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The impact of dilute sulfuric acid on the selectivity of xylooligomer depolymerization to monomers

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Abstract—The disappearance of xylose and xylooligosaccharides with degrees of polymerization (DP) ranging from 2 to 5 was followed at 160 °C with sulfuric acid added to adjust the pH from near neutral to 1.45, and the impact on the yields of lower DP xylooligomers and xylose monomer was determined. In addition, the experimental data for the disappearance of these xylooligomers was kinetically modeled assuming first-order reaction kinetics for xylose degradation and xylooligomer hydrolysis to evaluate how the pH affected the selectivity of monomer formation from xylooligomers and direct oligomer degradation to unknown products. The yield of xylose from xylooligomers increased appreciably with increasing acid concentration but decreased with increasing xylooligomer DP at a given acid concentration, resulting in more acid being required to realize the same xylose yields for higher DP species. For example, the maximum xylose yields were 49.6%, 28.0%, 13.2% and 3.2% for DP values of 2, 3, 4, and 5, respectively, at pH 4.75. Kinetic modeling revealed that all the xylooligomers disappeared at a higher rate compared to xylose monomer and the disappearance rate constant increased with DP at all pH. The kinetics for lower DP oligomers of 2 and 3 showed that these species directly degrade to unknown compounds in the absence of acid. On the other hand, higher oligomers of DP 4 and 5 exhibited negligible losses to degradation products at all pH. Therefore, only xylooligomers of DP 2 and 3 were found to directly degrade to undesired products in the absence of acid, but more work is needed to determine how higher DP species behave. This study also revealed that the source of water and the material used for the construction of the reactor impacted xylose degradation kinetics.

Keywords: Depolymerization; Degradation; Selectivity; Xylooligomers; Xylose; Yield

1. Introduction

Pretreatment is essential to realize the high yields vital to economic success for biological processing of lignocellulosic biomass to fuels (e.g., ethanol) and chemicals, and dilute acid treatments have been favored by many for near term applications.[1–5] At relatively low concentrations of 0.5–4.0% and temperatures of 140–220 °C, dilute acid hydrolyzes the hemicellulose in biomass to release primarily xylose monomers and some soluble xylooligosaccharides with high yields and produce highly digestible cellulose in the residual solids.[6,7] However, although sulfuric acid is low in cost itself, the overall process is still quite expensive due to the combined costs for the acid used, the base needed to neutralize it, the requirements for hydrolyzate conditioning, the expected exotic materials of construction, and the losses in sugars during pretreatment and hydrolyzate conditioning. Thus, it is valuable to better understand the role that the acid plays in hemicellulose hydrolysis if we are to reduce its use while still achieving high yields essential to economic success.

Numerous studies of hemicellulose hydrolysis using just water or with sulfuric and other acids give some indications of the effect of acid on hemicellulose sugar yields.[3,8,9] In addition, many models have been developed for reaction kinetics considering various possible routes to the formation and the degradation of

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oligomers and monomer in the complex hemicellulose hydrolysis pathway.[10,11] Extensive studies of xylose degradation in an acidic medium showed its loss to be consistent with a unimolecular reaction mechanism.[12–14] However, hemicellulose hydrolysis involves a complex insoluble solid breaking down to release numerous soluble oligomers, monomers, lignin decomposition products, and other compounds. Thus, following the concentrations of all the species involved is difficult, if not impossible, limiting insight into controlling mechanisms.

In an alternate approach, following the reaction of soluble oligomers of known DP provides a model system that allows us to accurately track the loss and the formation of oligomers and monomers that can then be applied to understand hemicellulose hydrolysis pathways. One such study focused on determining how the reactivity of xylooligomer bonds changed with the degree of polymerization (DP) for the hydrolysis of low DP xylooligomers from xylobiose to xylopentose as model compounds with dilute sulfuric acid (0.025, 0.05, and 0.25 M) at low temperatures (80 and 100 °C).[15] Under these conditions, xylose degradation was negligible, and it was determined that the xylosidic linkage adjacent to the non-reducing terminal group reacted almost 1.8 times faster than interior links.[15] Research by our group with low DP soluble oligomers at a higher temperature of 200 °C where xylose degradation is important showed that the selectivity of xylose monomer formation from oligomers was low in the absence of added sulfuric acid and that the low yields of xylose monomer could not be adequately explained without the incorporation of reactions for direct degradation of oligomers prior to monomer formation.[16] More recent research on xylobiose degradation revealed that sulfuric acid dramatically increased the ratio of the depolymerization rate constant to the overall disappearance rate constant to close to one, significantly enhancing the selectivity of xylose formation. In addition, the optimum acid concentration to maximize xylose monomer recovery during acid hydrolysis of xylan rich hemicellulose was determined to be between pH 2 and 3.[17]

