EXCELLIMS

Innovation for Excellence in Detection Technology

MA3100: High Resolution Ion Mobility Analysis for Thermo Scientific™ Linear Ion Trap and Orbitrap Mass Spectrometers

Ion mobility spectrometry (IMS) separates molecules by shape and size through their interaction with a drift gas. Coupled with mass spectrometry (MS), IMS provides an additional dimension of separation for the differentiation of isomers, 2D-confirmation of compounds, detailed molecular characterization, and fingerprinting of complex mixtures. With the MA3100, Excellims has developed a high resolution, compact IMS device that can be easily added to mass spectrometers like Thermo Fisher Scientific™ Orbitraps and linear ion traps (**Fig. 1**).

Designed for Easy Integration

In recent years, IMS-MS analysis has gained strong popularity as shown by a steadily growing number of publications. IMS-MS systems are often custom-built, requiring a significant time-investment on the part of the researcher. Fully integrated commercial IMS-MS systems have become available but are typically large and may be cost-prohibitive. With its high-performance atmospheric drift tube and modular design, the MA3100 offers a straightforward and cost-effective alternative for researchers who want to add IMS capability without changing mass spectrometer. The MA3100 is designed to replace the Ion Max and compatible ion sources in as little as 30 seconds, without modifications to the instrument or a need to break vacuum, thus providing IMS capabilities whenever needed.

Ions are generated using the Excellims Directspray™ electrospray ionization (ESI) source or a Thermal Desorber (using corona discharge ionization) and are introduced into the MA3100's drift tube via a pulsed Bradbury-Nielsen type ion gate (**Fig. 2**). Acceleration in the drift tube's field is counteracted by collisions with the drift gas, typically air, N_2 or CO_2. The resulting constant travelling speed of the ions depends on their collisional cross section (CCS) and thus on their shape and size.

While only 10.5 cm long, the high performance drift tube in the MA3100 offers excellent IMS resolution (resolving power > 70) due to its design and the high number of collisions at atmospheric pressure. The drift time spectrum can be visualized using the MA3100's Faraday detector. As drift times are directly related to the molecule's CCS and thus to their shape and size, drift tube IMS-spectra are easy to interpret (larger molecules – longer drift times), unlike in DMS/FAIMS-based mobility devices.

A second (exit) gate is located directly behind the Faraday detector, allowing the user to selectively introduce one or multiple species into the mass spectrometer based on their drift time and to run combined IMS-MS experiments, as will be discussed in the next section. All necessary control electronics and software are provided as part of the MA3100 package.

Fig. 1 MA3100 IMS analyzer and control electronics shown on a Q Exactive™ Orbitrap MS.

