



Mass spectrometry

## Scale up your science

### Orbitrap Ascend Editions Tribrid mass spectrometer

#### Benefits

- Improved capture of labile compounds while maintaining optimal ion transmission
- Exceptional performance with shorter gradients due to higher MS/MS sensitivity and faster scan rates
- Unmatched depth, accuracy, and precision for Tandem Mass Tag™ (TMT™) analysis
- Optional electron transfer dissociation (ETD) for orthogonal fragmentation capabilities
- The fully integrated Auto-Ready ion source allows for complete, unattended system calibration, without the need for manual hardware reconfiguration
- Full experimental flexibility with revolutionary instrument control software
- A wide range of optional functionalities for unprecedented versatility
- Common user interface with Thermo Scientific™ Orbitrap Astral™, Thermo Scientific™ Orbitrap Exploris™ and Thermo Scientific™ TSQ™ Triple Quadrupole mass spectrometers

Unleash the power of multiomics and native protein characterization with the Thermo Scientific™ Orbitrap™ Ascend Editions Tribrid™ mass spectrometers. Quantify more samples at lower concentrations using intelligent acquisition and faster Real-Time Search. Explore new frontiers with a cutting-edge hardware design featuring two ion routing multipoles for greater coverage. Characterize even the largest biopharmaceuticals with an optional mass range up to  $m/z$  16,000. Unlock the potential of alternate fragmentation techniques to decipher the most challenging molecules. Streamline instrument setup with automated Auto-Ready ion source calibration and pre-built method templates.



Be ready for tomorrow's most challenging demands with the Orbitrap Ascend Tribrid mass spectrometer

## Key capabilities

- Improved high-sensitivity atmospheric pressure interface combines a high-capacity transfer tube and a modified electrodynamic ion funnel, providing an increased ion flux and lower limits of detection for a wider range of molecules
- Active Ion Management (AIM+) Technology that includes the QR5 segmented quadrupole mass filter with hyperbolic surfaces, maximizes ion transmission to achieve unprecedented levels of qualitative and quantitative performance
- Dual Ion Routing Multipoles (IRMs) improves ion transfer, capture and dissociation
- Automated, remote, and hands-free calibration with the Auto-Ready ion source, allowing the user to spend more time obtaining results and less time on instrument setup
- Optional Native MS mode provides expanded mass range capabilities for comprehensive MS<sup>n</sup> characterization of native proteins and protein complexes
- Extended quadrupole isolation range with Native MS option for higher selectivity at high *m/z*, including native structural analytes
- Real-Time Search of a database or a spectral library enables faster data acquisitions and improved specificity, significantly increasing analytical performance, including TMT-based quantification experiments
- Thermo Scientific™ TurboTMT mode improves HRAM MS<sup>n</sup> acquisition rates in TMT experiments
- Thermo Scientific™ Precursor Fit filter enables precursor ion selection for a defined specificity
- Enhanced Vacuum Technology improves Thermo Scientific™ Orbitrap™ mass analyzer performance
- Environmentally friendly oil-free dry pumps for less maintenance and leak-free vacuum performance
- Improved accumulation and transmission of high molecular weight ions to the ion trap and Orbitrap mass analyzers
- Unique Tribrid architecture includes a quadrupole mass filter, dual-pressure linear ion trap and Orbitrap mass analyzers, allowing for high acquisition rates to provide maximum experimental flexibility for dissociation and detection of fragment ions
- DDA MS/MS acquisition rates of up to 45 Hz with 7500 resolution @ *m/z* 200 for Orbitrap mass analyzer and 50 Hz in the linear ion trap experiments
- Orbitrap resolution up to 480,000 FWHM at *m/z* 200
- Large surface area ion trap detector for significantly improved robustness and lifetime
- Full parallelization of MS and MS<sup>n</sup> analyses with Dynamic Maximum Injection Time
- Synchronous Precursor Selection (SPS) for MS<sup>n</sup> experiments
- Optional compact Electron Transfer Dissociation (ETD) and optional Proton Transfer Charge Reduction (PTCR) ion source based on Townsend discharge with extremely stable anion flux for improved usability and reagent longevity
- Multiple fragmentation techniques: collision-induced dissociation (CID), higher-energy collisional dissociation (HCD), optional electron-transfer dissociation (ETD, including electron-transfer higher-energy collision dissociation, or EThcD, and electron-transfer collision-induced dissociation, or ETciD) and optional ultraviolet photodissociation (UVPD), available at any stage of MS<sup>n</sup> with detection in the Orbitrap mass or linear ion trap analyzers
- Optional Proton Transfer Charge Reduction (PTCR) simplifies MS<sup>n</sup> spectra of multiply charged analytes
- Advanced Peak Determination (APD) for improved precursor annotation in data-dependent experiments
- Universal method maximizes peptide identifications without method optimization when sample concentration is unknown
- Intuitive, flexible drag-and-drop instrument method editor simplifies method development and creation of unique analytical workflows
- Extensive pre-built and optimized method templates that are fully customizable for a variety of application workflows
- Thermo Scientific™ AcquireX™ data acquisition software workflow provides all-inclusive, automated small molecule sample profiling on an LC timescale
- Streamlined calibration routines with improved Orbitrap calibration for ions with *m/z* <200

