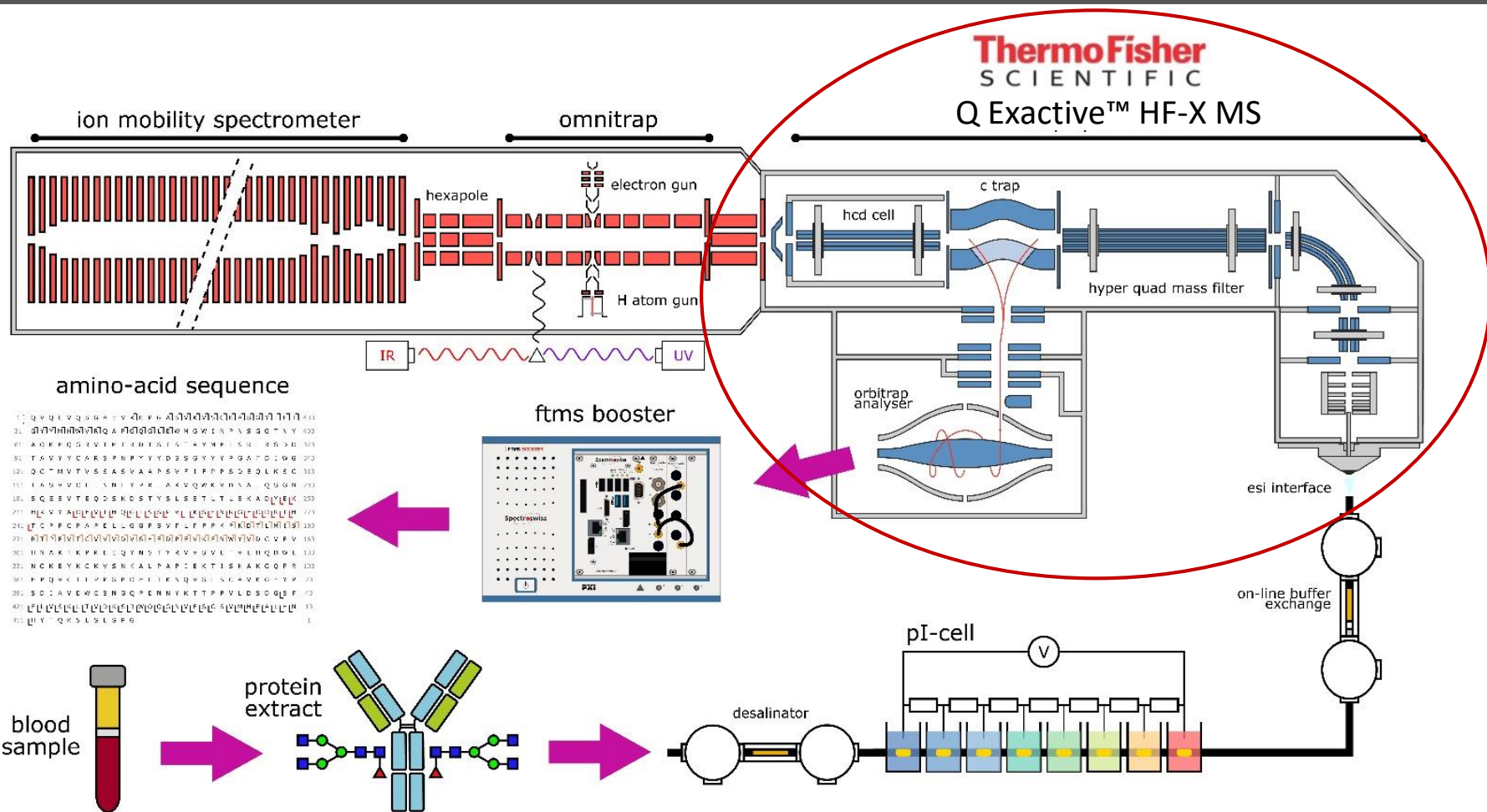




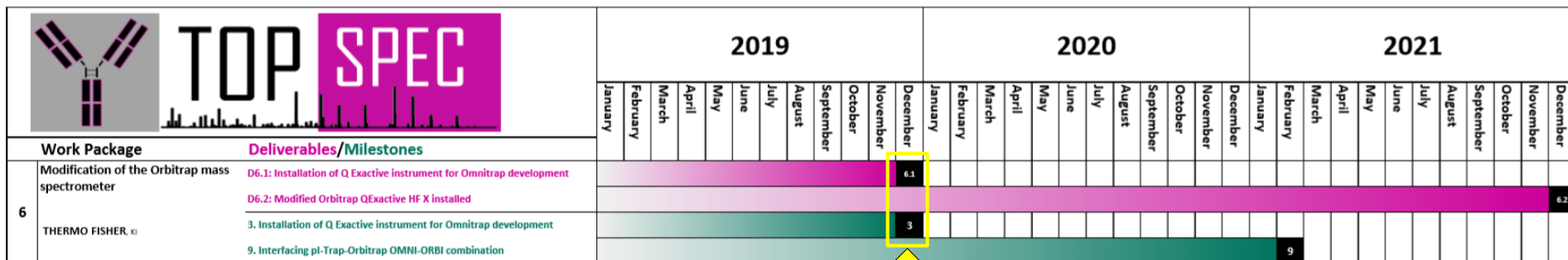
## **WP6. Modification of the Orbitrap mass spectrometer**

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# WP6 Modification of the Orbitrap mass spectrometer



| Deliverable | Deliverable Title  | Status    | Completion |
|-------------|--|-----------|------------|
| D6.1        | Installation of Q Exactive instrument for Orbitrap development | Completed | 20-12-2019 |
| D6.2        | Modified Orbitrap QExactive HF X installed                     | Ongoing   | 31-12-2021 |
| Milestone   | Deliverable Title  | Status    | Completion |
| 3           | Installation of Q Exactive instrument for Orbitrap development | Completed | 20-12-2019 |
| 9           | Interfacing pl-Trap-Orbitrap OMNI-ORBI combination             | Ongoing   | 28-02-2021 |

## Task 6.1: Description

- Installation of a **loaned Q Exactive instrument** to Fasmatech to support Omnitrap development.
- Configured to be **interfaced to Omnitrap** by removing the charge detector on the back of the HCD cell and adding functionality of ion transfer to and from the Omnitrap
- Dedicated trigger signals will be provided to initiate the operational sequence of the Omnitrap
- Software training and support will be provided to Fasmatech and Spectroswiss
- The instrument will be focused on optimizing Omnitrap functionalities



## Task 6.1: Action

- Difficulties with Source turbopump were overcome by improved instrument ventilation
- Instrument passed standard acceptance specifications, making it available for Omnitrap installation
