install SII for Xcalibur 1.7 (or later) and or LC Devices 3.2 SP3 (or later).
5. Install Orbitrap Tribrid Series 4.0 SP1.
6. Launch the Thermo Foundation Instrument Configuration, then add and configure the system devices.
7. Open the Tune application and follow the Upgrade Diagnostic prompts:
 - a. Run **Upgrade Diagnostic – No Sample**.
 - b. Infuse the calibration solution.
 - c. With a stable spray of calibration solution, run **Upgrade Diagnostic – Calmix/FlexMix**.
8. After completing the upgrade diagnostics, run the Check Calibration tests in the Tune application. If any failures occur, run the applicable calibration procedure.

Note Contact Thermo Fisher Scientific Technical Support if your system does not pass all calibrations.

Resolved issues

Table 2 lists defects that were resolved between the Orbitrap Tribrid Series 4.0 and the Orbitrap Tribrid Series 4.0 SP1 applications.

The table excludes Help issues and any cosmetic fixes. In some cases, the abstract has been amended or extended from the original to better describe the reported issue. Both an engineering fix and follow-up testing (verified by our product evaluation department) have resolved these issues.

Table 1. Resolved issues between Orbitrap Tribrid Series 4.0 and Orbitrap Tribrid Series 4.0 SP1

Item ID	Abstract
357099	Addressed an issue in which injection time may oscillate during LC-MS runs of intact proteins on Orbitrap Ascend system
357152	Addressed an issue in which Discharge Pressure Check fails due to readback error on Orbitrap Fusion ETD system
357636	Modified accepted range for energy dependency on Orbitrap Ascend system
358602	Addressed incorrect application of isolation waveforms in MS ⁿ experiments when ion trap isolation is used for more than one stage
363649	Addressed an issue in which new changes in “Peak Selection and Threshold Settings” properties are not properly synchronized across multiple Real-Time Search filters after re-opening a method previously saved with “Use Common Peak Selection and Threshold Settings” option selected

Table 2. Resolved issues between Orbitrap Tribrid Series 3.5 SP2 and Orbitrap Tribrid Series 4.0 (Sheet 1 of 2)

Item ID	Abstract
219580	Corrected IQ-X 7.5k RP default maximum injection time to 11ms (instead of 22ms).
262323	Addressed an issue preventing the display of Method Editor menu bar in Chromeleon.
266405	Modified reference volume of Auto-Ready vial to 4mL (instead of 5mL).
281784	Addressed an issue related to mislabeled Contact Closure options in Instrument Configuration.
296415	Addressed an issue related to the validation of methods including Dynamic Exclusion filter.
315166	Modified messages in calibration reports generated with Auto-Ready Ion Source.
320242	Added ‘Calmix Evaluation Toggle’ in Diagnostics.

Known issues

Table 2. Resolved issues between Orbitrap Tribrid Series 3.5 SP2 and Orbitrap Tribrid Series 4.0 (Sheet 2 of 2)

Item ID	Abstract
326968	Addressed an issue in which workstation method editor could not be launched without Foundation installed (affects Chromeleon Enterprise).
328502	Added 'Set Spray Voltage' in Diagnostics for Auto-Ready.
276030	Addressed an issue in which ion transfer efficiency may be reduced during acquisition from Tune following the execution of a "View EASY-ETD Reagent" scan in Tune ("Define Scan" tab).
300995	Addressed an issue in which ion current maybe lower than expected in polarity switch experiments.
279631	Addressed a memory consumption issue with methods using Assisted Collision Energy in conjunction with Easy-IC.
279373	Addressed an issue related to inaccurate HCD fragment efficiency calculation.
215549	Addressed an issue in which multiple API licenses are reported in Tune.
269263	Addressed an issue related to FAIMS Pro fan speed out of range.
270254	Addressed an issue related to FAIMS DV status check.

Recommended recovery actions

- For issues such as connectivity problems, restart the application.
- For issues that arise during data acquisition, restarting the application might not ensure a complete recovery (e.g. restarting the Xcalibur data system). In this case, restarting the data system computer typically resolves the issue; some devices displaying error conditions may also require a power cycle.
- Reinstalling the software or the operating system is not recommended as a general fix. Please contact Thermo Fisher Scientific Technical support for assistance.

Feature requests and other removed items

- We do not include issues where there is insufficient information logged to successfully reproduce the reported problem.
- We do not list feature requests as software issues, regardless of the reported significance or severity of the request. Product managers evaluate logged feature requests for future releases.
- We report only discrepancies in the documented software as known issues.

Terminology

Table 3. Severity levels

Severity	Interpretation
Crash/Data Loss	A problem that renders the system unusable because either an entire function is unusable and no workaround exists, or use of the current system compromises data integrity or results in data loss. Catastrophic problems also include significant and non-obvious quantitative errors, and all human and instrument safety issues.
Major Problem	A serious issue that does not affect data integrity (meaning data loss, corruption of data, or the wrong answer), but affects the customer's ability to use the product as designed. It can be a failure, design issue, or documentation error or omission. A workaround might or might not exist.
Minor Problem	A minor error or poor behavior of a product feature. There is probably a workaround.
Cosmetic	An issue that has a limited effect on customer usage of the product; for defects with visibility so low that a customer might never see it; or for ease of use issues or other items not causing any performance degradation.