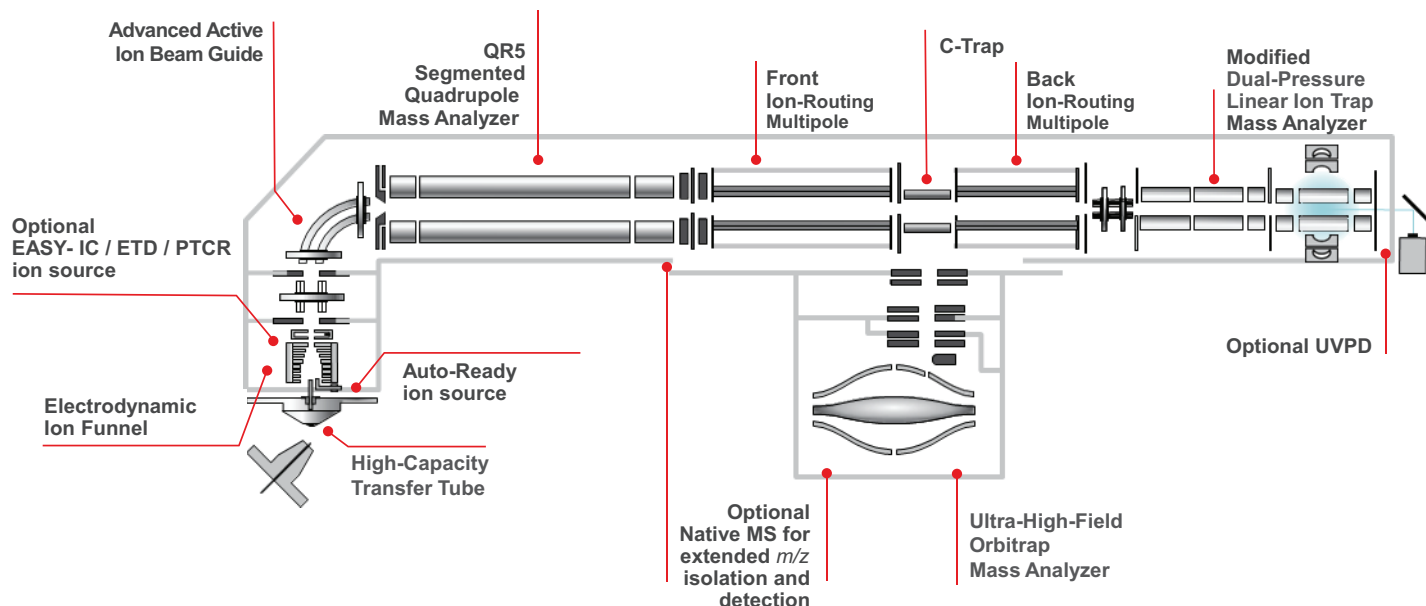


Figure 1. Orbitrap Ascend MultiOmics Tribrid mass spectrometer ion path

## Hardware features

### Active Ion Management (AIM+) Technology

AIM+ maximizes ion transmission, from injection to detection, using a novel hardware design for precise management of electrical fields and removal of noise to achieve unprecedented levels of quantitative performance.