- Needed modification: Extension of the mass range of Orbitrap detection towards  $m/z > 10,000$  to cover intact native proteins, especially intact antibodies sprayed under native conditions
- Joint experiments on the combined hybrid instrument took place in January 2020 at Fasmatech site



## Task 6.2: Description

- An **Orbitrap Q Exactive HF-X (or a similar high-end instrument)** will be modified to improve its performance for desolvation and transmission of intact antibodies
- There is a reserve for optimizing the desolvation region of the atmosphere-to-vacuum interface that deserves a more detailed exploration
- **Integration of instrument control software** using application programming interface (API). Development of tuning and calibration procedures specific for antibody analysis in order to ensure best top-down performance, integration of data for all fragmentation methods and cross-section measurements. (Joint work with Spectroswiss and Fasmatech)
- After testing of all functional units, the resulting instrument will be delivered and installed at KI and performance protocol will be completed for a test set of compounds.

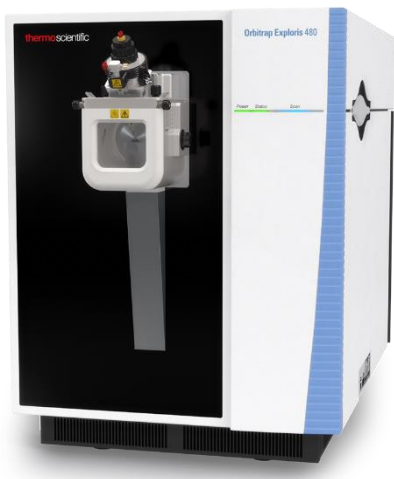
Q Exactive™ HF-X MS



Orbitrap Exploris™ 480 MS



- Since the **new Orbitrap Exploris 480 supersede the Q Exactive HF-X** instrument it was proposed to also base the TopSpec platform on this latest product
- Most functionalities are transferrable from Q Exactive HF to Orbitrap Exploris platform
- Some increase of project scope will be addressed by joint efforts with Fasmatech



