


 Mass spectrometry

Scale up your biopharma experiments

Orbitrap Ascend BioPharma Tribrid mass spectrometer

Benefits

- Exceptional performance with shorter gradients due to higher MS/MS sensitivity and faster scan rates
- Recommended electron transfer dissociation (ETD) and UVPD for orthogonal fragmentation capabilities
- Recommended Native MS option for comprehensive analysis of native protein complexes
- Recommended proton transfer charge reduction (PTCR) for enhanced ion-ion interactions
- The fully integrated Auto-Ready ion source allows for system calibration without hardware reconfiguration
- Full experimental flexibility with revolutionary instrument control software
- Common user interface with Thermo Scientific™ Orbitrap Astral™, Thermo Scientific™ Orbitrap Exploris™ and Thermo Scientific™ TSQ™ Triple Quadrupole mass spectrometers

With new capabilities for biotherapeutic products and native protein characterization, the Thermo Scientific™ Orbitrap™ Ascend BioPharma Tribrid™ mass spectrometer delivers ultimate experimental throughput, versatility, and ease-of-use. Achieve greater coverage using a revolutionary hardware design that features two ion routing multipoles. Characterize intact biopharmaceuticals with improved native MS workflows. Perform sensitive and deep peptide-based analyses and improve your oligonucleotide characterization with the combination of recommended alternate fragmentation techniques that can help elucidate the most difficult-to-characterize molecules. All while producing more with less instrument setup featuring automated Auto-Ready ion source for calibration and pre-built, optimized method templates to maximize convenience and ease-of-use. Be ready for tomorrow's most challenging demands.



Be ready for tomorrow's most challenging demands with the Orbitrap Ascend BioPharma 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
- Recommended Native MS mode provides expanded mass range capabilities for comprehensive MSⁿ characterization of native proteins and protein complexes
- Extended quadrupole isolation range with recommended Native MS mode 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 MSⁿ 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 MSⁿ analyses with Dynamic Maximum Injection Time
- Synchronous Precursor Selection (SPS) for MSⁿ experiments
- Recommended compact Electron Transfer Dissociation (ETD) and 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), recommended electron-transfer dissociation (ETD, including electron-transfer higher-energy collision dissociation, or EThcD, and electron-transfer collision-induced dissociation, or ETciD) and recommended ultraviolet photodissociation (UVPD), available at any stage of MSⁿ with detection in the Orbitrap mass or linear ion trap analyzers
- Recommended Proton Transfer Charge Reduction (PTCR) simplifies MSⁿ 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 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