### Ion source

#### Thermo Scientific™ Ion Max NG™ ion source

- Adjustable heated electrospray ionization (HESI) probe provides ultimate sensitivity
- Flow rates 1–2000  $\mu\text{L}/\text{min}$
- Designed for maximum performance for a broad range of molecular ions
- Enhanced ruggedness
- Sweep gas reduces chemical noise
- Optional APCI probe compatible with liquid flow rates of 50–2000  $\mu\text{L}/\text{min}$  without splitting; the APCI probe can be upgraded to optional APPI capability

#### Thermo Scientific™ Auto-Ready ion source

- Integrated source with dedicated emitter and ion transfer tube simplifies maintenance and calibration
- Trigger on-demand or scheduled system checks and calibrations without disruption to your LC-MS configuration
- Calibration and maintenance results are controlled and reported directly in the instrument control software interface

- Internal Thermo Scientific™ Pierce™ FlexMix™ Calibration solution consumable lasts  $\geq 3$  months with weekly mass calibration and monthly system calibration, plus intermittent checks

### Generation V ion optics and mass analyzers

The new API interface consists of a High-Capacity Transfer Tube (HCTT) and an Electrodynamic Ion Funnel (EDIF)

#### High-capacity ion transfer tube (HCTT)

- The HCTT increases ion flux into the vacuum system for improved sensitivity
- Vent-free maintenance

#### Electrodynamic Ion Funnel

- A modified ion funnel captures the ions under more gentle conditions, improving the transfer of labile compounds while maintaining optimal transmission
- Automatic tuning results in broad ion transmission curves with reduced ion losses, increasing sensitivity

#### Advanced active ion beam guide (AABG)

- AABG with an axial field and low-pass filter prevents neutrals and high-velocity clusters from entering the quadrupole mass filter

#### QR5 segmented quadrupole mass filter with hyperbolic surfaces

- Next generation quadrupole mass filter with 5.25 mm field radius delivers industry leading ion isolation across  $m/z$  50–2000 range for efficient precursor selection with  $m/z$  0.4–1200 isolation width.

## New front ion-routing multipole

- Ion storage capabilities
- Higher-energy collisional dissociation (HCD) on MS<sup>2</sup>
- Variable pressure control from 0.5 to 20 mTorr with Native MS option, for small and large molecule workflows
- Efficient and reliable ion transfer between the ion-routing multipole, Orbitrap mass analyzer and linear ion trap mass analyzer

## Orbitrap mass analyzer

- Next-generation ultra-high-field Orbitrap mass analyzer
- Low noise detection pre-amplifier
- Automatic calibration of all ion transfer parameters

## Dual-pressure linear ion trap mass analyzer

- MS<sup>n</sup>, n = 1 through 10 using any available fragmentation technique
- Extended front section of the high-pressure cell for improved ETD and PTCR reaction control
- Precursor isolation width range from 0.2-4000 *m/z* for precursors between 50 and 4000 *m/z* for normal mass range isolations
- Collision-induced dissociation (CID)
- Optional ETD/ET<sub>h</sub>CD/ET<sub>c</sub>iD and UVPD fragmentation techniques
- Optional PTCR ion-ion reaction
- Low pressure cell for improved scan rates, resolving power, and mass accuracy
- Dual-dynode detector with high linear dynamic range for improved quantification, and a large surface area for increased lifetime and robustness

## Vacuum system

- Multiple turbomolecular pumps create vacuum stages ranging from >1 Torr down to ~1x10<sup>-10</sup> Torr
- Stainless steel and aluminum high-vacuum analyzer chambers
- Enhanced vacuum technology reduces the pressure in the ultra-high vacuum region to <10<sup>-10</sup> Torr
- Improved Orbitrap mass analyzer performance at a wider range of collision gas settings

## Analog inputs

- Channel 1 analog input (0–10 V); Channel 2 analog (0–2 V)

## Optional hardware

### Thermo Scientific™ NanoSpray Flex NG™ ion source

- Supports static and dynamic electrospray experiments
- Compatible with liquid flow rates of 50 nL/min–2 μL/min

