Analysis of Sugars in Glycosylated Woody Biomass with the Agilent 1200 Series LC System

Application Note

Biofuels and Alternative Energy

Author

Atsuko Naito
Agilent Technologies Japan
Hachioji, Tokyo

Abstract

Although bioethanol is currently produced mainly from edible plants, research is underway into production methods based on non-edible plants, for example, wood. When wood is used as a raw material, glycosylation and fermentation are carried out by turning cellulose, hemicellulose, and lignin into small molecule compounds and subjecting the cellulose and hemicellulose to the action of enzymes. This is an example of analysis of sugars in wood sugar solutions obtained by low environmental impact hydrothermal treatment and mechanochemical treatment followed by enzyme-based glycosylation. The samples were kindly provided by Mr. Shigeki Sawayma, Head of the Research Team, and Mr. Katsuji Murakami, Chief Researcher, from the Biomass Research Center of the National Institute of Advanced Industrial Science and Technology.

Agilent Technologies

Configuration

Agilent 1200 Series LC System

- Agilent 1200 Series Quaternary Pump (G1354A)
- Agilent 1200 Series Standard Autosampler (G1329A)
- Agilent 1200 Series Thermostatted Column Compartment (G1316A)
- Agilent 1200 Series Evaporative Light Scattering Detector (G4218A)

Analytical Conditions

Column:	Shodex Asahipak NH2P-50 4E
Mobile phase:	Water/acetonitrile = 20/80
Flow rate:	1.0 mL/min
Column temperature:	30 °C
Injection volume:	20 µL
Sample concentration:	1000 ng/µL

A chromatogram of the reference solutions of sugars typically detected in wood sugar solutions is shown in Figure 1. Figures 2–6 show analytical results for wood sugar solutions obtained using different pre-treatment methods and raw materials. The samples were obtained by diluting wood sugar solutions with a mixture of water and acetonitrile (1:1) and passing the diluted solutions through a 0.22-µm filter.

The amount of the produced sugars and their ratios varied greatly depending on whether hydrothermal treatment or ball mill treatment was used. In addition, the amount of the produced sugars varied depending on the raw materials.

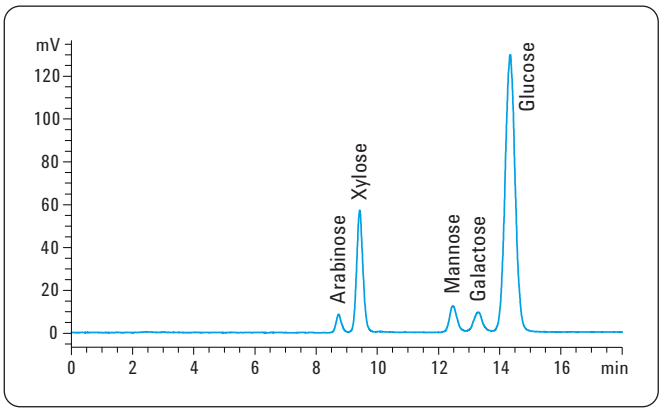

Figure 1
Chromatogram of reference solutions (1000 ng/µL each).

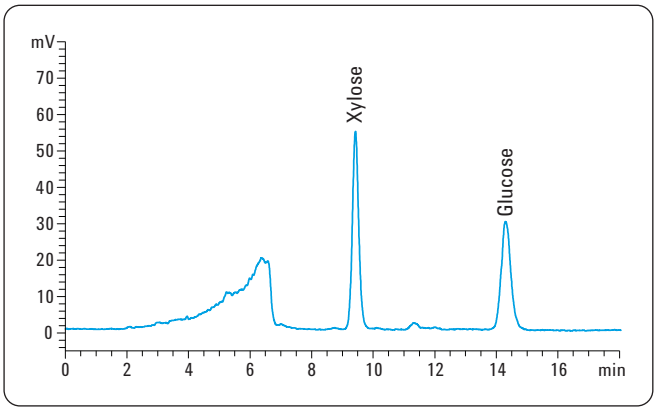

Figure 2
Bagasse, hydrothermal 180 °C 5 min.

