Comparative Enzyme Biodegradability of Xylan, Cellulose, and Starch Derivatives†

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The enzyme biodegradability of xylan and of two of its prominent derivatives, (hydroxypropyl)xylan and xylan acetate, were examined comparatively vis-à-vis similar cellulose and starch derivatives, using a commercially available xylanase-rich enzyme preparation. The results indicate that xylan, like cellulose and its derivatives, is subject to enzymatic depolymerization in relation to degree of substitution. Degradation paralleled the presence of unmodified xylose units in the polysaccharide. Degradation proceeded rapidly, with greater than 80% of total degradation being complete after only 1 day. A comparison with starch and cellulose derivatives revealed that hydroxypropyl xylan suffers less biodegradability loss as substitution rises to 0.25 or 0.4, after which loss of degradability parallels that of the corresponding cellulose and starch derivatives. This apparent tolerance to low degrees of substitution is attributed to molecular heterogeneity which suggests that constituents other than xylose become propoxylated initially. Xylan acetate was found to lose enzymatic biodegradability at a rate which parallels that of cellulose acetate, and this is 35% slower (with respect to DS) than corresponding starch acetates. Size exclusion chromatography results suggest that xylan degrades to monosaccharides, that xylan derivatives degrade to monosaccharides and oligosaccharides, and that substitution with lignin prevents xylan from being recognized by xylanolytic enzymes, resulting in loss of degradability.

Introduction

Xylan is a representative of a class of branched heteropolysaccharides consisting of poly(anhydroxylose) with substituents of 4-*O*-methyl glucuronic acid, acetyl groups, and anhydroarabinose units. Xylans from lignifying plants are rarely free of lignin, which is attached covalently (*1*). Many agricultural residues, such as sugar cane bagasse, corn cobs, straw, etc., have as much xylan (25–35%) as cellulose (30–40%) (*2–4*). Xylans are easily degraded when lignified biomass is processed by chemical means (*5, 6*). The isolation of xylans using different isolation options, and the chemical modification by ester and ether-forming reactions, have been described in recent publications (*7, 8*).

Chemically, xylan resembles cellulose, both being β-1,4-linked polyglycopyranosides. In morphological terms and properties-wise, xylan is more like starch, partially crystalline (and branched), and highly interactive with water (*9, 10*).

The enzymatic degradation of fiber-bound xylans has recently received much attention in relation to pulp-bleaching studies (*11, 12*). Whereas xylans attached to pulp fibers resisted solubilization by xylanases, presumably due to limited enzyme accessibility (*11*), isolated xylan substrates underwent solubilization in relation to their degree of substitution (*11*).

Cellulose and starch derivatives have recently found attention as potential biodegradable plastics and components for biodegradable plastics (*13–18*). The biodegradability of cellulose esters has been examined using a simplified enzyme biodegradability assay based on commercially available cellulase preparations (*19*).

The objectives of this study revolve around the comparative evaluation of xylan, cellulose, and starch derivatives using an enzyme biodegradability assay developed in earlier work (*19*).

Materials and Methods

A. Materials. ***a. Xylans.*** Xylans were isolated in accordance with earlier work by alkali extraction of barley husks (*7*). This extraction involved (a) direct extraction with aqueous alkali from barley husks (X_A), (b) alkali extraction of barley husks previously delignified using a conventional chlorite treatment (X_C), and (c) alkali extraction of an organosolv-delignified barley husk resource (X_O). Oat spelt xylan (X_{OS}) was obtained from Sigma Chemicals (St. Louis, MO). Cellulose was obtained from Whatman under the designation CF-11 (Whatman Chemicals, Hillsboro, OR). Food grade corn starch was used in all the starch preparations.

b. Xylan Derivatives. (Hydroxypropyl)xylan (HPX) was obtained by mild propoxylation of alkaline xylan solutions in accordance with earlier work (*8*).

