

# Lipid Separation and Structural Characterization Using Hybrid Surface Technology and Travelling Wave Cyclic Ion Mobility

Waters

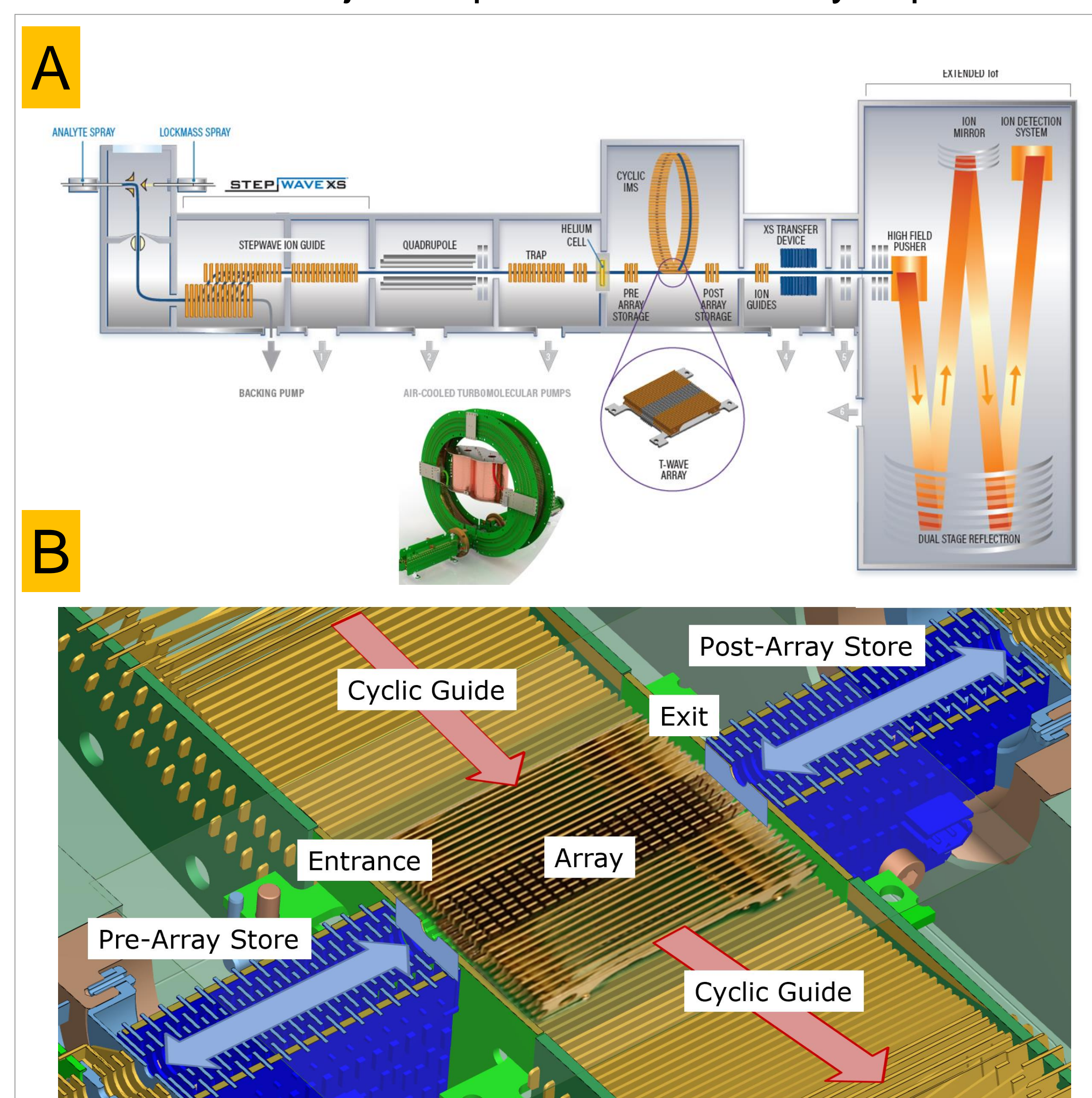
THE SCIENCE OF WHAT'S POSSIBLE.™

Giorgis Isaac, Nyasha Munjoma, Martin Palmer, Lee A. Gethings, Robert S. Plumb

<sup>1</sup> Waters Corporation

## INTRODUCTION

- The analysis and structural characterization of lipids remain challenging due to the chemical structure diversity and isobaric nature of lipids.
- Ion mobility spectrometry (IMS) applicability has been widely used for the characterization of biomolecules and small molecules such as lipids.
- The main objective of the presentation is to provide an overview of cyclic ion mobility (cIM) separation technology to enhance separation and structural characterization of lipids.
