An Unusual Lignin From Kenaf

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Introduction

In a collaborative project with the U.S. Forest Products Laboratory, we have been examining the structure of the lignin from kenaf, a dicot. Although it does not have much to do with forages, the novel lignin is helping us to assign and determine more about the structure of lignins in forage grasses and legumes. Lignins have been characterized from such a variety of plant materials that it seems unlikely for a new type of lignin to be revealed. The lignin from kenaf is, however, strikingly different from any other lignin that has been characterized. Its uniqueness is revealed both in terms of its high syringyl content and in the extensive acetylation of the side-chain hydroxyls.

Experimental

Kenaf lignin was isolated by traditional procedures from bast fibers from the core of Tainung Kenaf stems. NMR spectra were run on the Center's Bruker AMX-360.

Results and Discussion

The kenaf lignin was initially intriguing because of its high syringyl content. Dicots typically have a rather even syringyl:guaiacyl distribution, whereas nitrobenzene oxidation of the kenaf dioxane lignin gave a molar syringaldehyde:vanillin ratio of 6.0 (without loss corrections). Syringaldehyde:vanillin ratios in hardwoods have been reported up to *ca* 5.2 and a thesis describes an 8.4 ratio. The dominance of syringyl units in this kenaf leads to a very simple lignin as seen from the ^{13}C-NMR of the acetylated kenaf dioxane lignin, Fig. 1a. What is striking, in addition to the obviously high syringyl:guaiacyl ratio, is that β-ethers predominate significantly over other interunit linkage types (cf. the β-β units at *ca* 55.5 ppm), and that the material is highly etherified (since the proportion of phenolic endgroups, seen from the aromatic acetate carbonyl peaks, is low). These values have been estimated from quantitative ^{13}C NMR at ~80% β-ether units and 14% phenols. The aliphatic region of the 2D HMQC-TOCSY spectrum, Fig. 2, confirms the assignment of

sidechain protons, again shows that syringyl β-ether units predominate, and beautifully disperses the isomers. The prior observations that *erythro* β-ether isomers predominate in syringyl units is strikingly revealed in this spectrum; there is little diastereoselectivity in guaiacyl lignins.