### Orbitrap Exploris 480 MS

#### Improved features:

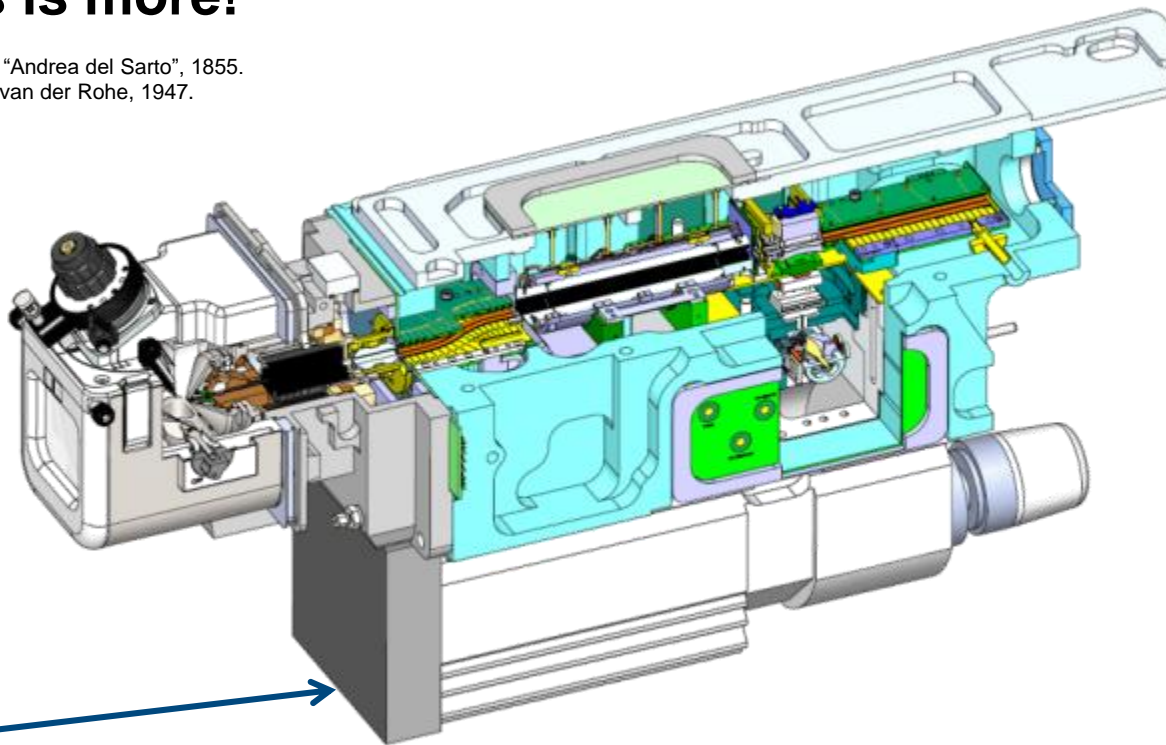
- Increased robustness, higher resolution and easier access for service and Omnitrap upgrade
- Field-asymmetric waveform ion mobility (FAIMS) front-end option, which opens new capabilities for additional specificity of analysis