- The cIM device consists of a 100 cm path length RF ion guide comprising over 600 electrodes around which T-Waves circulate to provide mobility separation (Figure 1A). It provides the option to perform either a single pass, or multiple passes until the desired resolution is achieved.
- Figure 1B shows a closer look at the cIM device with the option to “slice” and store ions in the Pre-Array store and re-inject to perform ion mobility separation.



## RESULTS

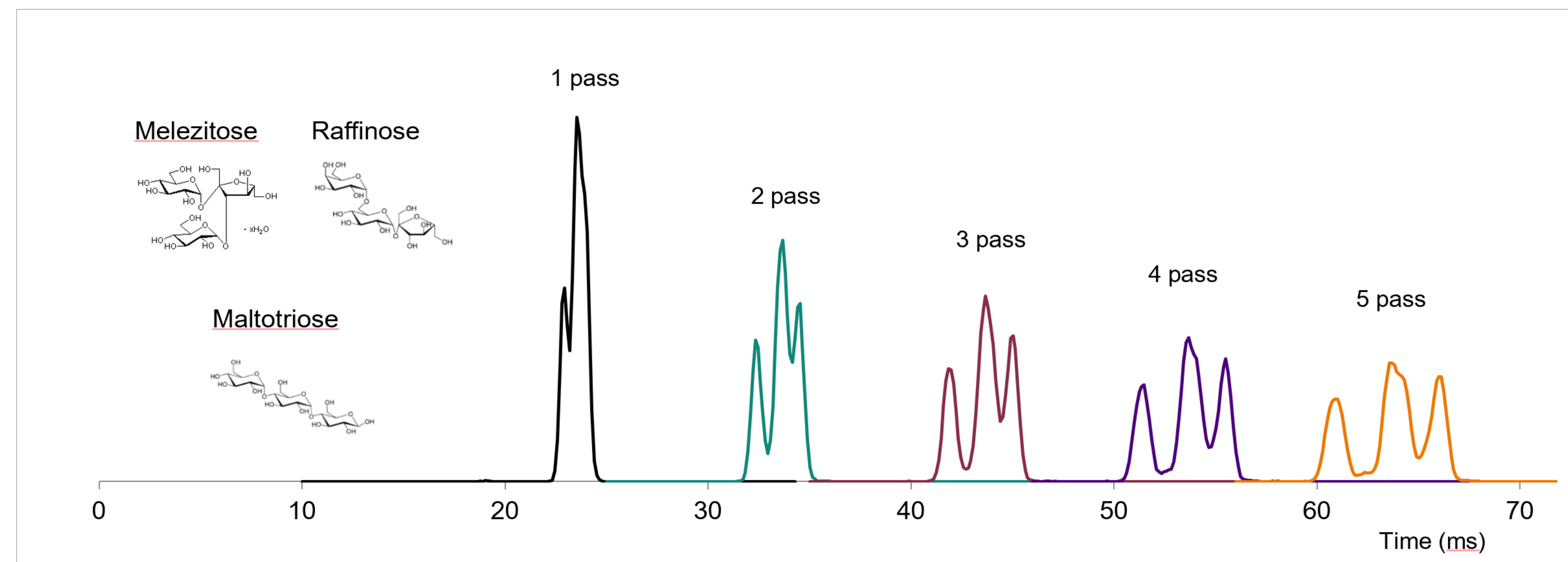


Figure 2. cIMS separation of trisaccharides. More features visible at subsequent passes. CCS Melezitose < Raffinose < Maltotriose.