Table 4. Risk levels

Risk	Interpretation
High	Occurrence is likely to happen and can compromise operation.
Medium	Occurrence is uncommon, but could compromise operation if it occurs.
Low	Issue is minor; however, the software could operate differently from a user's expectations. A workaround might be available.
No Risk	This issue causes no problem but is commonly an inconsistency or cosmetic issue.

Known defects

Table 5 contains a known defect in the software, categorized by software section, with a brief abstract and information related to the defect's severity and risk, if applicable. The Item ID is the internal number assigned to the issue, if applicable. Product management assesses risk, which can differ significantly from the reported severity.

Table 5. Known software issues (Sheet 1 of 2)

Item ID	Severity	Abstract	Risk
57094 (73337)	Minor Problem	Raw files under Windows 10 do not open properly by using the View Raw File button or by directly double-clicking the raw file. To open a raw file, navigate to it, right-click the file, and then browse to the FreeStyle application. Select the check box to always open a raw file with FreeStyle. FreeStyle is now the default application to open raw files.	Low
210023	Minor Problem	When running Orbitrap Tribrid Series software under Chromeleon, an error message may appear in the Chromeleon audit trail: "Double Property Bearing_Temperature cannot be updated with TP_RUNNING". The error occurs when first opening Chromeleon or when adding the mass spectrometer to the Chromeleon instrument configuration. Note This error does not affect the operation of the Orbitrap Tribrid instrument.	Low
330846	Minor Problem	Sequence submission validation in Chromeleon 7.2.10 MUF with Orbitrap Tribrid Series 4.0 fails when using a MS method initially created from a previous version of Orbitrap Tribrid Series and a different Tribrid model even after modifying some parameters and re-saving the method. Workaround: Make a copy of the method using the "save as" function. Copied method passes sequence submission validation step and acquisition is performed importing a method created.	Low
331725	Minor Problem	When creating a method with parallel branches and SPS-ddMS3 as top scan node in at least one of them, unchecking Synchronous Precursor Selection property has no effect on the ddMDMS3 scan user interface. Workaround: Delete the ddMS3 scan and add the scan node again.	Low

Table 5. Known software issues (Sheet 2 of 2)

Item ID	Severity	Abstract	Risk
332582	Minor Problem	<p>After downgrading from Orbitrap Tribrid Series (OTS) 4.0 to an older version, reports are no longer generated. This is because the OTS 4.0 installer updates the MongoDB from version 4.06 to 4.0.28, but the TNG uninstaller does not uninstall the MongoDB during a downgrade.</p> <p>Workaround: Manually uninstall the MongoDB when downgrading OTS 4.0 to an older version by using the MongoDB uninstaller provided with OTS 4.0. Follow these steps:</p> <ul style="list-style-type: none"> • Double click on MongoDBUninstaller.exe to uninstall MongoDB from the Programs folder • To uninstall the MongoDB data and log files located at: C:\ProgramData\Thermo\TNG\MongoDB <ul style="list-style-type: none"> – Open a command prompt – Navigate to the folder containing the uninstaller – Type MongoDBUninstaller -f and press Enter (e.g., C:\[folder]\MongoDBUninstaller -f) 	Low
336445	Minor Problem	<p>On Orbitrap Ascend and Orbitrap IQ-X MSs controlled under Chromeleon, changing the System Self-Check Options triggers a configuration mismatch error when running the next analysis.</p> <p>Workaround: Remove and then re-add the mass spectrometer in instrument configuration as indicated in the error message.</p>	Low
347016	Minor Problem	<p>Performing MSⁿ acquisition (with n > 8) may result in firmware crash and acquisition freeze, thus requiring instrument reset.</p> <p>Workaround: Do not perform MSⁿ acquisition with n > 8.</p>	Low
344407	Minor Problem	<p>When operating ion source with Vaporizer Temperature set above 100 °C while "Enable Minimum Gas Flow Requirement for Ion Source" is disabled in Instrument Configuration, there is a risk to damage the heater if Auxiliary gas is too low, i.e., below 3 units (arb).</p> <p>Workaround: Set Auxiliary gas higher than 3 units (arb) when operating ion source with Vaporizer Temperature above 100 °C, or activate "Enable Minimum Gas Flow Requirement for Ion Source" in Instrument Configuration.</p>	Low
362116	Minor Problem	<p>Updating ftboot files of Orbitrap Ascend (e.g., as part of the installation of Orbitrap Tribrid Series version 4.0 SP1) can cause a crash.</p> <p>Workaround: Restart the instrument (by resetting the electronics) after the installation of Orbitrap Tribrid Series version 4.0 SP1</p>	Low

Trademarks

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Orbitrap Ascend, Orbitrap Eclipse, Orbitrap Fusion Lumos, Orbitrap IQ-X, Orbitrap Fusion, Orbitrap ID-X, Orbitrap, Tribrid, Thermo Scientific, Xcalibur, FreeStyle, AcquireX, Almanac, Chromeleon, and FlexMix are trademarks of Thermo Fisher Scientific Inc. and its subsidiaries.

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