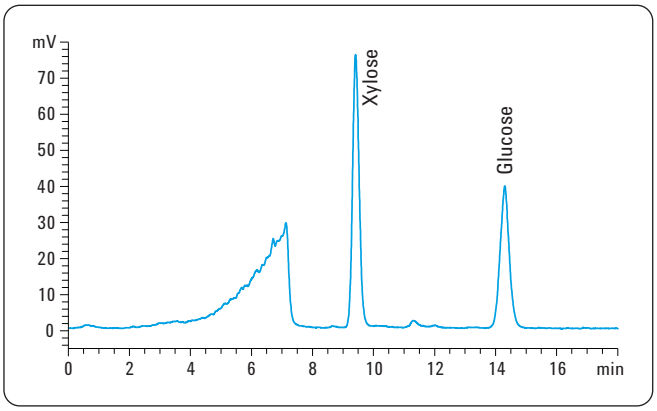

Figure 3
Bagasse, hydrothermal 160 °C 15 min.

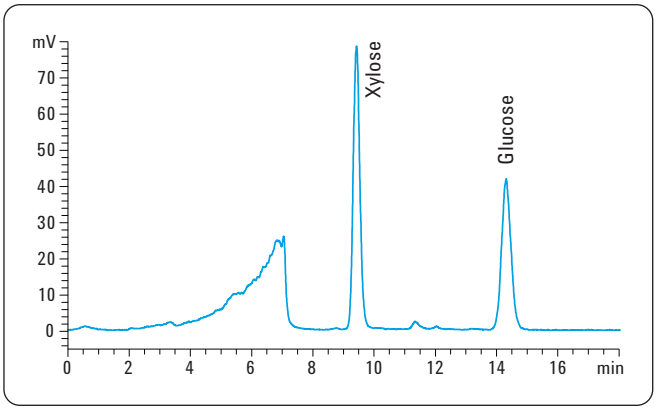

Figure 4
Bagasse, hydrothermal 160 °C 30 min, w/phosphoric acid.

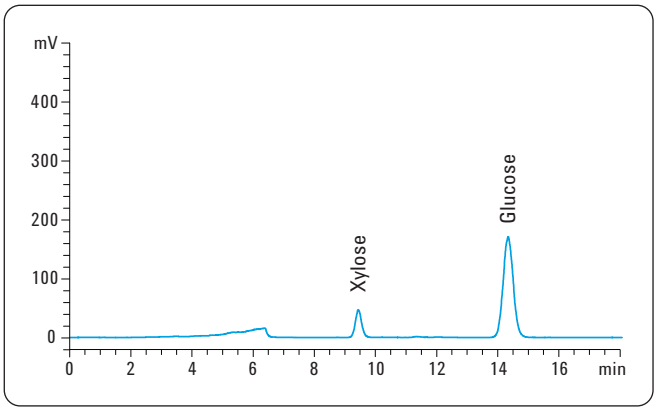

Figure 5
Bagasse, ball mill.

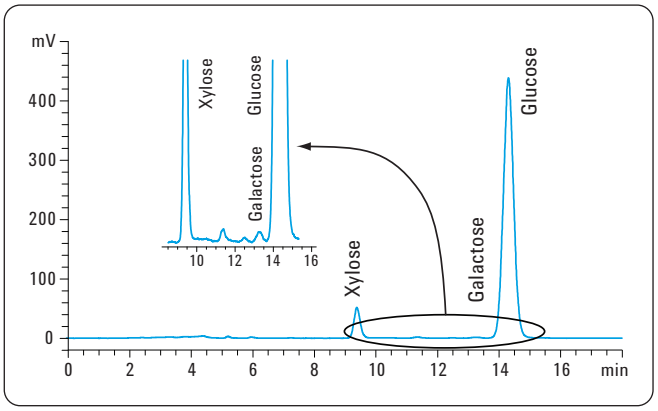

Figure 6
Eucalyptus, ball mill.

Figures 7–10 show analytical results for 2-fold dilution. A mannose peak was detected and several peaks believed to belong to oligosugars were observed subsequent to glucose elution. In addition, an unknown peak was detected prior to the mannose peak. It was found that there were few peaks believed to belong to oligosugars when the hydrothermal treatment was used and there were many peaks when the ball mill treatment was used.