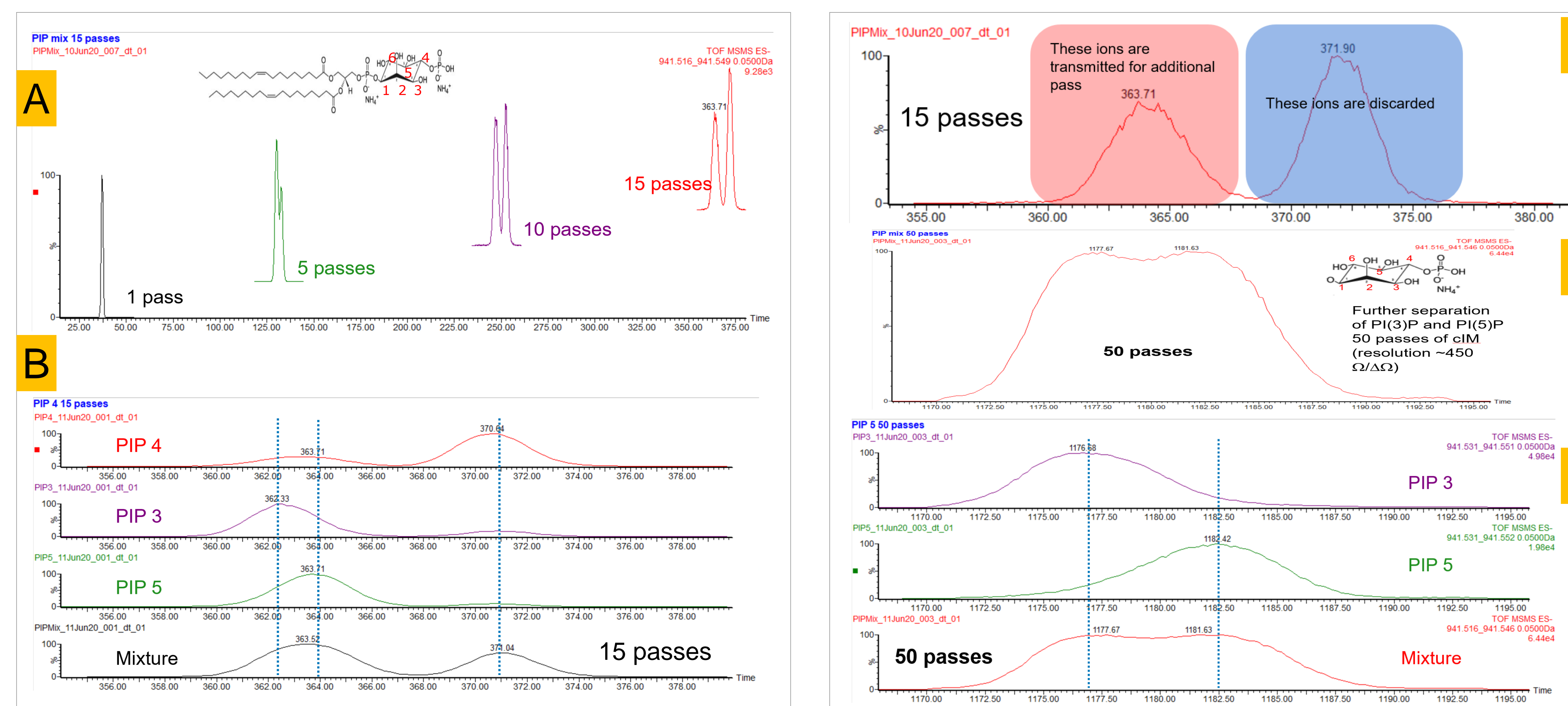


Figure 3. (A) Separation of PI(3)P, PI(4)P and PI(5)P mixture with 1 to 15 passes (IMD resolution ~65 to 250  $\Omega/\Delta\Omega$ ). The mixture contains three PIP isomers and expected to see three peaks but only two isomers separated at 15 passes. (B) To identify which PIP peaks are co-eluting, individual PIP species of PI(3)P, PI(4)P and PI(5)P standards were infused. PI(3)P in purple and PI(5)P in green didn't separate from each other. Only PI(4)P in red resolved from the other two PIPs at 15 passes. (C) In order to separate PI(3)P and PI(5)P; the PI(4)P ions (highlighted in blue) were discarded, and the co-eluting PI(3)P & PI(4)P ions (highlighted in red) are sliced, stored in the Pre-Array store and transmitted to the cyclic ion mobility for additional passes.