Xylan acetates were prepared by the following procedure. Xylan extracted from barley husks (15 g) was dissolved in 60 mL of formamide. Acetic anhydride (30 mL) was added slowly at ice-cold temperature. The mixture was stirred at room temperature for 12 h, and then an additional 30 mL of acetic anhydride was added slowly. The reaction was continued for a total period of 24 h and then heated at 80 °C for 1 h. The mixture was precipitated in ice-cold 0.5% hydrochloric acid. The

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precipitate was filtered, washed well with water, and dried. Total yield of xylan acetate was 17 g. The xylan acetate was saponified in ethanolic alcohol to the desired DS according to ASTM D871 method.

c. Cellulosic Derivatives. Cellulose acetates of different degrees of substitution (DS 1–3) were prepared by dissolving cellulose in dimethylacetamide/lithium chloride (DMAC/LiCl) and subsequently treating this solution with acetic anhydride in pyridine as reported earlier (*20*). Heterogeneously prepared cellulose acetates were obtained from Eastman Kodak (Kingsport, TN). Hydroxypropyl cellulose (HPC, Klucel) used in this study was supplied by Aqualon, Inc., Wilmington, DE.

d. Starch Derivatives. For the preparation of starch acetates, 450 g of commercial corn starch was solvent-exchanged first with methanol, three times, an hour each time, and then with DMAC three times, an hour the first two times, followed by overnight soaking. The solvent-exchanged starch (82 g) was stirred into DMAC/LiCl (2 L/140 g) overnight under dry nitrogen until completely dissolved. Acetylation was accomplished by treating a 4% starch solution with acetic anhydride in pyridine at 60 °C for 24 h under nitrogen. Starch acetate was precipitated and washed several times with methanol and dried overnight under vacuum at 60 °C. Starch acetates with a wide range of substitution were prepared by stoichiometric control of acetic anhydride.

B. Biodegradability Assay. The assay procedure by Glasser et al. (*19*) was followed. In summary, polysaccharides and their derivatives were incubated with the appropriate enzyme preparations until three consecutive readings were similar. The incubation mixture was tested for free monosaccharides by the Stanbio assay (Stanbio Direct Glucose procedure No. 0270, Stanbio Lab., Inc., San Antonio, TX).

Xylanolytic enzyme biodegradability (XEB) is defined as weight percent xylose equivalent yield and is calculated using eq 1

$$\text{XEB (\%)} = \frac{132.1 + xp}{150.1} y \qquad (1)$$

$$\text{calcd xylose yield (\%)} = \frac{(1-p) \times 63.9 \times 150.1 \times 100}{100 \times 132.1 + 63.9 \times 58p} \qquad (2)$$

where

63.9 = wt % anhydroxylose in X_c

58 = molecular weight of propylene oxide

150.1 = molecular weight of xylose

132.1 = molecular weight of anhydroxylose

x = net increase in the molecular weight per hydroxyl group

p = degree of substitution

y = experimental xylose equivalent yield (%) (ash free)

Xylan and hydroxypropyl xylans were incubated with Ecopulp-60 (predominantly xylanase; ALKO Ltd., Helsinki, Finland) in 20 mM acetate buffer (pH 5.3), whereas xylan acetate (XA) was treated with Econase C.E. (containing both cellulase and hemicellulase, ALKO Ltd., Helsinki, Finland) in 20 mM acetate buffer (pH 4.8). Starch acetates (SA), cellulose acetates (CA), and hydroxypropyl cellulose were incubated also with Econase C.E. in 20 mM acetate buffer (pH 4.8). Econase C.E. contains endo-1,4-β-D-glucanase, cellobiohydrolase, and *exo*-1,4-β-D-glucosidase as principal enzymes, and it contains hemicellulase, β-glucanase, protease, and amyloglucosidase as minor components. Amyloglucosidase can hydrolyze both α-1,4 and α-1,6 bonds in starch. This is why Econase C.E. was selected as enzyme for the biodegradability assay of both starch acetates as well as xylan and cellulose acetates. This choice was tested by comparative degradations of starch acetate with (a) α-amylase only, (b) amyloglucosidase only, and (c) combinations of α-amylase and amylo-glucosidase. Since the results obtained in the latter three experiments were essentially identical to those obtained with Econase C.E., all assays were conducted using the latter at its optimum pH.

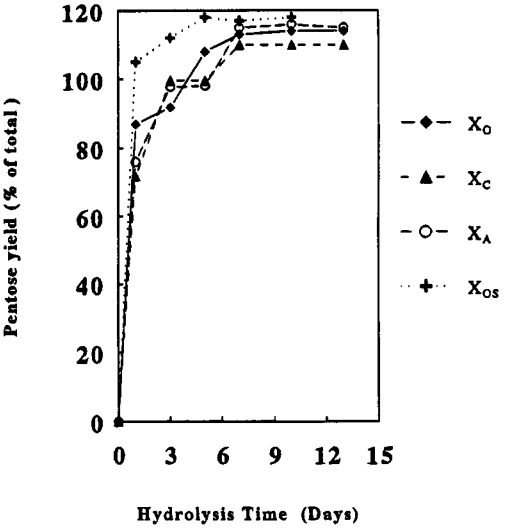

Figure 1. Enzymatic hydrolysis of xylans from different sources: X_o, organosolv-delignified barley husk; X_c, chlorite-treated barley husk; X_A, barley husk alkali extraction; X_{os}, Oat spelts.