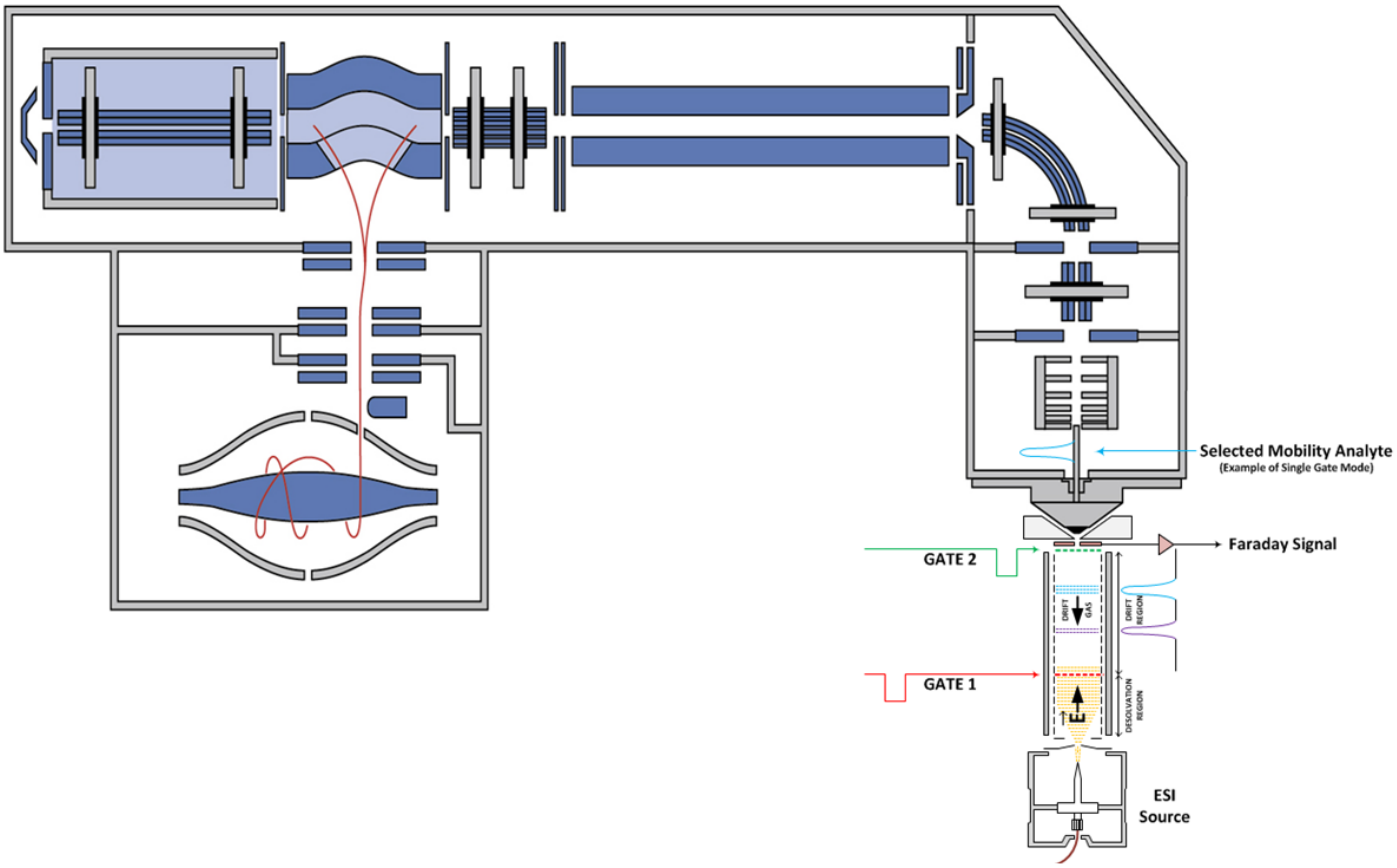

Fig. 2 The Excellims dual-gate MA3100 is easily coupled with Thermo Fisher Scientific™ mass spectrometers utilizing the Ion Max source design. Four different control modes support simultaneous collection of IMS and MS spectra and selective inclusion or removal of targeted ions based on their drift time.

Multiple Modes of Operation

A dynamic array of methodologies can be developed from four available operating modes:

- *OPEN mode*: Ions flow continuously through both ion gates into the mass spectrometer without IMS separation. Conventional mass spectral data can thus still be obtained without physical removal of the MA3100.
- *FARADAY mode*: Parallel ion mobility and mass spectra are generated. Mobility information obtained from the Faraday plate detector can be used to rapidly change gate settings for targeted IMS-MS analysis (currently only supported within the visIon Trap software).
- *SCAN mode*: A window of variable width is sequentially stepped across a chosen drift time range. This mode creates 2D IMS-MS maps and also allows for individual m/z ion mobility profiles to be extracted.
- *GATED mode*: Single or multiple mobility window(s) can be specified for targeted ion transmission or removal (**Fig. 3**), e.g. to quickly confirm the presence of a particular isomer or to selectively remove a species from ion accumulation in the trap.