### Thermo Scientific™ EASY-Spray™ ion source

- Maximum nanoelectrospray performance with no need for adjustments

### Thermo Scientific™ EASY-ETD™ ion source (optional)

- Generates fluoranthene anions for ETD
- Townsend discharge provides extremely stable and robust ionization
- Compact size, located entirely within the footprint of the instrument
- Active reagent ion filtering using the quadrupole mass filter
- Charge-state dependent calibration of reaction time to maximize spectral quality
- Improved dynamic range and signal-to-noise ratio (S/N) for fragments using high dynamic range EASY-ETD

### Thermo Scientific™ PTCR ion source (optional)

- Extension of the EASY-ETD ion source
- Generates perfluoroperhydrophenanthrene (PFPP) ions for subsequent gas-phase ion-ion reactions
- Simplifies interpretation of complex spectra by decreasing the charge state of precursors and/or product ions

### Thermo Scientific™ EASY-IC™ ion source (optional)

- Generates internal calibrant ions for real-time mass calibration of spectra in positive and negative modes
- Provides <1 ppm RMS drift over 24 hours, measured with FlexMix Calibration Solution

### Thermo Scientific™ UVPD ion source (optional)

- Includes a Class 1, 213 nm CryLaS laser system with 2.5 kHz repetition rate, delivering >1.2 μJ per pulse
- Performs dissociation of precursors at any stage of MS<sup>n</sup>, with detection in either the ion trap or Orbitrap mass analyzer
- Compact size, located entirely within the footprint of the instrument

### Thermo Scientific™ FAIMS Pro Duo interface (optional)

- Performs online gas-phase fractionation based on differential ion mobility
- Optimized performance for 100 nL/min to 1 mL/min flow rates

### Native MS option

- Extends the mass range to  $m/z$  16,000 for detection of precursor and product ions in the Orbitrap mass analyzer
- Ion isolation up to  $m/z$  8,000 in the ion trap with isolation widths  $> m/z$  100
- Extends the ion isolation range in the quadrupole to include  $m/z$  2,000 to 8,000 with  $m/z$  5 to 3,000 isolation width

## Software features

### Data system

- High-performance PC with Intel® microprocessor
- High-resolution LED color monitor
- Microsoft® Windows® 10 operating system

### Thermo Scientific™ Xcalibur™ software

- Xcalibur software is the control software for the next-generation Thermo Scientific mass spectrometer portfolio
- Accelerates familiarization and reduces training needs

### Orbitrap Ascend Tribid MS instrument control software

- Tune application for instrument calibrations and checks, diagnostics, and manual data acquisition
- Method editor with a comprehensive application-specific template library, method setup supported by tooltips, and a drag-and-drop user interface to facilitate method development

## Optional software

### Thermo Scientific™ Proteome Discoverer™ software

Flexible, expandable platform for the qualitative and quantitative analysis of proteomics data with intelligent INFERYYS™ Rescoring and the novel CHIMERYS™ intelligent search algorithm by MSAID.

### Thermo Scientific™ ProSightPD™ software

Processing node within Proteome Discoverer software that allows execution of ProSight searches of top-down and middle-down data.

### Thermo Scientific™ BioPharma Finder™ software

Integrated solution for protein-based biotherapeutic analysis through molecular and sub-structural data processing utilizing deconvolution and predictive fragmentation pattern algorithms.

### Thermo Scientific™ Compound Discoverer™ software

Platform for small-molecule structural identification, qualitative, and quantitative data analysis.

## Operation modes

### Top speed

Maximizes the number of high-quality MS<sup>n</sup> spectra in each cycle by intelligently scheduling MS and data-dependent MS<sup>n</sup> scans based on a user-defined time between adjacent survey spectra.

### TopN

Enables a user-defined maximum number of high-quality MS<sup>n</sup> spectra from each cycle.

### Dynamic maximum injection time

Maximizes the amount of high-quality data acquired by synchronizing operations of ion injection, ion transfers within the instrument, and operation of the linear ion trap and Orbitrap mass analyzers.

### Advanced Peak Determination (APD)

Precursor annotation algorithm for improved charge state assignment to increase the number of precursors available for data-dependent analysis.

## Exclusive technologies

### Dynamic scan management

Enables intelligent, real-time scheduling, parallelization, and prioritization of acquisition events, and selection, sorting, and routing of precursors to different fragmentation modes and analyzers based on user-selected parameters, including precursor  $m/z$ , intensity, and/or charge.