Size exclusion chromatography was performed using a Waters HPLC System consisting of a UV detector (Waters Lambda Max model 481) in series with a differential refractive index detector (Anspec Co., Erma ERC 7512) and three Biogel columns (Biorad model TSK 20-XL and SEC 30-XL) with pore sizes of 120, 120, and 250 Å. Separation conditions involved ambient temperature and a flow rate of 0.6 mL/min with distilled, degassed, filtered water as the solvent. All analyses were conducted in duplicate.

The acetyl contents of all polysaccharide derivatives were determined by ^{1}H-NMR (Bruker 270 MHz and Varian 400 MHz) and gas chromatography (*21*). Ash contents were determined by a Perkin-Elmer TGS-2 thermogravimetric analyzer. The samples were scanned, 10 °C/min, under air constantly flowing at 45 mL/min.

Results and Discussion

The enzymatic biodegradation (EB) of isolated (and partly commercially available) xylan preparations from different sources using commercially available xylanase-rich polysaccharidase enzyme mixtures reveals the usual degradation behavior with time (Figure 1). Regardless of purity and method of isolation, xylans degrade rapidly with more than 80% of total degradation being complete within one day. Almost lignin-free xylan from oat spelts (X_{os}) exceeds the degradation rate of all other xylans by ca. 20% in day one. All preparations were found to

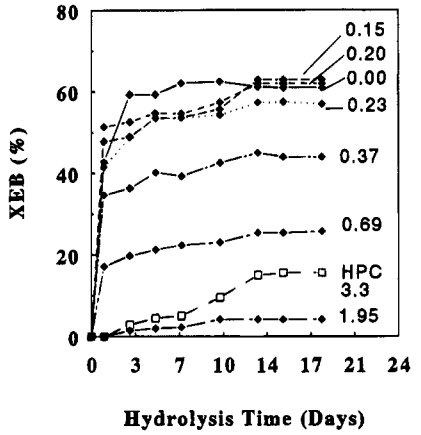

Figure 2. Xylanolytic enzyme biodegradability (XEB) of (hydroxypropyl)xylan (HPX_C) with different degrees of substitution (DS). HPC, hydroxypropyl cellulose.

produce pentoses in virtually quantitative yield (114% by weight considering the uptake of 1 equiv of water/equiv of anhydropentose unit). Theoretical yields are achieved in approximately 7 days of incubation with enzyme.

Xylanolytic enzyme biodegradability (XEB) of (hydroxypropyl)xylan (HPX) preparations vs time, measured as pentose yield per total mass (corrected for ash content) (Figure 2), reveals that as DS rises, XEB declines at a rate equivalent to DS and that XEB falls below 10% when DS rises above 1.5. DS has no significant effect on rate, as most derivatives complete greater than 80% of their total degradability within 1 day.

Hydroxypropyl cellulose (HPC) (with molar substitution, MS, of 3.3) differed from HPX by displaying accelerated degradation after day seven. Total degradation, however, remained below 20% after 18 days. No explanation can be given.

The relationship between XEB and DS for three (hydroxypropyl)xylans that were isolated by different methods (Figure 3) reveals consistent and virtually exponential loss of degradability with DS rising beyond a threshold DS (Figure 3). This distinct two-phase behavior, with a slow degradability loss gradient below a DS of between 0.25 and 0.4 prior to a rapid degradability loss, has not been observed with any other polysaccharide or polysaccharide derivative. This unusual tolerance to biodegradability suggests that a modest degree of propoxylation results in little or no loss of enzymatic recognition of xylans.