# Overview of Orbitrap Exploris 480 Mass Spectrometer



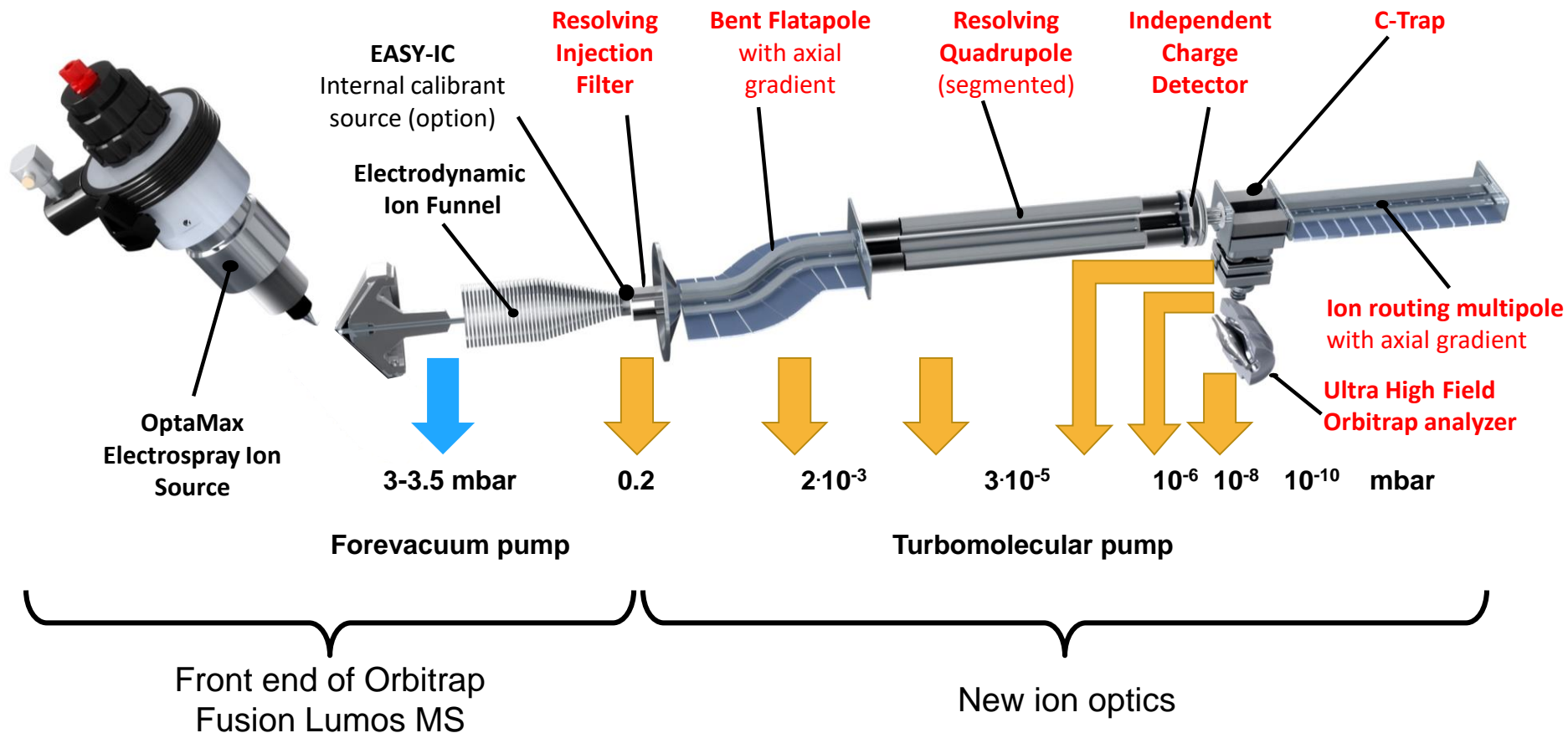
## Less is more!

R. Browning, "Andrea del Sarto", 1855.  
L. Mies van der Rohe, 1947.



>2-fold size reduction relatively to Q Exactive family

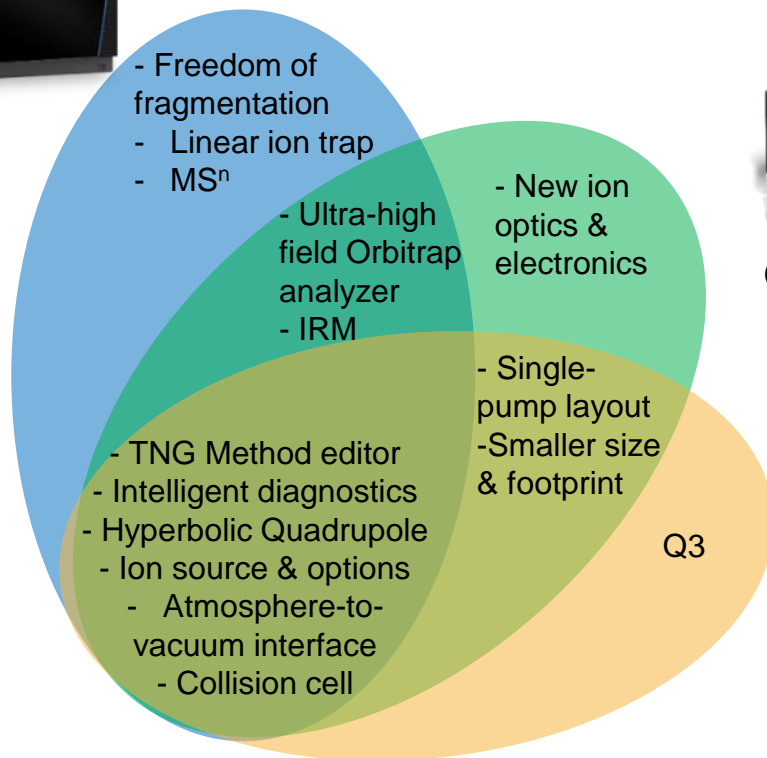
# Layout of Orbitrap Exploris 480 Instrument



