Differentiation of Oligosaccharide Linkage Isomers by Electron Activated Dissociation

Xiang Yu, Cheng Lin, Catherine E. Costello

Center for Biomedical Mass Spectrometry, Boston University School of Medicine, Boston, MA USA

Overview

Objective

- To investigate the potential of Electron Activated Dissociation (ExD) techniques in the differentiation of oligosaccharide linkage isomers with different charge carriers.

Methods

- ExD on a 12-T FT-ICR Mass Spectrometer.

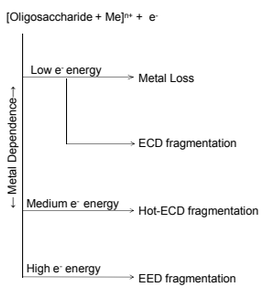
Results

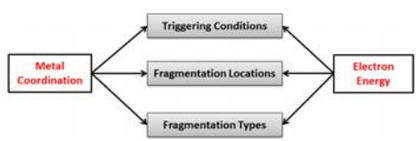
- Electronic excitation dissociation (EED) produced linkage-specific cleavages for each linkage isomer tested in this study.

Introduction

- The Metal charge carrier is one of the experimental variables that influence the electron capture dissociation (ECD) behavior of native oligosaccharides [1].

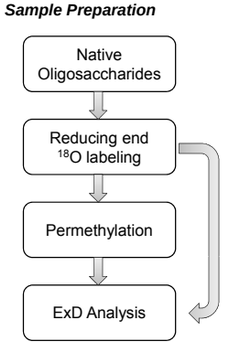
- Electron activated dissociation techniques (ExD), including ECD, hot ECD (hECD) [2] and electronic excitation dissociation (EED), produce different fragmentation patterns at different electron energies.





Methods

Sample Preparation



Data Analysis

- Data Analyses were carried out with the assistance of a Visual Basic program that has been modified from Glyco-Peakfinder.

- Mass accuracy was better than 5 ppm.

- The nomenclature used for describing glycan fragment ions was the one introduced by Domon and Costello.

Results: Differentiation of Disaccharide Linkage Isomers and Anomers by EED

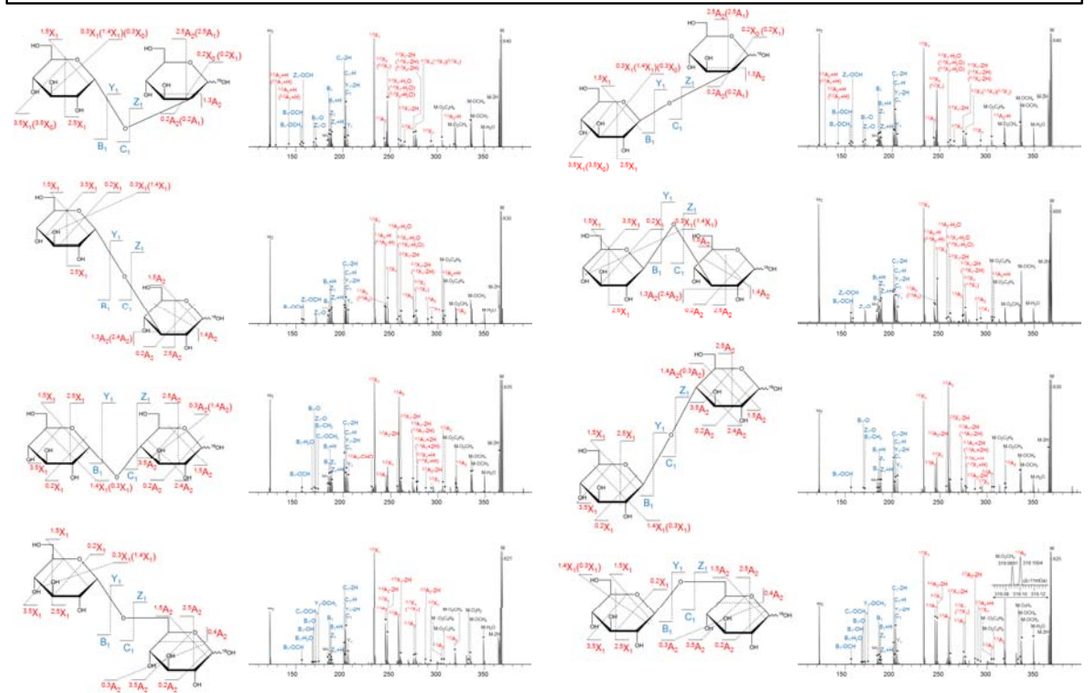


Figure 1. EED spectra and cleavage maps for the singly sodiated disaccharide linkage isomers (columns) and anomers (rows).