A comparison of enzyme degradability (EB) in relation to DS for several other types of polysaccharide derivatives, starch acetate, cellulose acetate (both prepared by homogeneous and by heterogeneous methods) (*19*, *20*), and xylan acetate reveals a steady loss of degradability with rising DS (Figure 4). The rate of EB loss is virtually identical for cellulose and xylan acetates but slower than that for corresponding starch acetates. Starch acetates lose their enzyme biodegradability 150 and 120% faster with DS rising than corresponding cellulose and xylan acetates. A slope of d(EB)/d(DS) of −100 would indicate theory assuming that all monosubstituted anhydromonosaccharide units become unrecognizable by enzymes following monosubstitution. These differences in degradability were established for the same enzyme system (Econase C.E.) operating at a constant pH of 4.8. This pH level provides optimum conditions for enzyme activity, but it is considered inoptimal for deacetylation. Buchanan et al. (*13*) found that cellulose diacetate degradation by an *in vitro* enrichment cultivation technique required a pH of 7.1 for maximal degradation rates, apparently by promoting hydrolytic deacetylation. The two isolated (but mixed) polysaccharidase enzyme systems employed in this study had optimum activities at pH 4.8 and 5.1, and no effect of pH on degradability was noticed. These pH levels were virtually identical to that used by Viikari et al. (*11*) with xylanase (pH 5).

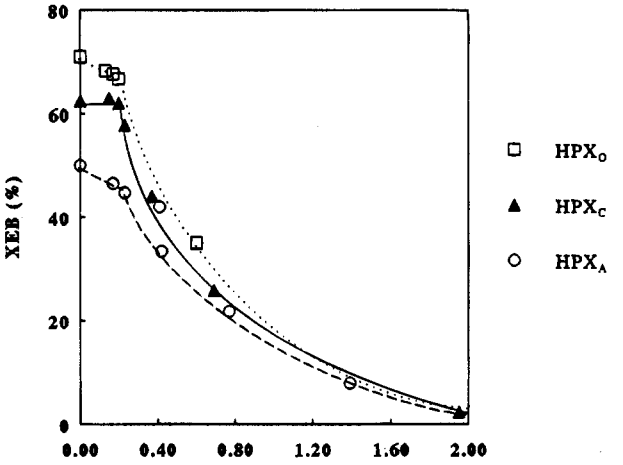

Figure 3. Xylanolytic enzyme biodegradability (XEB) of HPX from different sources in relation to DS. (See Figure 1 for sources of xylan.)

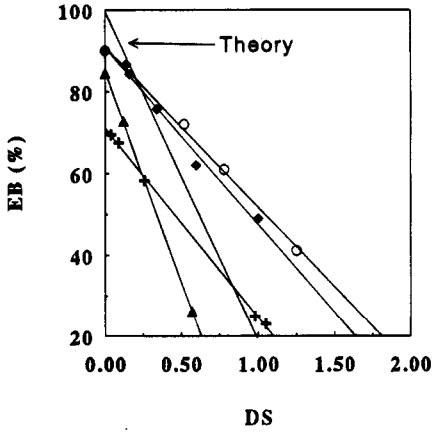

Figure 4. Enzyme biodegradability (EB) of different polysaccharide acetates in relation to DS. (▲) Starch acetate; (♦) cellulose acetate (homogeneous); (○) cellulose acetate (heterogeneous); (+) xylan acetate.

This behavior, linear loss of EB with rising DS (−d(EB)/d(DS)), exhibited by polysaccharide acetates is in accordance with theory and consistent with the degradability behavior of cellulose esters (*19*). This behavior, however, is contradicted by data reported by Mitchell et al. (*22*), who found no loss of degradability of xylan acetates with degree of substitution rising from 0 to 0.8 (Figure 5). This difference can be explained with differences in assay procedure, method of xylan acetate preparation, or differences in enzyme or microbiological activity.

Three different behaviors can now be distinguished with respect to EB vs DS. These involve linear loss of EB (as in the case of cellulose, starch, and xylan acetates); complete insensitivity of degradability (and enzyme recognition) to DS with DS rising from 0 to nearly 1 (as suggested by Mitchell et al. (22) for xylan acetate); and a mixed behavior, slow loss of degradability followed by rapid (and exponential) loss (as in the case of HPX) (Figure 5).