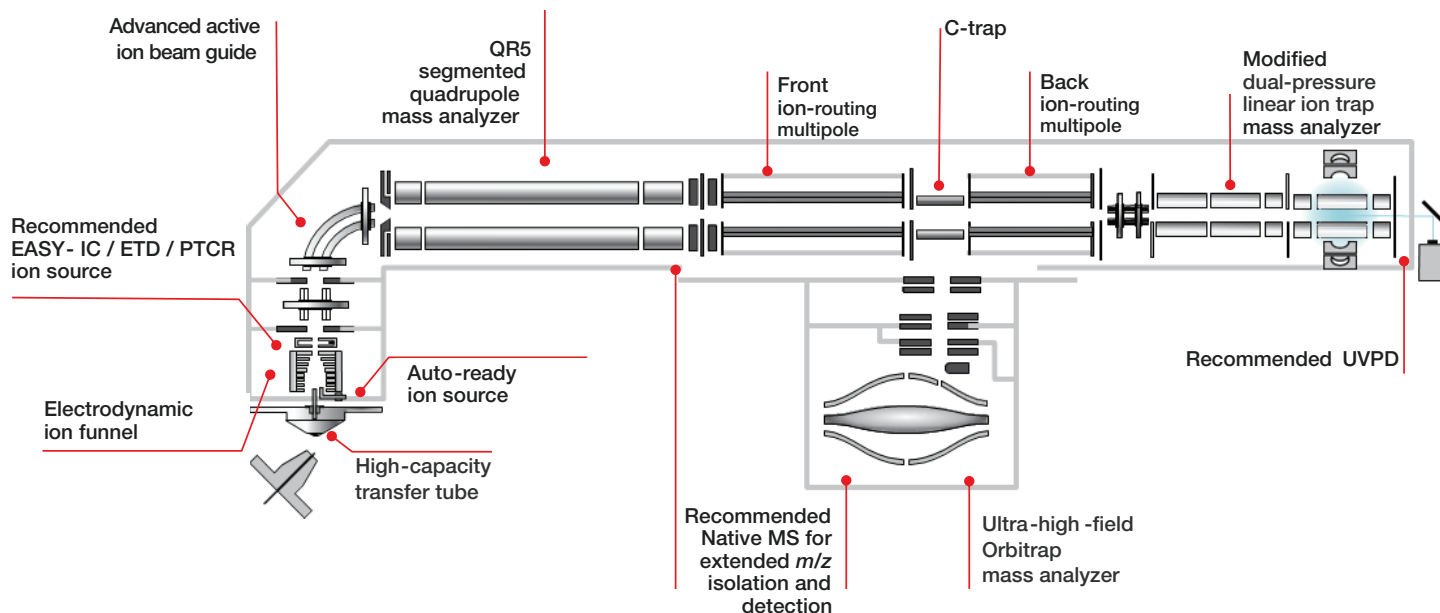


Figure 1. Orbitrap Ascend BioPharma Tribrid mass spectrometer ion path

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 ≥ 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 MS²
- 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

- MSⁿ, 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)
- Recommended ETD/ET_hcD/ET_ciD and UVPD fragmentation techniques
- Recommended PTCR ion-ion reaction capability
- 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 ~1x10⁻¹⁰ Torr
- Stainless steel and aluminum high-vacuum analyzer chambers
- Enhanced vacuum technology reduces the pressure in the ultra-high vacuum region to ~1x10⁻¹⁰ 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 (recommended)

- 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 (recommended)

- 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 (recommended)

- 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 (recommended)

- 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 MSⁿ, 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 (recommended)

- 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 ProSightPD software 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^n spectra in each cycle by intelligently scheduling MS and data-dependent MS^n scans based on a user-defined time between adjacent survey spectra.

TopN

Enables a user-defined maximum number of high-quality MS^n 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 techniques

Precursor dissociation can be performed in the ion-routing multipoles (HCD), the linear ion trap (CID, recommended ETD/ETciD, recommended UVPD) or both (ETHCD), with product ion detection in the linear ion trap or Orbitrap mass analyzer at any stage of MS^n 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 (recommended)

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

Real-time database search

Searches user-defined database in real time to then select identified/or not identified precursors for further MSⁿ experimentation. Example: Selection of identified precursors only for SPS MS³ 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 MSⁿ acquisition events. It enables library directed MSⁿ 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 (ϕ SDM) 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 recommended Native MS option 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 ⁿ 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 ⁿ 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 solution; internal calibration achieves <1 ppm RMS drift over 24 hours, measured with FlexMix solution
MS/MS Electrospray Ionization (ESI) ion trap sensitivity	2 μ L of a 50 fg/ μ L solution of reserpine (100 femtograms total) injected at a flow rate of 500 μ L/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 ⁿ , 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 ⁿ 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. Recommended Native MS option 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
Orbitrap resolution	Reaction with ETD anions dissociates the isolated 2 ⁺ MRFA precursor (<i>m/z</i> 262) to generate a series of fragments that have a summed relative abundance \geq 15% to the precursor measured in the linear ion trap, using FlexMix solution infused at 6 μ L/min
UVPD recommended 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 \geq 25% to the precursor measured in the linear ion trap, using FlexMix solution infused at 6 μ L/min
PTCR recommended option efficiency	Reaction time of 100 ms causes isolated MRFA 2 ⁺ 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 \geq 25% to the precursor measured in the linear ion trap, using FlexMix calibration solution infused at 6 μ L/min

* Under defined conditions

Advanced data-dependent experiments

TMT SPS MS³ aided by Real-Time Search

TMT SPS MS³ experiment with real-time protein database search to perform SPS MS³ only on identified precursors.

Universal method

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

Product-ion triggered-MSⁿ

Fragment ion or neutral loss-triggered MSⁿ experiment, including at the same MSⁿ level.

Isolation offset

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

Quanfirmation 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 pump 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 recommended

- Nitrogen supply for EASY-IC/ETD ion source: 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/OrbitrapAscendBioPharma