Evaluation of Ion Mobility Mass Spectrometry (IMMS) for Determining the Isomeric Heterogeneity of Bovine Submaxillary Mucin (BSM)

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Abstract

- Rapid separation and analysis of isomeric species are essential steps in the analytical protocol of carbohydrate structure analysis for glycomics research.
- This study evaluated the use of ion mobility spectrometry (IMS) prior to MS as an additional rapid isomer separation step prior to mass spectral analysis.
- Neutral oligosaccharide alditols of BSM were investigated in positive mode.
- Multiple isomers were detected for specific m/z on millisecond time scale, which would be difficult or impossible using MS alone or single LC column.
- Tandem MS provide valuable information for elucidating glycan structures and understanding the isomeric nature of glycans.

Experimental

Figure 2: Schematic diagram of resistive glass tube on mobility spectrometer interfaced to a time-of-flight mass spectrometer.

Ion mobility quadrupole ion trap mass spectrometer was also employed in this study.

Results –Part I

Figure 1: HPLC elution profile of neutral oligosaccharide-aldehydes isolated from BSM and mass spectra of BSM mixture and individual HPLC fractions.

Results –Part II

Figure 3: Two-Dimensional IM-MS plots of different compounds in BSM fractions:
- (a): m/z 449 in BSM Fr. 2; (b): m/z 757 in BSM Fr. 7; (c): m/z 814 in BSM Fr. 7;
- (d): m/z 919 in BSM Fr. 6 & 9; (e): m/z 960 in BSM Fr. 8 & 9; (f): m/z 1065 in BSM Fr. 9.

Results –Part III

Figure 4: Ion mobility and MS/MS analysis of m/z 611 from HPLC fraction 4, 5 and 6.
- (a): Overlaid 2D IMMS spectra of m/z 611 from BSM Fr. 6 (black trace) and Fr. 5 (red trace); (b): Overlaid 2D IMMS spectra of m/z 611 from BSM Fr. 5 (red trace) and Fr. 6 (black trace); (c): The MS/MS spectrum of m/z 611 eluted from HPLC fraction 4; (d): The MS/MS spectrum of m/z 611 eluted from HPLC fraction 5; (e): The MS/MS spectrum of m/z 611 eluted from HPLC fraction 6.

Conclusions

- It is demonstrated that IMS has the capability to rapidly differentiate glycan isomers, ranging from disaccharides (m/z 449) to hexasaccharides (m/z 1065).
- IMS resolved isomeric peaks for the m/z species that appeared as single peak in a HPLC fraction.
- The number of isomers of a given m/z species able to be identified by LC/MS proved to be very limited compared to IMMS.
- Ion mobility coupled to mass spectrometry represents a significant advancement in biological glycan analysis.

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