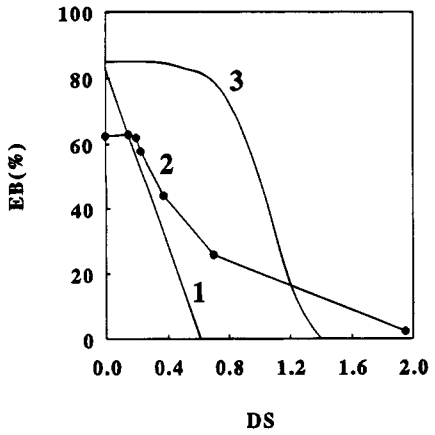

Figure 5. Summary of enzyme biodegradability (EB) of different polysaccharide derivatives in relation to DS. Note: (1) a cellulose hexanoate of Glasser et al. (*19*); (2) the author's data HPX_C, and (3) from Mitchell et al. (*22*) for XA.

Assuming uniform substitution of all xylan constituents by propylene oxide (PO) during propoxylation and all monosubstituted xylose units becoming unrecognizable by enzymes, loss of enzyme recognition of the derivative should be related only to DS. Thus, a linear relationship is expected for XEB with DS rising from 0 to 1 (Figure 5). This behavior was observed, more or less, by xylan acetates studied here and cellulose esters described in the previous study (*19*). Data for HPX by contrast are a poor representation of this theory (see Figure 3). A comparison of the xylose content (determined by acid hydrolysis and by enzymatic digestion) with that calculated from DS data (Figure 6) led to a possible interpretation of the anomalous behavior.

The calculated curve was constructed using eq 2. It assumes that (a) the composition of starting material, X_c (63.9% AHX plus others, see Table 1), does not change during hydroxypropylation, (b) hydroxypropylation occurs only on xylan, and (c) only monosubstitution of anhydroxylan units takes place between the DS of 0 and 1. The experimental data representing the enzymatic and acid hydrolysis values, however, always exceed the calculated values (Figure 6). In other words, both enzyme and acid hydrolysis indicate more free xylose units than are predicated by calculation. This observation suggests that xylan propoxylation is heterogeneous in nature. At low levels of DS, the modification takes place preferentially on nonxylose components. This seems to be the case at high levels of DS as well, where chain extension of PO units is more prevalent than ring substitution. Nonuniformity of propoxylation in relation to xylan components therefore must be held responsible for the observed noncompliance of XEB behavior with DS of HPX preparations. Since xylan contains small amounts of lignin and since phenolic OH groups react more rapidly with PO in alkali, the presence of lignin may well explain the apparent greater tolerance of xylans to low levels of chemical modification by PO. This is further supported by the observed differences between HPX_A, HPX_O, and HPX_C, the latter (which has been isolated after a bleaching process) having a less pronounced tolerance phase than the former, probably on account of oxidative degradation of phenolic aromatic rings during isolation.

SEC Analysis

An analysis of the water-soluble enzyme incubation-mixture by size exclusion chromatography (SEC) produced evidence for the stepwise hydrolysis of xylan to mixtures of monosaccharides and oligosaccharides (Figure 7). The hydrolysis mixture of xylan analyzed by UV and RI sequential monitoring revealed three distinct component groups different by retention time. These were (A) low molecular weight compounds detected by both UV_{280} and RI (40–46 min retention time) (glucose and xylose eluting at 42 and 43.5 min, respectively, under the same conditions); (B) substances of medium molecular weight, such as oligosaccharides (35 to 40 min of retention time) which are detectable only by RI (cellobiose is retained for 40 min under the same conditions); and (C) a group of high molecular weight substances (20–30 min retention time) which are often split into two separate peaks, one strongly UV_{280}-absorbing (at 20 to 25 min retention time) and one absorbing both UV and RI (at 26 min) (Figure 7). The most highly retained substances represent monosaccharides and buffer, followed by oligosaccharides at shorter retention times and followed by (water-soluble) lignin (which elutes at the same retention time as a nonionic, water-soluble hydroxyethyl lignin, source: Westvaco, Charleston, S. C., 20 min) as well as a lignin–xylan complex rich in carbohydrates (and therefore raising a strong RI signal). Whereas the starting material consists primarily of a polymeric lignin-xylan complex and, on occasion, of barley husks' water-soluble lignin responsible for a strong UV peak at 22 min (which disappears after two hours of incubation with xylanolytic enzymes), and of buffer (at ca. 46 min), the 2-h incubation product displays a greatly enhanced monosaccharide peak, a very distinguishable oligosaccharide peak, and a reduced polymer peak. After 17 days of incubation, the water-soluble product mixture represents primarily monosaccharides followed by a weak polymeric signal representing a lignin–xylan complex and only a trace of oligosaccharides (Figure 7). (The polymeric lignin fraction at 22 min retention time is very pronounced with X_A and X_C preparations in which this

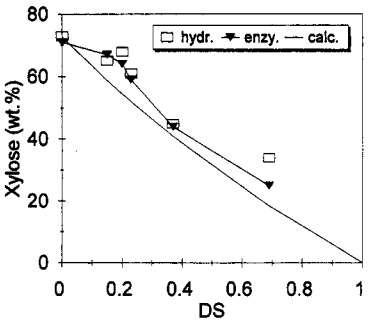

Figure 6. Xylose content of HPX_C in relation to DS. Theoretical line is based on eq 2. Enzymatic hydrolysis data from Figure 3.