## RESULTS

(D) After 50 passes (approximately at 450 IMS resolution) PI(3)P and PI(5)P were successfully separated. (E) To identify the PI(3)P and PI(5)P in the mixture, individual standards of each species were infused into cIM at 50 passes. PI(3)P in purple at a drift time of 1176 milli sec and PI(5)P in green eluted at a drift time of 1176 and 1182 milli sec, respectively.

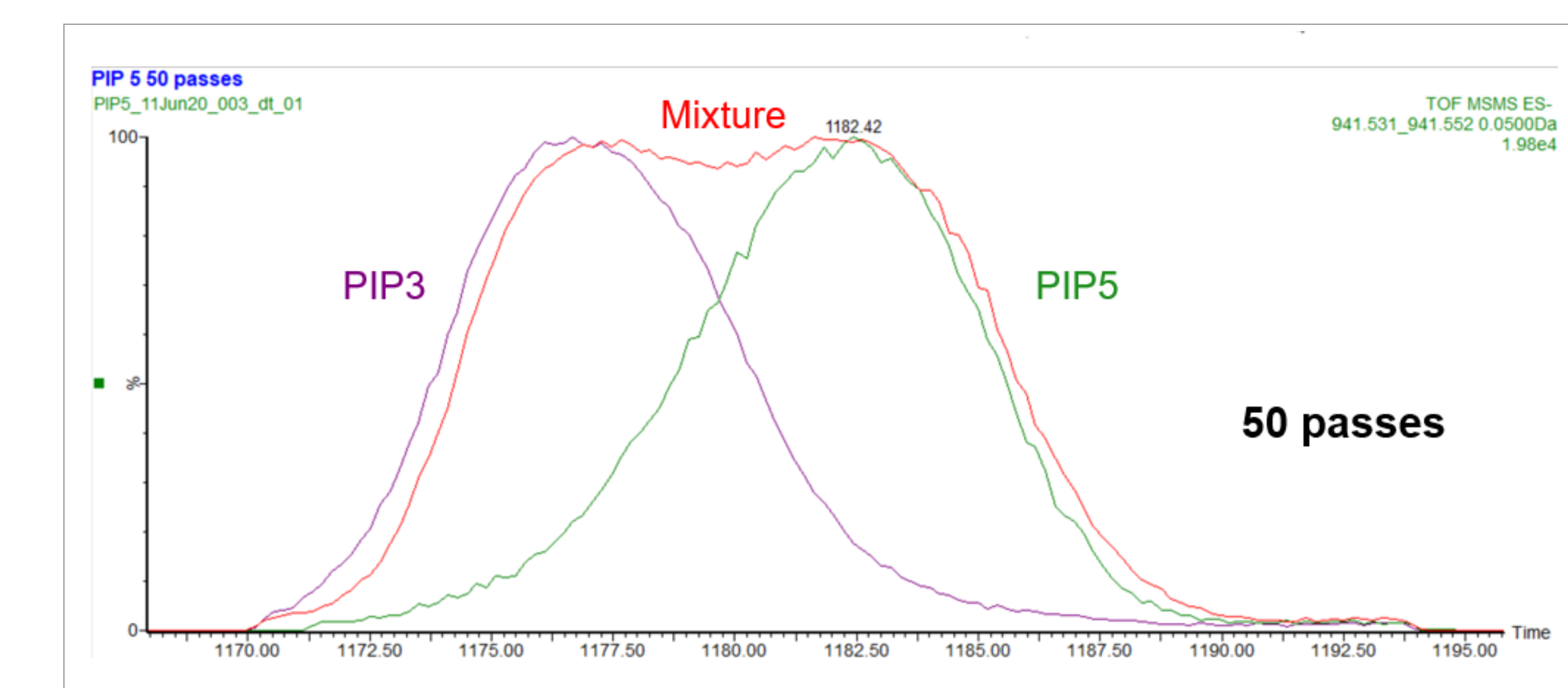


Figure 4. Overlaid mobilogram of individual PI(3)P (purple), PI(4)P (green) and their mixtures (red) after 50 passes.