The results for xylooligomer reactions reported to date suggest that acid enhances the selectivity of xylooligomer breakdown to monomers through either dramatically reducing the role of side reactions that directly degrade oligomers or by significantly speeding the reactions to monomers relative to these direct degradation reactions. However, because published results are limited to a few temperatures, acid concentrations, and oligomer chain lengths, it is difficult to draw broad conclusions. Thus, in this study, xylose, xylobiose, xylotriose, xylotetrose, and xylopentose decomposition were followed at 160 °C with sulfuric acid added to cover a pH range from near neutral to 1.45, and the selectivity and the yields of xylose formation from xylooligomers were determined. In addition, xylooligomer degradation was modeled with and without incorporating a parallel degradation pathway, and the influence of reactor materials and water source on xylose degradation was determined based on the concerns that the materials of construction could impact the results.[18]

2. Materials and methods

2.1. Materials

Xylobiose (lot 10702), xylotriose (lot 51003), xylotetrose (lot 10804), and xylopentose (lot 10205) were obtained from Megazyme International Ireland, Limited (Bray Business Park, Bray, Co. Wicklow, Republic of Ireland). Monomeric xylose was obtained from Fisher Biotech (lot 006640, Fair Lawn, NJ, 07410).

2.2. Analytical procedures

All chemical analyses of xylooligomers and xylose were performed using a Waters Alliance HPLC system (Model 2695, Waters Corporation, Milford, MA) employing Aminex-HP 42 A and HPX-87P columns (Bio-Rad, Hercules, CA) for oligomer and xylose quantification, respectively, equipped with appropriate accessories. Samples were processed at an eluent flow rate of 0.60 mL/min using a refractive index (RI) detector (model 2414, Waters Corporation, Milford, MA).

2.3. Parameter estimation

Parameter estimation was performed by fitting the data by least-square method using the Excel Solver Routine and Berkeley Madonna software from the University of California, Berkeley.

2.4. Experimental system

Xylooligomer degradation experiments were performed using clear glass crimp vials with flat bottoms and aluminum crimp tops with TFE/silicone liners (7 mm × 40 mm, 800 μL, Alltech Associates Inc; Deerfield, IL) into which 500–600 μL of xylooligomer solution (0.25–1 mg/mL) was pipetted and sealed using a quarter-inch crimping tool. To prevent rupturing, the TFE/silicone septa glass reactor caps at high reaction pressures, a disc punched from a 25 μm thick brass sheet was inserted between the cap and the septa.[17] In addition, 316 stainless steel tubes of 0.5 in. OD, 5 in. length, and 0.035 in. wall thickness from Swagelok (Bangor, ME) were employed as reactors to study the effect of materials of construction and xylose concentration on xylose degradation. In this case, 7 mL of xylose solution (1–10 mg/mL) was pipetted into these tubes which were