Table 1. Composition of Xylan and (Hydroxypropyl)xylan

	carbohydrate composition[a]					DP		
materials	AH	AHG	Σpent	Σhex	lig	DP_n	DP_o	DS
				Xylan				
X_c	68.9	–	69.6	–	25.6	130	360	
X_o	69.0	–	75.6	–	11.2	50	130	
X_A	50.5	5.8	60.9	8.7	22.5	140	1550	
X_{os}	62.1	11.7	69.7	11.7	3.3	595	2075	
				(Hydroxypropyl)xylan (HPX_c)				
	57.5	0.9	60.0	0.9	34.3			0.15
	59.8	0.9	62.9	0.9	32.3			0.20
	53.5	1.6	56.6	1.6	34.8			0.23
	41.9	1.7	43.7	1.7	31.9			0.37
	24.7	2.0.	30.6	2.0	29.8			0.69
	12.7	1.1	19.4	1.1	30.8			1.95

[a] AHX, anhydroxylose; AHG, anhydroglucose; Σpent, AHX + anhydroarabinose; Σhex, AHG + anhydrogalactose; lig, lignin.

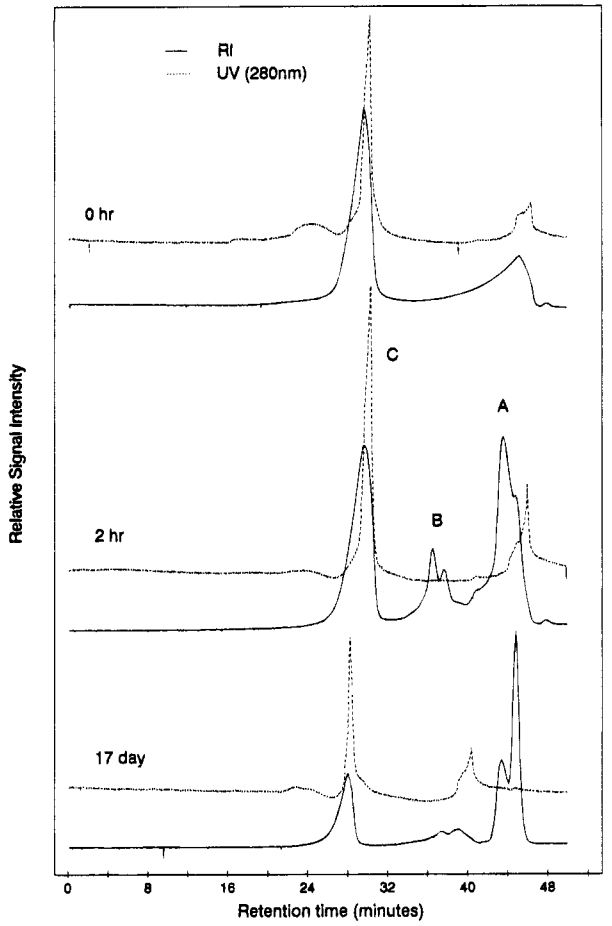

Figure 7. SEC of X_O in relation to enzyme hydrolysis time.

disappears after a very brief incubation period with enzyme. This phenomenon can be explained with the loss of water solubility of lignin–xylan complexes rich in lignin after xylan is removed enzymatically.)

The same overall pattern of molecular degradation by enzyme incubation is observed with all xylan preparations. These observations are consistent with the understanding that xylan is degraded by xylanolytic enzymes to a mixture of monosaccharides via the intermediate oligosaccharidic mixture which is free of lignin. These results are also consistent with the view that xylan is covalently associated with lignin and that lignin protects xylan against enzymatic degradation, much like other substituents prohibit recognition of xylan by xylanolytic enzymes. Thirdly, these results are consistent with the view that totally xylan-free lignin components are water insoluble and that cleaving the lignin–xylan bonds liberates small amounts of low molecular weight substances (i.e., monolignols) which elute together with monosaccharides.