### Multiple dissociation options

Precursor dissociation can be performed in the ion-routing multipoles (HCD), the linear ion trap (CID, optional ETD/ETciD, optional UVPD) or both (ETHcD), with product ion detection in the linear ion trap or Orbitrap mass analyzer at any stage of MS<sup>n</sup> analysis.

### Automatic gain control

Ensures that the ion-routing multipole is always filled with the optimum number of ions for any acquisition type.

### Proton transfer charge reduction option

Performs ion-ion reaction to reduce charge states of selected precursors or product ions and can be applied at any stage of MS<sup>n</sup> analysis.

### Real-time database search

Searches user-defined database in real time to then select identified/or not identified precursors for further MS<sup>n</sup> experimentation. Example: Selection of identified precursors only for SPS MS<sup>3</sup> quantitation increases the accuracy and proteome coverage of TMT experiments.

### Dual mass range quadrupole isolation

Optimal isolation performance at togglable low and high *m/z* range using enhanced, novel dual residence RF capabilities.

### Real-Time Library Search

Intelligent acquisition provides higher confidence with real-time spectral matching against a local, customizable spectral library

for decision-based triggering of MS<sup>n</sup> acquisition events. It enables library directed MS<sup>n</sup> data acquisition to increase confidence in annotation, including structurally related compounds and improve structured assignment of isomeric species, unknown characterization, and other applications.

### TurboTMT mode

Novel processing mode powered by Phase-Constrained Spectrum Deconvolution Method (ϕSDM) designed to improve the acquisition rate of TMT experiments.

### Precursor Fit filter

Allows selection of precursors with defined specificity.

## Performance specifications

Orbitrap mass range	Standard mass range <i>m/z</i> 40–2000, mid-mass range <i>m/z</i> 200–6000, and optional Native MS mass range <i>m/z</i> 500–16,000
Orbitrap resolution	Standard resolution settings range from 7500 to 480,000 (FWHM) at <i>m/z</i> 200, with isotopic fidelity up to 240,000 FWHM
Acquisition rate*	Orbitrap MS <sup>n</sup> acquisition rates up to 45 Hz when measured using a data-dependent experiment with FlexMix calibration solution, MS and HCD MS/MS resolution setting of 7500 @ <i>m/z</i> 200, with a maximum ion fill time of 11 milliseconds; linear ion trap MS <sup>n</sup> acquisition rates up to 50 Hz using a data dependent experiment with FlexMix calibration solution, Orbitrap mass analyzer resolution of 240,000 and HCD MS/MS ion trap detection range of <i>m/z</i> 200–1400, with a MS/MS maximum ion fill time of 10 milliseconds
Orbitrap mass accuracy*	External calibration achieves <3 ppm RMS drift over 24 hours, measured with FlexMix; internal calibration achieves <1 ppm RMS drift over 24 hours, measured with FlexMix
MS/MS Electrospray Ionization (ESI) ion trap sensitivity	2 µL of a 50 fg/µL solution of reserpine (100 femtograms total) injected at a flow rate of 500 µL/min produces a minimum S/N ratio of 200:1 for the transition of the isolated protonated molecular ions at <i>m/z</i> 609 to the largest two product ions, <i>m/z</i> 397 and <i>m/z</i> 448, when the mass spectrometer is operated at unit resolution in the full scan MS/MS mode, <i>m/z</i> 165–615; the test requires the HESI II probe
Dynamic range	>5000 within a single Orbitrap mass spectrum
MS scan power	Collects MS <sup>n</sup> , for n = 1 through 10, detected in either the Orbitrap or linear ion trap mass analyzer
Synchronous precursor isolation	Samples up to 20 precursors per MS <sup>n</sup> scan
Precursor multiplexing	Up to 20 precursors can be multiplexed using the quadrupole mass filter and front ion-routing multipole to perform selected ion monitoring or tandem MS
Quadrupole isolation range	Standard quadrupole with 5.25 mm field radius isolates from <i>m/z</i> 50-2000 with isolation width of <i>m/z</i> 0.4-1200. Optional Native MS extends the quadrupole isolation range to <i>m/z</i> 2000-8000 with isolation width range of <i>m/z</i> 5-3000
Polarity switching	One full experimental cycle acquired in <1.1 seconds where the cycle consists of acquiring one full MS scan in positive and negative polarity at a resolution setting of 30,000
ETD option efficiency	Reaction with ETD anions dissociates the isolated 2 <sup>+</sup> MRFA precursor ( <i>m/z</i> 262) to generate a series of fragments that have a summed relative abundance ≥15% to the precursor measured in the linear ion trap, using FlexMix solution infused at 6 µL/min
UVPD option efficiency	Exposure of 100 ms dissociates the isolated caffeine molecular ion ( <i>m/z</i> 195) to generate the <i>m/z</i> 138 product ion with a relative abundance of ≥25% to the precursor measured in the linear ion trap, using FlexMix solution infused at 6 µL/min
PTCR option efficiency	Reaction time of 100 ms causes isolated MRFA 2 <sup>+</sup> molecular ion ( <i>m/z</i> 262) to convert to a charged reduced ion at <i>m/z</i> 524, with a relative abundance of ≥25% to the precursor measured in the linear ion trap, using FlexMix calibration solution infused at 6 µL/min