The NMR spectrum of the un-derivatized lignin, Fig. 1b, provided even more startling information — the lignin was extensively acetylated (*ca* 50%). The spectrum shows that acetylation was almost entirely (*ca* 95%) at the primary γ-position of the sidechain, as was confirmed by an HMBC experiment correlating the acetate carbonyl carbon with γ-protons on lignin (not shown). Although acylation of dicot lignins by hydroxycinnamic acids has previously been reported to be almost insignificant, grasses have proportions of sidechain alcohol groups esterified by *p*-coumaric acid; such esterification has recently been shown to be exclusively at the γ-position in a maize lignin isolate and indications are that other grasses, both C_3 and C_4, imply that grasses pre-esterify lignin monomers to produce the hydroxycinnamyl *p*-coumarates which are then exported and incorporated into a lignin complex by the traditional oxidative coupling mechanisms. Presumably, the same can be concluded in the case of the kenaf in this study; the acetate is not so cleanly at the γ-position (*ca* 95%), but we have previously observed acetate migration in lignin model compounds. We suspect that, in kenaf, sinapyl (and perhaps coniferyl) acetates are enzymatically produced and exported for lignification with sinapyl and coniferyl alcohols. It is most likely that the small amount of α-acetate derives from internal migration rather than from the alternate mechanism of acetate addition to quinone methide lignin intermediates, a mechanism over which the plant has no direct control. The presence of such large amounts of acetate on lignin was sufficiently novel that our procedure for the lignin isolation came under close scrutiny. The isolation was repeated under conditions in which no reagents or solvents could conceivably produce