# How This Instrument Fits with Others?



Orbitrap Eclipse Tribrid MS

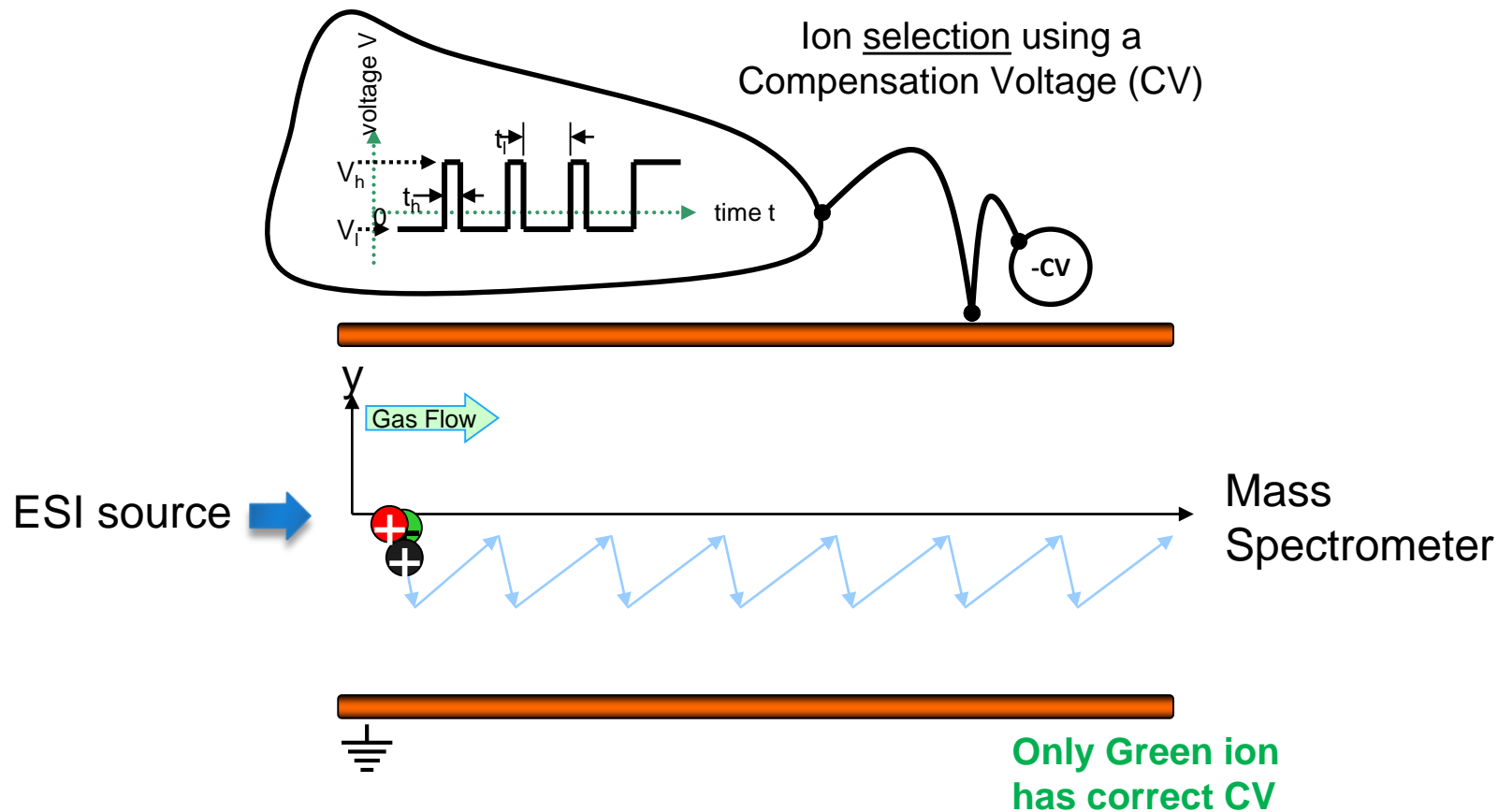


Orbitrap Exploris 480 HR/AM MS



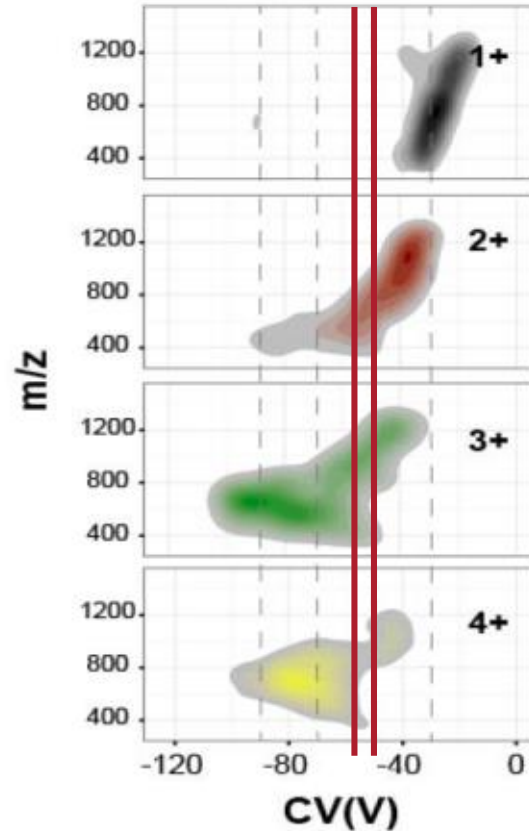
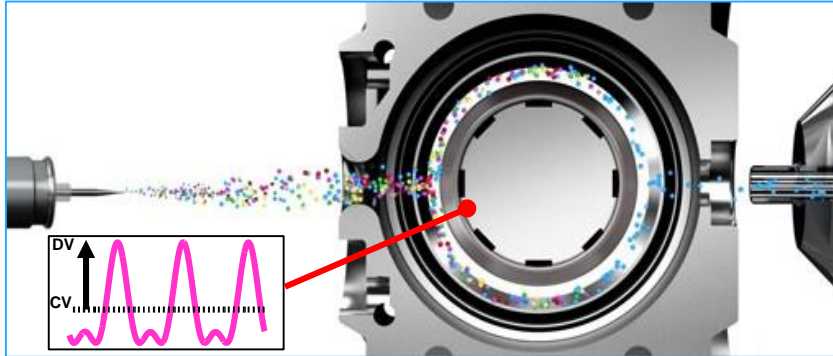
TSQ Altis Triple Quad MS

# High-Field Asymmetric Waveform Ion Mobility Spectrometry (FAIMS)



# What is Special about FAIMS with Cylindrical Electrodes?

- Cylindrical Electrodes help focus ions through the electrode assembly
- Nitrogen carrier gas moves the ions through from front to back
- The result is better ion transmission into the MS compared to parallel, planar electrodes
- The inner electrode blocks “line of sight”, but the gas and fields direct ions to the MS inlet