\* Under defined conditions

## Advanced data-dependent experiments

### TMT SPS MS<sup>3</sup> aided by Real-Time Search

TMT SPS MS<sup>3</sup> experiment with real-time protein database search to perform SPS MS<sup>3</sup> only on identified precursors.

### Universal method

Powered by Dynamic Maximum Injection Time, ensures the best results from samples with unknown concentrations.

### Product-ion triggered-MS<sup>n</sup>

Fragment ion or neutral loss-triggered MS<sup>n</sup> experiment, including at the same MS<sup>n</sup> level.

### Isolation offset

Custom centering of the precursor isolation window, optimized for broad isotopic distributions.

### Quantification workflow

Synchronous acquisition of HRAM SIM with full scan MS/MS in the linear ion trap for precursor identity confirmation.

### Thermo Scientific™ SureQuant™ internal standard (IS) targeted protein quantitation workflow

Enhanced PRM experiment that leverages internal standards to guide and automatically maximize outcomes and data quality for real-time targeted proteomics analysis.

## Installation requirements

### Power

- 230 Vac ±10% single phase, 50/60 Hz, with ground, 2x outlets at 15 A each
- 120 or 230 Vac single phase with earth group for the data system

### Gas

- Ultra-high purity Helium (99.999%) with less than 1 ppm each of water, oxygen, and total hydrocarbons
- High purity Nitrogen (99.5% pure, flow rate 15 L/min) gas supply for the API source and ion routing multipole (IRM)

### Dimensions (w × d × h)

- 1270 × 805 × 703 mm (50 × 31.7 × 27.7 in)

### Weight

- 180 kg (400 lb) without data system, vacuum rough pumps and optional items

### Environment

- System averages 1400 W (4800 Btu/hr) and rough pump averages 570 W (1950 Btu/hr) output when considering air conditioning needs
- Operating environment must be 16–26 °C (59–78 °F) and relative humidity must be 50–80% with no condensation
- Optimum operating temperature is 18–21 °C (65–70 °F)

### EASY-IC/ETD/PTCR ion source options

- Nitrogen supply for EASY-IC/ETD ion source options: Ultra-high purity nitrogen (UHP, 99.999%) with less than 1 ppm each water and oxygen

### FAIMS Pro Duo interface option

- Nitrogen supply for FAIMS Pro Duo interface is 99.5% pure nitrogen at >20 L/min (100 psi)

Learn more at [thermofisher.com/OrbitrapAscend](https://thermofisher.com/OrbitrapAscend)

**Research Use Only – Not For Diagnostic Procedures** © 2024 Thermo Fisher Scientific Inc. All rights reserved. Intel is a registered trademark of Intel Corporation. Microsoft and Windows are registered trademarks of Microsoft Corporation. CHIMERYS and INFERYYS are trademarks of MSAID. TMT is a trademark of Proteome Sciences. All other trademarks are the property of Thermo Fisher Scientific and its subsidiaries. This information is presented as an example of the capabilities of Thermo Fisher Scientific products. It is not intended to encourage use of these products in any manner that might infringe the intellectual property rights of others. Specifications, terms and pricing are subject to change. Not all products are available in all countries. Please consult your local sales representative for details.

PS002899 0524S

**thermo** scientific