The corresponding incubation mixtures of (hydroxypropyl)xylan (HPX_C), DS 0.69, are shown in Figure 8. The same overall pattern emerges with an apparently greater stability of the oligosaccharide component. This suggests that HPX is indeed molecularly degraded to smaller molecular components and that saccharidic constituents, which fail to be recognized as degradable substances by the xylanase, accumulate in an undegradable pool of modified (presumably hydroxypropyl) xylan oligosaccharides. This result is expected, and it explains the apparent loss of biodegradability in view of a prevailing reduction of molecular weight.

These data demonstrate that chemically modified (hydroxypropyl)xylan continues to be biodegradable even

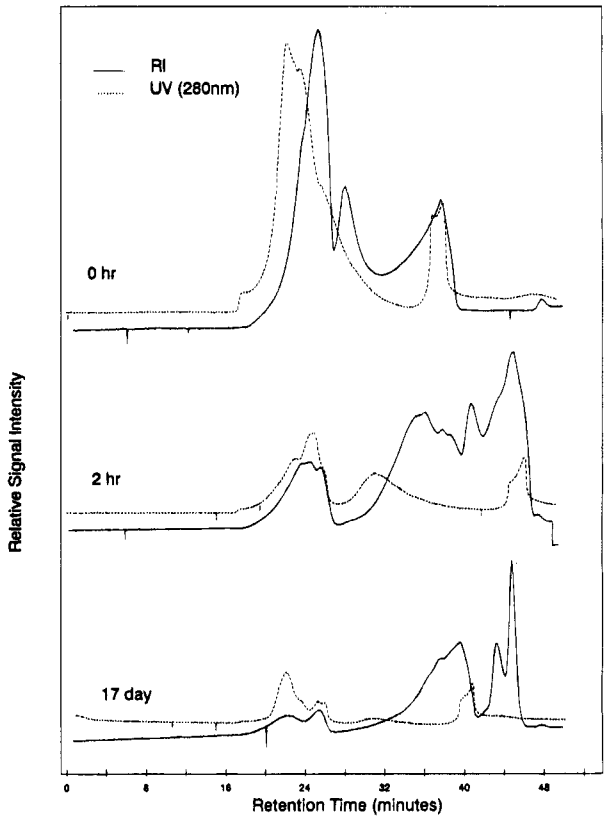

Figure 8. SEC and HPX_C (DS 0.5) in relation to enzyme hydrolysis time.

following chemical modification to extents which prevent a significant positive response to the xylose assay. The polymer seems to be degraded to a mixture of oligosaccharides instead of reducing monosaccharides.

Conclusions

1. Xylan represents the most readily biodegraded polymeric constituent of woody biomass. It produces xylose in virtually theoretical yields within 1–7 days of incubation with xylanase. There are no significant differences between different xylans isolated by different methods.

2. Xylanolytic enzyme biodegradability (XEB) as defined previously with corresponding cellulose derivatives declines in relation to degree of substitution. Whereas this decline was found to parallel that of cellulose esters for xylan acetate, (hydroxypropyl)xylan was found to follow a distinctly biphasic degradation pattern.

3. Cellulose and xylan acetates lost their EB more slowly than corresponding starch acetates as degree of substitution rises.

4. The surprising tolerance of HPX-derivatives with low DS to chemical substitution was explained with component-specific substitution. Reaction with propylene oxide occurs preferentially with non-xylose constituents of xylan (presumably phenolic lignin) before xylose units are reacted. This makes xylans with low degrees of substitution by propylene oxide especially attractive as biodegradable constituents for polymeric materials.

5. The degradation of xylans to monosaccharides by xylanolytic enzymes proceeds to completion only with unsubstituted xylans. Xylans carrying lignin or hydroxypropyl substituents fail to reach the monomeric level, and partially degraded substituted components accumulate in a pool of oligosaccharides.