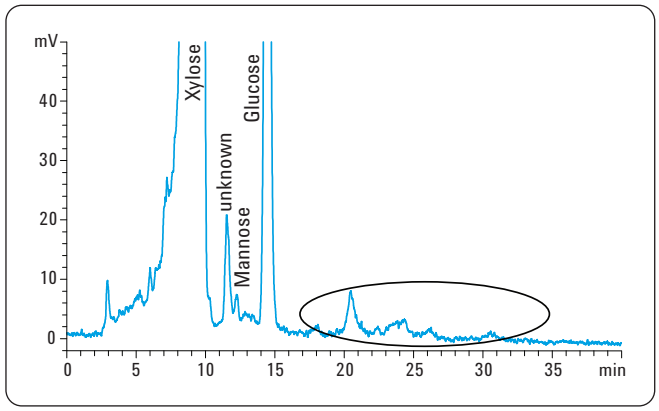

Figure 7
Bagasse, hydrothermal 180 °C 5 min (2-fold dilution).

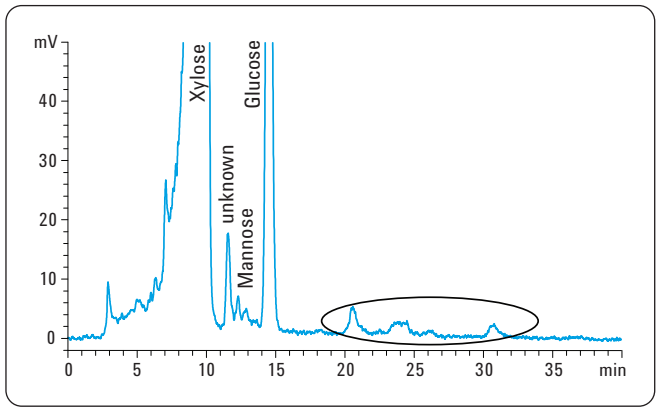

Figure 8
Bagasse, hydrothermal 160 °C 15 min (2-fold dilution).

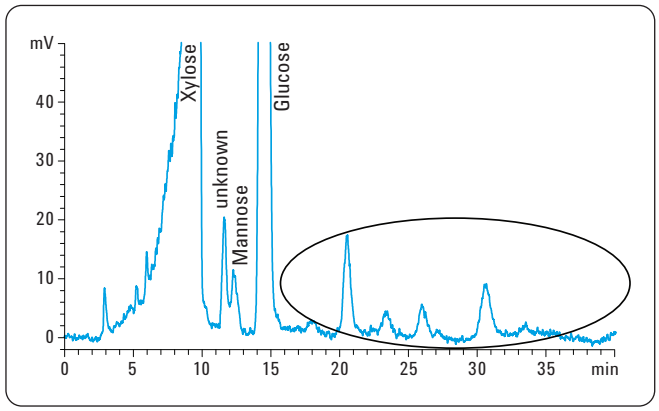

Figure 9
Bagasse, ball mill (2-fold dilution).

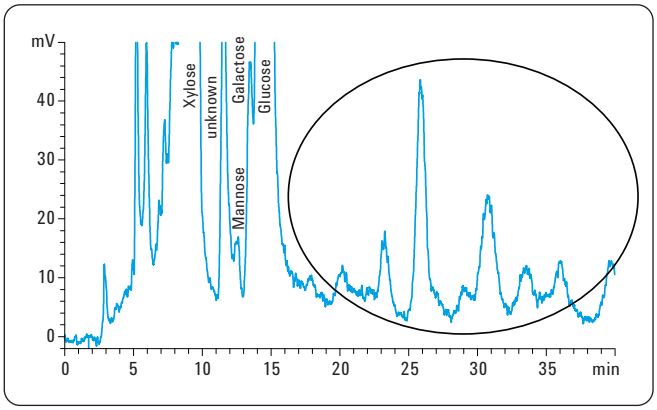

Figure 10
Eucalyptus, ball mill (2-fold dilution).

Conclusion

Sugars in glycosylated woody biomass are mainly xylose and glucose, but the concentration depends on the pre-treatment process. The Agilent 1200 Series LC system with the evaporative light scattering detector is suitable for sugar analysis in glycosylated woody biomass due to good sensitivity and good usability.

www.agilent.com/chem/lc

Published February 1, 2011
Publication Number 5990-7318EN

Agilent Technologies