Potential Monolignol Replacement: Synthesis of Feruloyl Quinic Acids and Feruloyl Threitols

Martina Opitnik, Paul Schatz, Hoon Kim, Christy Davidson, John Grabber, and John Ralph

1Department of Biochemistry, University of Wisconsin–Madison; 2US Dairy Forage Research Center, USDA-ARS

Introduction
Evidence suggests that the lignification is particularly malleable. Many monomers besides the three traditional monolignols are now well authenticated as being incorporated into the polymer in a variety of natural and transgenic plants. Introduction of new monomers is seen as a possible means for changing the structure of lignin. Some structural changes can result in lignins that contribute less to the recalcitrance of cell walls toward saccharification, benefiting either microbial digestion of forages in ruminants or industrial enzymatic saccharification of cell wall polysaccharides to sugars destined for fermentation to liquid biofuels.

Two partial monomer-replacement strategies (we contend that complete replacement of lignin monomers is unwise) are being considered here. The first is to reduce the hydrophobicity of fiber lignins to allow improved enzyme accessibility to the polysaccharides. Here we explore the introduction of hydrophilic groups via quinic acid and threitol esters of ferulate – ferulates are known to incorporate well into lignins. The second is to use biphenolic conjugates (here we explore 1,5-di-feruloyl quinic acid and 1,4-O-di-feruloyl threitol) to introduce readily cleavable ester linkages into the backbone of the lignin polymer. Cleaving the esters will result in lignin depolymerization under milder conditions than is possible for ‘normal’ lignins, helping extract the lignins away from the polysaccharides, again improving enzymatic access to those polysaccharides.

Monolignol Substitutes

1,5-Di-feruloyl quinic acid (1)
- Increased hydrophobicity
- Novel predetermined breaking point (ester)
- Difunctional lignification model

1,4-Disubstituted feruloyl threitol (3)
- Increased hydrophobicity
- Novel predetermined breaking point (ester)

5-Feruloyl quinic acid (2)
- Increased hydrophobicity
- Novel predetermined breaking point (ester)

1-O-Feruloyl threitol (4)
- Increased hydrophobicity
- Novel predetermined breaking point (ester)

1-O-Disubstituted feruloyl threitol (8)
- Increased hydrophobicity
- Novel predetermined breaking point (ester)

Syntheses

Feruloyl quinic acids (1&2)
Feruloyl threitols (3&4)

NMR spectra (HSQC) of whole cell walls
Inclusion of the model compounds into the lignification process.

The spectra from cell walls lignified with added 5-feruloyl quinic acid 2 surprisingly contained only correlations of guaiacyl units (derived from coniferyl alcohol) and syringyl units (derived from sinapyl alcohol). The absence of ferulate correlations indicates that 2 was not effectively incorporated into the wall bound lignin. In the case of added 1,4-di-O-feruloyl threitol 3, correlations from the ferulate units are readily observed. These correlations indicate the integral incorporation of the monomer-substitute into the wall bound lignin. A minor concern is that difeurolyl polyols depleted peroxydase activity, resulting in decreased lignification.

Conclusion
Our objective is to find the best lignin monomer substitutes to allow simpler biomass processing.
- Straightforward pathways for the synthesis of the required monomer-substitutes were developed, allowing the syntheses of these compounds on a gram scale.
- 2D NMR methods allow their incorporation into artificially lignified cell walls to be tracked. Feruloyl quinic acids do not appear to provide a promising strategy as incorporation was not observed.
- Incorporation of difeurolyl threitol appears to be worth pursuing if saccharification of the walls is enhanced (with or without pretreatment).
- In ongoing work the incorporation of these strategic monolignol derivatives into the lignin is being analyzed and the effects on cell wall degradability are being studied.

References