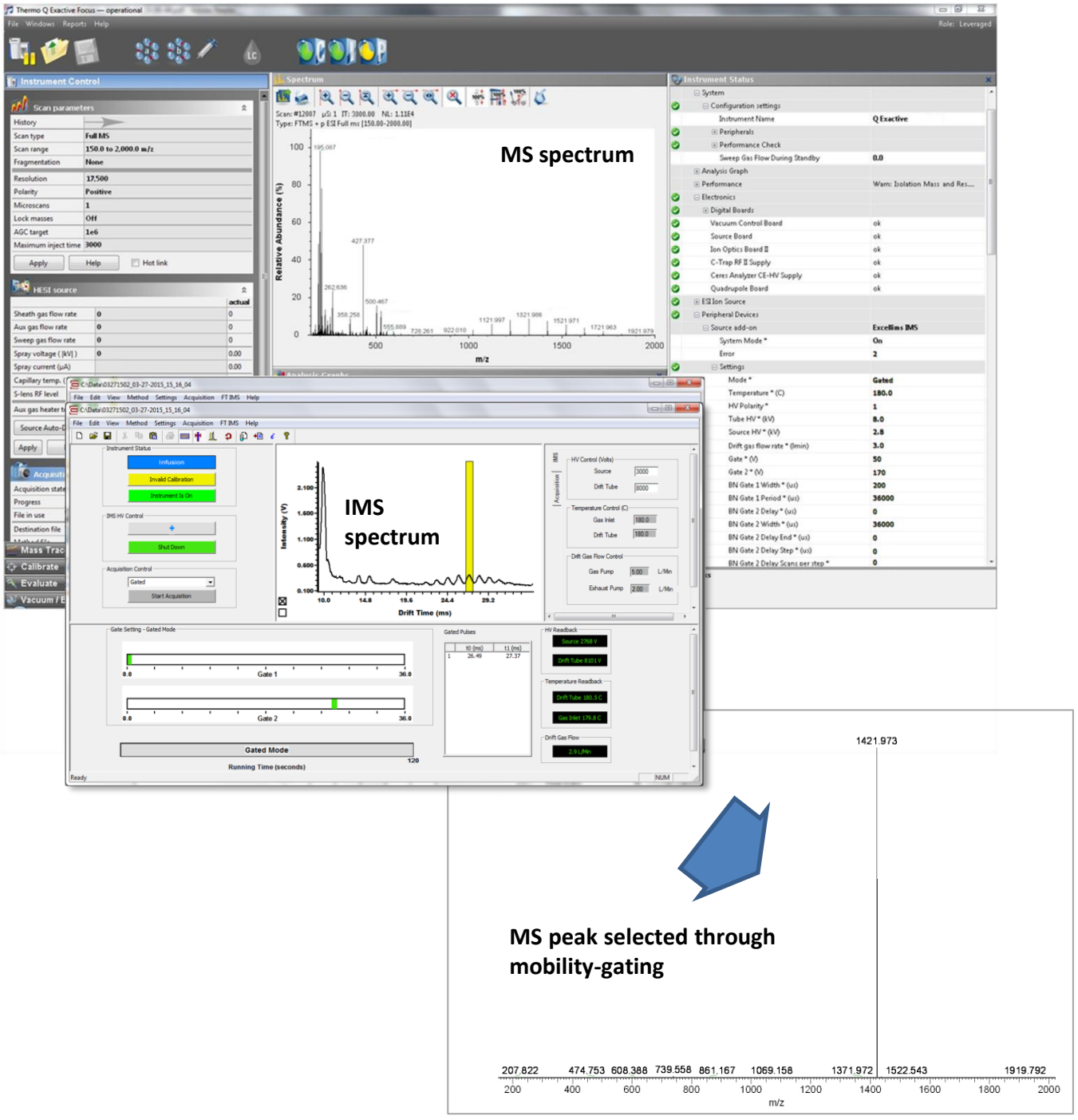

Fig. 3 Illustration of the MA3100 operating modes: the Tune window (top) shows the entire MS spectrum in OPEN mode while the visIon window (middle) displays the corresponding IMS spectrum in FARADAY mode. The yellow marker indicates the selection of one IMS peak selected for further analysis in GATED mode. The GATED mode MS spectrum (bottom) confirms that all but the selected IMS peak have been filtered out successfully.

Isomer differentiation and 2D confirmation

A principal benefit of IMS is its ability to easily distinguish between isomers that are hard to separate by MS/MS or by chromatography. Fig. 4–6 show several examples in IMS-only or extracted IMS-MS scan mode.