Acknowledgment

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Literature Cited

(1) Meier, H. Chemical and Morphological Aspects of the Fine Structure of Wood. *Pure Appl. Chem.* **1962**, *5*, 37–52.
(2) Whistler, R. L. Xylan. *Adv. Carbohydr. Chem.* **1950**, *5*, 269–290.
(3) Rexen, F. Low-quality Forages Improve with Alkali Treatment. *Feedstuffs* **1979**, *51* (42), 33–34.
(4) Wilkie, K. C. B. The Hemicelluloses of Grasses and Cereals. *Adv. Carbohydr. Chem. Biochem.* **1979**, *36*, 215–264.
(5) Aspinall, G. O. Structural Chemistry of the Hemicelluloses. *Adv. Carbohydr. Chem.* **1959**, *14*, 429–468.
(6) Timell, T. E. Wood Hemicelluloses: Part I. *Adv. Carbohydr. Chem.* **1964**, *19*, 247–302.
(7) Kaar, W. E.; Jain, R. K.; Sealey, J. E.; Glasser, W. G. Xylan I. Isolation Options from Biomass on Pilot Scale. Manuscript in preparation.
(8) Jain, R. K.; Glasser, W. G. Xylan. II. Structure and Properties of Hydroxypropyl Xylan. Manuscript in preparation.
(9) Joseleau, J. P.; Comtat, J.; Ruel, K. Chemical Structure of Xylans and their Interaction in the Plant Cell Walls. In *Xylans and Xylanases*, Visser, J., et al., Eds.; Elsevier Science Publishers B.V.: 1992; pp 1–15.
(10) Voragen, A. G. J.; Gruppen, H.; Verbruggen, M. A.; Viëtor, R. J. Characterization of Cereal Arabinoxylans. In *Xylans and Xylanases*, Visser, J., et al., Eds.; Elsevier Science Publishers B.V.: 1992; pp 51–67.
(11) Viikari, J.; Kantelinen, A.; Buchert, J.; Puls, J. Enzymatic Accessibility of Xylans in Lignocellulosic Materials. *Appl. Microbiol. Biotechnol.* **1994**, *41*, 124–129.
(12) Ratto, M.; Mathrani, I. M.; Ahring, B.; Viikari, L. Application of Thermostable Xylanase of Dictyoglomus Sp. in Enzymatic Treatment of Kraft Pulps. *Appl. Microbiol. Biotechnol.* **1994**, *41*, 130–133.
(13) Buchanan, C. M.; Gardner, R. M.; Komarek, R. J. Aerobic Biodegradation of Cellulose Acetate. *J. Appl. Polym. Sci.* **1993**, *47*, 1709–1719.
(14) Parandoosh, S.; Hudson, S. M. The Acetylation and Enzymatic Degradation of Starch Films. *J. Appl. Polym. Sci.* **1993**, *48*, 787–791.
(15) Gu, J. D.; Eberiel, D.; McCarthy, S. P.; Gross, R. A. Cellulose Acetate Biodegradability Upon Exposure to Simulated Composting and Anaerobic Bioreactor Environments. *J. Environ. Polym. Degrad.* **1993**, *1* (2), 143–153.
(16) Narayan, R. Preparation of Bio-Based Polymers for Materials Applications. *Appl. Biochem. Biotechnol.* **1988**, *17*, 7–22.
(17) Penn, B. G.; Stannett, V. T.; Gilbert, R. D. Biodegradable Cellulose Graft Copolymers. I. Condensation-Type Graft Reactions. *J. Macromol. Sci.—Chem.* **1981**, *A16*, 473–479.
(18) Lynn, M. M.; Stannett, V. T.; Gilbert, R. D. Synthesis and Biodegradability of Amylose Block CoPolymers. *J. Polym. Sci. Polym. Chem.* **1980**, *18*, 1967–1977.
(19) Glasser, W. G.; McCartney, B. K.; Samaranayake, G. Cellulose Derivatives with Low Degree of Substitution. III. The Biodegradability of Cellulose Esters Using a Simple Enzyme Assay. *Biotechnol. Prog.* **1994**, *10*, 214–219.
(20) Samaranayake, G.; Glasser, W. G. Cellulose Derivatives with Low Degree of Substitution. I. Novel Acylation System. *Carbohydr. Polym.* **1993**, *22*, 1–7.
(21) Samaranayake, G.; Glasser, W. G. Cellulose Derivatives with Low Degree of Substitution. II. Analysis of Alkanoates. *Carbohydr. Polym.* **1993**, *22*, 79–86.
(22) Mitchell, D.; Grohmann, K.; Himmel, M. E. Effect of the Degree of Acetylation on the Enzymatic Digestion of Acetylated Xylans. *J. Wood Chem. Technol.* **1990**, *10*, 111–121.

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