Results: Comparison of ECD and EED in the linkage determination of LNT and LNnT

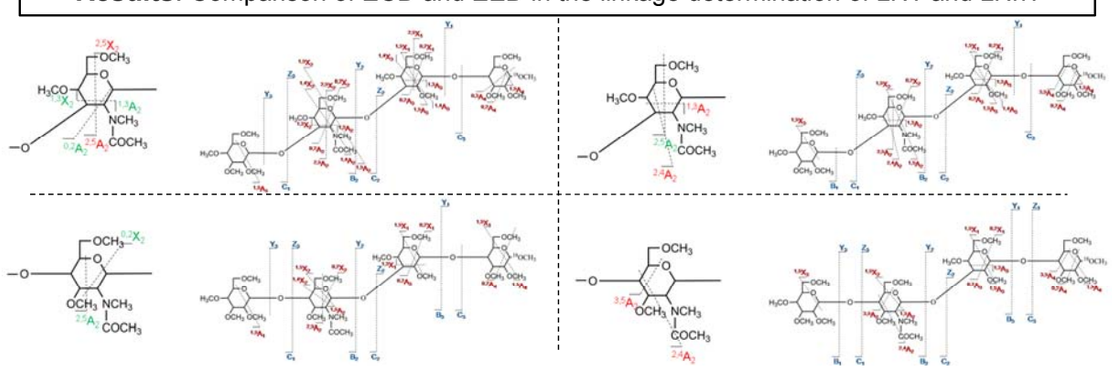
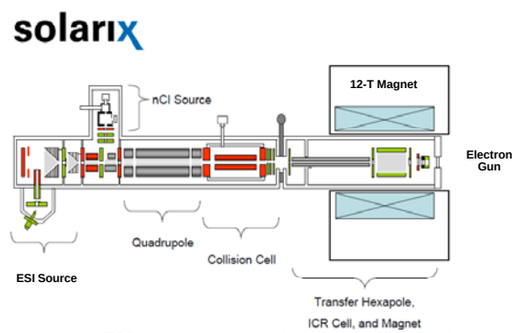


Figure 2. Cleavage patterns of the permethylated and doubly lithiated LNT (top) and LNnT (bottom) in ECD (left) and EED (right).

Methods (Continued)

FT-ICR MS Instrumentation



- For low-energy ECD analysis, precursor ions were irradiated with ~1.5 eV electrons for 100 ms.
- For higher energy EED experiments, precursor ions were irradiated with ~14 eV electrons for up to 1 s.
- The cathode heating current was 1.3 A for EED and 1.5 A for ECD.

Conclusions

- EED is capable of determining the linkage in all disaccharide linkage isomers tested here by producing signature cross-ring fragments.
- EED is applicable towards analysis of singly-charged precursor ions.
- EED is insensitive to the anomeric configuration.
- Compared to CID and ECD, EED is more informative for the linkage analysis of the model milk oligosaccharides, LNT and LNnT.
- Theoretical investigation of the EED mechanism is ongoing.

References

1, Adamson, J. T. and K. Hakansson (2007). Electron capture dissociation of oligosaccharides ionized with alkali, alkaline earth, and transition metals. *Analytical Chemistry* **79**, *7*, 2901-2910.

2, Zhao, C., B. Xie, C. E. Costello, and P. B. O'Connor (2008). Collisionally activated dissociation and electron capture dissociation provide complementary structural information for branched permethylated oligosaccharides. *Journal of the American Society for Mass Spectrometry* **19**, *1*, 138-150.

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