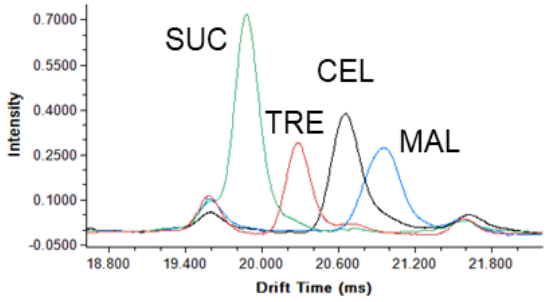

Fig. 4 Disaccharide isomers (sucrose, trehalose, cellobiose and maltose)

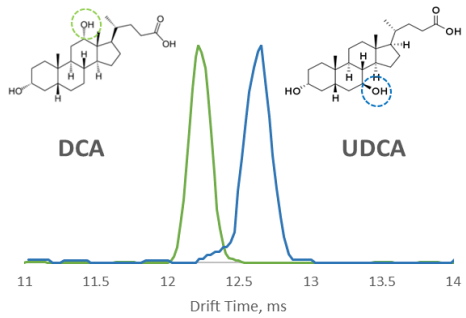

Fig. 5 Bile acids isomers on the MA3100 + Orbitrap

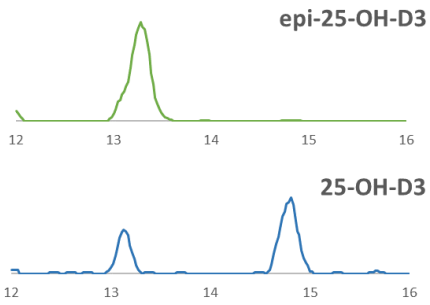

Fig. 6 Differentiation of Hydroxy-Vitamin D3 from its epimer (using a diagnostic secondary conformer)

The same capability to differentiate conformers and other isomers can also be applied to larger molecules, as shown in Fig. 7 and 8.

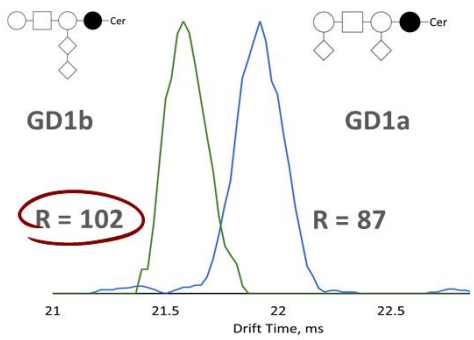

Fig. 7 Separation of two gangliosides by IMS on the MA3100 with resolving power 80 – 100

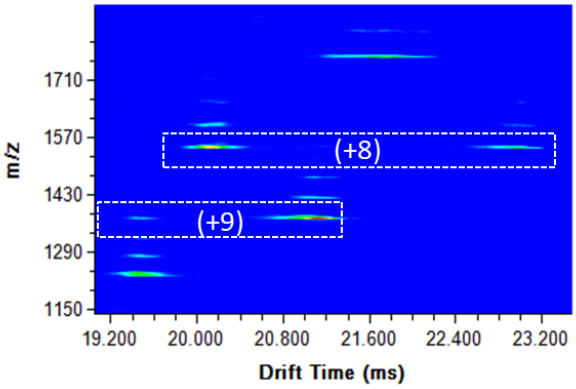

Fig. 8 Two charge states of ubiquitin (+8 and +9) showing multiple conformers (2D IMS-MS map on Orbitrap)

The MA3100 IMS-add on can thus be used to screen for the presence of isomers e.g. for new compounds in pharmaceutic development or for complex mixtures in petrochemical and polymer applications. Conformation states can be studied quickly under a variety of conditions. If multiple isomers with known drift times are expected, a quick succession of IMS filters can be set to isolate each isomer without chromatography or MS^2. The same IMS filter approach can be used for general 2D confirmation of compounds, using drift time and m/z as separate variables, as long as these have been first characterized by standards.