Pfammatter, S. *et.al.* *Mol Cell Proteomics*. 2018.

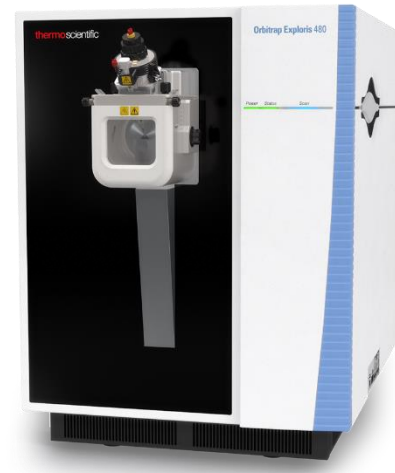
## Task 6.2: Open items

### Improved features:

- Increased robustness, higher resolution and easier access for service and Omnitrap upgrade
- Field-asymmetric waveform ion mobility (FAIMS) front-end option, which opens new capabilities for additional specificity of analysis

### Open questions:

- New mechanical interface (already designed by Fasmatech)
- Transferability of IAPI from Q Exactive platform to Exploris platform
- Interfacing of Omnitrap to Ion Routing Multipole/HCD cell – mass range, speed
- FAIMS performance for proteins- limited data available



Orbitrap Exploris 480 MS