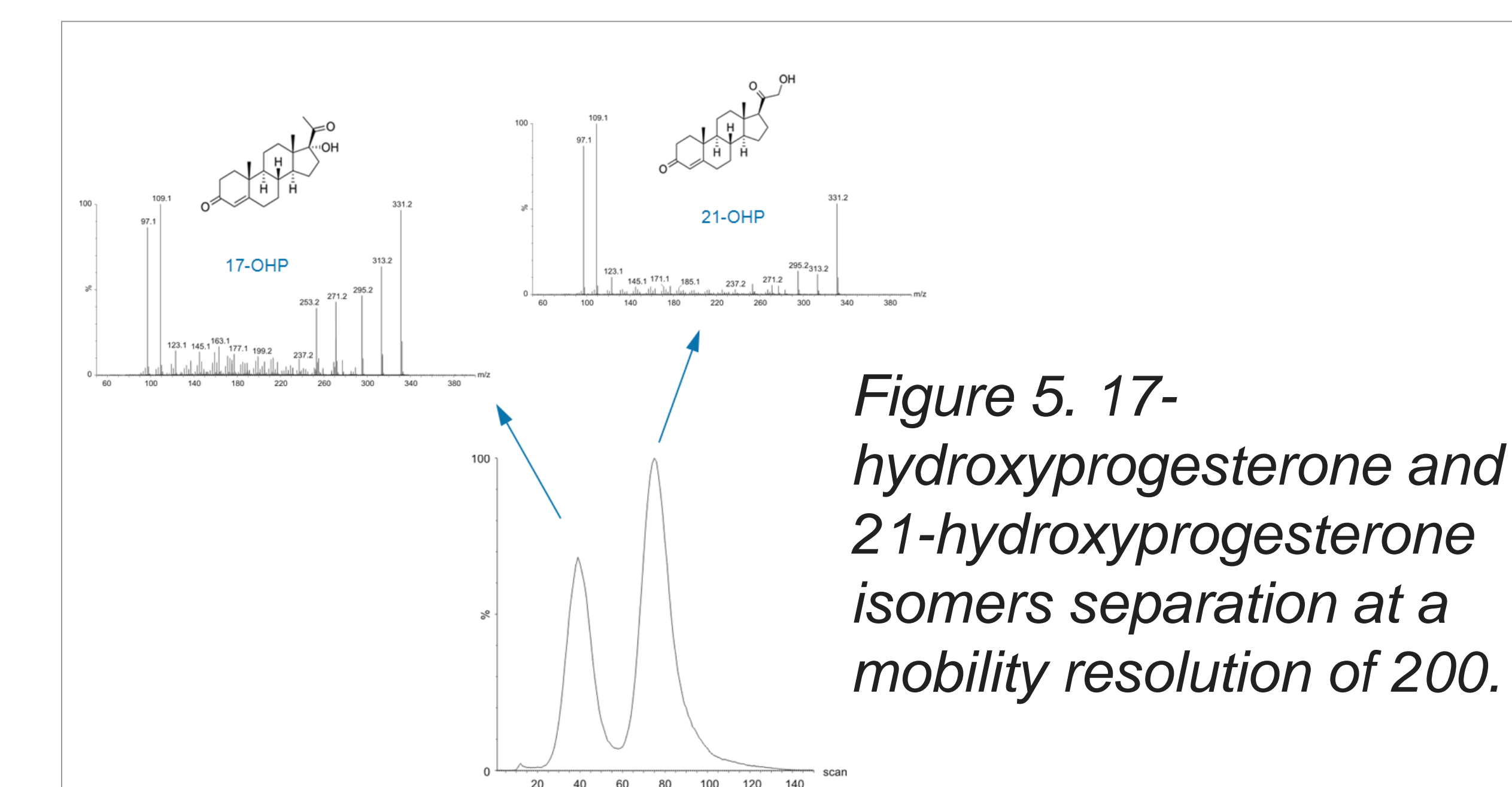


Figure 5. 17-hydroxyprogesterone and 21-hydroxyprogesterone isomers separation at a mobility resolution of 200.

## Conclusion

- Cyclic IMS provides novel, scalable ion mobility resolution (50 passes of cIM with resolution ~450  $\Omega/\Delta\Omega$ ).
- The increased resolution is useful to resolve and separate isobaric and isomeric lipids species.
- Advanced modes of operation with ion activation followed by ion mobility separation offers new insights into lipid structural characterization.

Figure 1. (A) Schematic of the cyclic IMS QTOF instrumentation. It contains three main regions: The trap region, the cyclic ion mobility device and the transfer region. (B) Zoom in of the cyclic ion mobility device.

To download a copy of this poster, Visit [www.waters.com/posters](http://www.waters.com/posters)