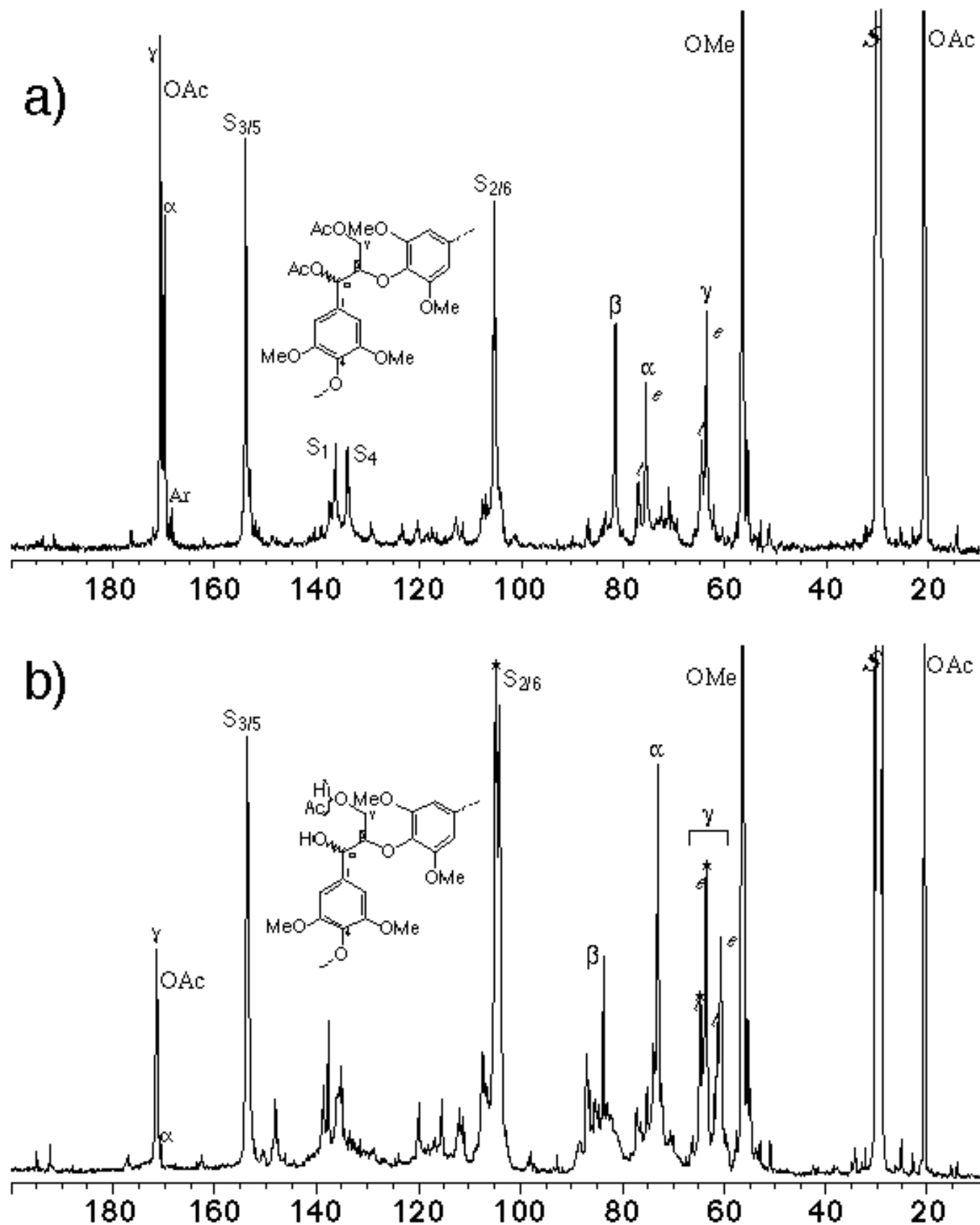
Figure 1. ^{13}C NMR spectra of the isolated Kenaf lignins showing the preponderance of syringyl peaks. Assignments are for β-ethers: a) acetylated, b) underivatized, * indicates easily identifiable peaks that result from acetate substitution.
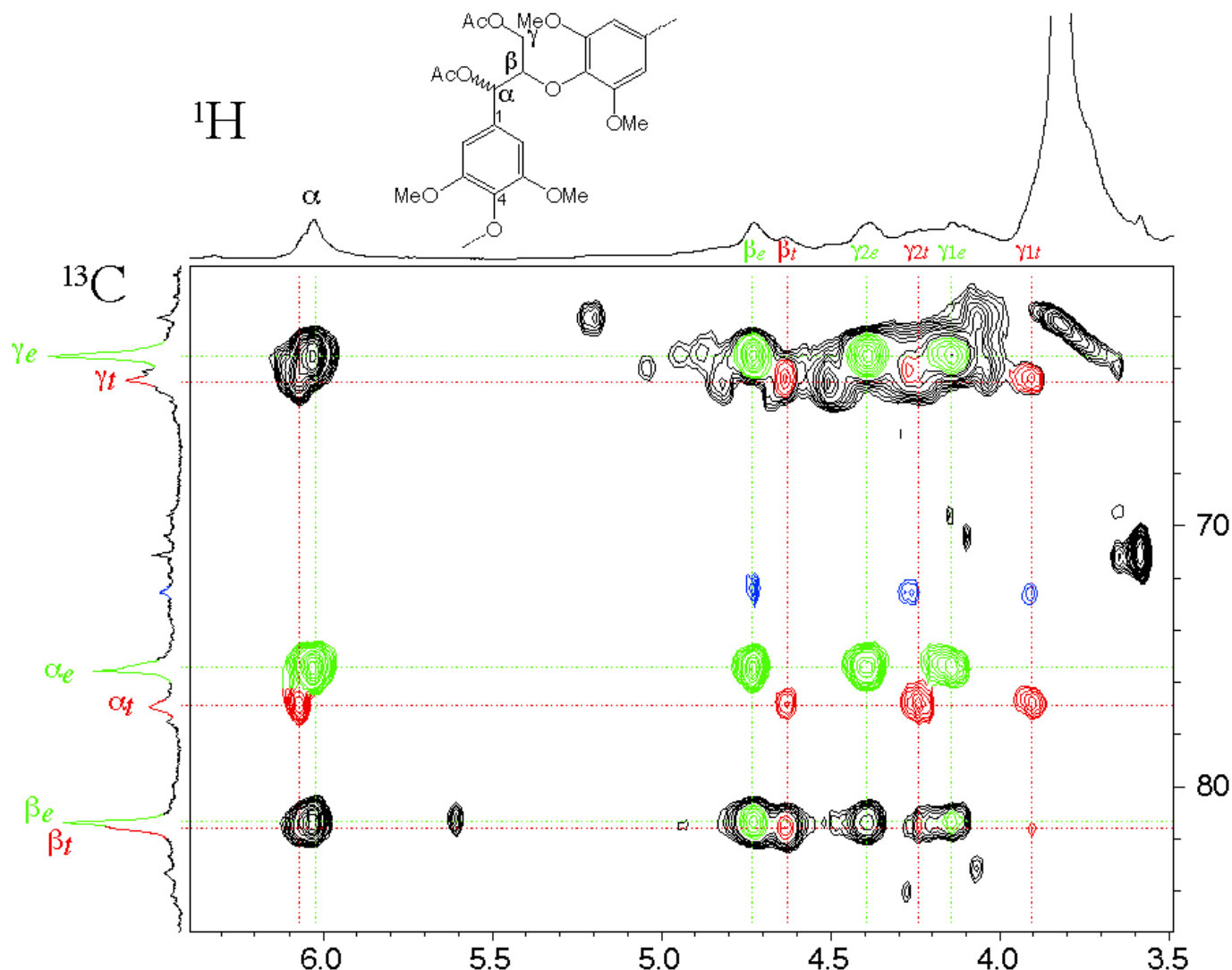
Figure 2. Aliphatic region of a 2D HMQC-TOCSY experiment showing syringyl β-ether units and the excellent dispersion of erythro- and threo-isomers.

acetylation artifacts; the lignin isolated in this manner retained the extensive acetylation.

We are currently intrigued by what might be the function of such an acetylated lignin for the plant, and if somehow (and for some reason) this is a mechanism to achieve high syringyl lignins — syringyl rich synthetic lignins with high molecular weights are difficult to prepare in vitro. Of practical relevance is that the presence of acetate groups on kenaf lignin will consume base during alkaline delignification, but that delignification of this lignin fraction should be relatively straightforward and extensive because the high-syringyl lignin is relatively unbranched and, as

seen from the ^{13}C-NMR spectra, contains a substantial proportion of high-temperature-base-cleavable β-aryl ether units.

In summary, kenaf lignin is unusual and perhaps unique. It actually represents a new lignin type implicating new monomers (the 4-hydroxycinnamyl acetates) in its synthesis. The NMR spectra, being relatively simple and syringyl rich, are providing data for syringyl residues that we did not have and are therefore valuable in aiding assignment and structural elcuidation of more traditional forage lignins.