IMS-MS fingerprints and CCS measurements

IMS-MS scans with the MA3100 can also be used to develop a 2D map of complex samples, dividing MS spectra into multiple trendlines representing series of molecules with different degrees of compactness or different charge states (see Fig. 9 for an example).

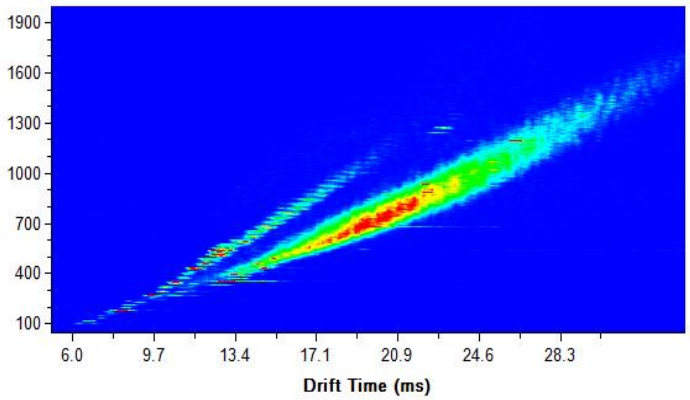

Fig. 9 Trendlines in a crude oil intermediate

2D IMS-MS mapping can thus be used to create a quick visual fingerprint of the sample, to reduce complexity by making distinct series visible and comparable (e.g. polymer branching) or to bring out hidden lower intensity series and outliers in a dense forest of MS peaks.

Due to the direct correlation between drift time and CCS in linear drift tube IMS, size differences between any two molecules can be directly visualized (example in Fig. 10)

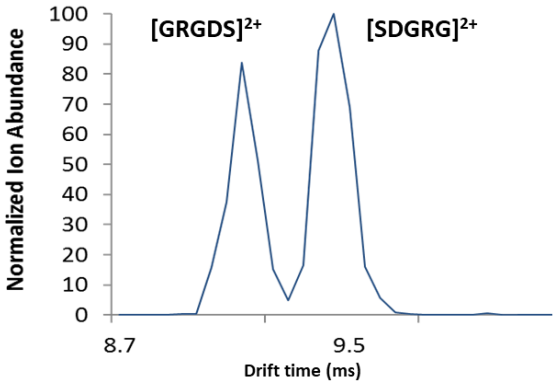

Fig. 10 SCAN-mode spectrum of a pair of reverse peptides

Using a reference compound with known CCS, drift times can then be converted to CCS without a need for substance-class specific calibrations.

Software integration and data processing

For the Exactive™ series (Orbitrap Q Exactive and higher), control of the MA3100 is embedded within the latest releases of Tune with the appropriate license. For other instruments, visIon Trap is supplied directly through Excellims for complete control of the MA3100 and all necessary communications with the mass spectrometer. visIon Trap automatically reads in the raw MS files after the acquisition and displays 2D IM-MS maps as well as extracted MS and IMS profiles.

The MA3100 thus offers a complete package of ionization source, high performance IMS separation with multiple modes for 2D analysis, control electronics and software, ready to provide ion mobility capabilities to your mass spectrometer as needed. Additional options including enhanced separation through liquid drift gas modifiers, nanospray adaptors and multiple other source options – please contact an Excellims application specialist at sales@excellims.com to discuss your requirements in detail or to schedule an MA3100 demonstration.

Application examples in the literature

- Kaszycki et al., *Separation of biologically relevant isomers on an Orbitrap mass spectrometer using high-resolution drift tube ion mobility and varied drift gas mixtures*, Rapid Commun. Mass Spectrom. 2019; 1– 8
- Oranzi et al., *Measuring the Integrity of Gas-Phase Conformers of Sodiated 25-Hydroxy-vitamin D3 by Drift Tube, Traveling Wave, Trapped, and High-Field Asymmetric Ion Mobility*., Anal. Chem. 2019 Mar 19; 91(6) : 4092-4099
- Kwantwi-Barima et al., *Tuning Mobility Separation Factors of Chemical Warfare Agent Degradation Products via Selective Ion-Neutral Clustering*, Anal. Chem. 2017, 89, 22, 12416-12424