made leak proof by wrapping their threads with Teflon tape and applying vacuum grease before sealing with a bolt fitted with a Teflon stopper. Both the glass vials and stainless steel reactor tubes were heated to reaction temperature by immersion in a 22.8 cm ID × 35 cm deep 4 kW model SBL-2D fluidized sand bath (Techne Corporation, Princeton, NJ). To hold the reactors in place, promote uniform heating, and simplify removal, a screen tied to a 3.5 cm diameter iron rod was immersed about half way into the depth of the sand bath. Heating times of 30 and 60 s for the glass and steel reactors, respectively, were adequate to reach reaction temperatures as determined by a thermocouple. The reaction vessels were removed from the sand bath at different reaction times with tongs and quenched in a cold water tub held at room temperature. To reduce breakage and loss of contents, the glass vials were air cooled for 10–20 s before immersing them in the tub. The contents of the reactors were then removed with a 1 mL syringe (Dickinson and Company, Franklin Lakes, NJ) and filtered through 0.2 μm nylon filter vials (Alltech Associates Inc., Deerfield, IL). About 100–150 mg of calcium carbonate was added to the filter vials to neutralize acidic samples before they were centrifuged using Eppendorf Centrifuge MiniSpin at 13,400 rpm for 2 min. Centrifugates were then pipetted into 500 μL polyethylene HPLC vials.

3. Results

3.1. Xylose degradation

All experiments were run at 160 °C, the temperature that is found to often give the highest xylose yields for dilute acid hydrolysis of hemicellulose.[4,19,20] First, we evaluated how the choice of reactor material (stainless steel or glass) influenced degradation kinetics. As shown in Figure 1a, the rate of xylose degradation in glass vials appeared to decrease somewhat with increasing xylose concentration. On the other hand, as shown in Figure 1b, little effect of xylose concentration was evident over the same concentration range in stainless steel reactors.

Table 1 shows the xylose degradation constants estimated by fitting the experimental data using Excel Solver assuming first-order reaction kinetics, shown below, for three different sugar concentrations and two different pH values (neutral and 2.75).

$$\text{Xylose} \xrightarrow{k_d} \text{Degradation products}$$

$$\frac{dX_1}{dt} = -k_d * X_1$$

$$\frac{X_1}{X_{1_0}} = \exp(-k_d * t)$$

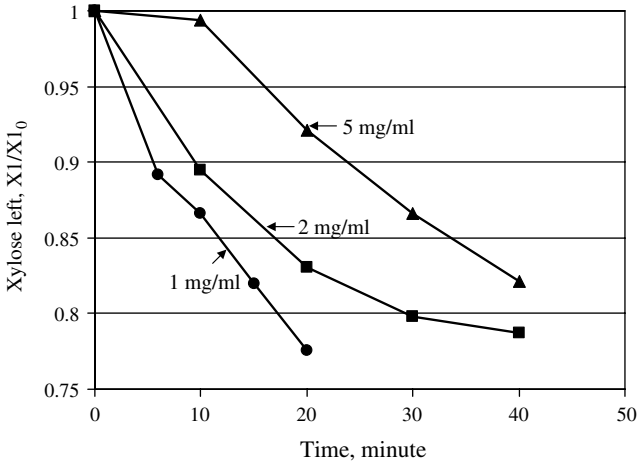


Figure 1a. Fraction of initial xylose left in solution over reaction time at 160 °C for reactors made from glass vials.

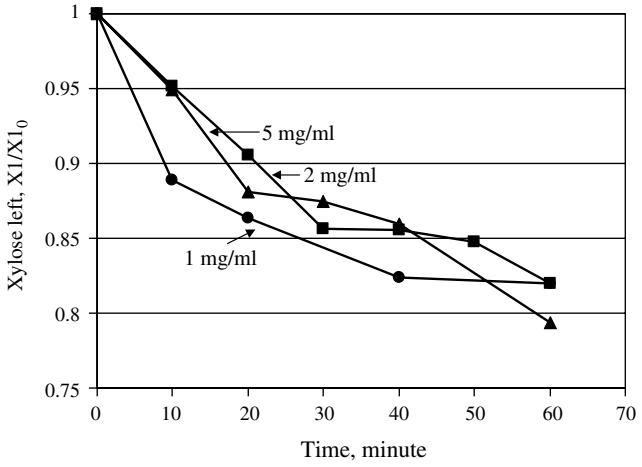


Figure 1b. Fraction of initial xylose left in solution over reaction time at 160 °C for reactors made from stainless steel.

Table 1. Xylose degradation rate constants (k_d) at different xylose concentrations and pH

Xylose concn (mg/ml)	pH neutral		pH 2.75	
	Steel tube	Glass vial	Steel tube	Glass vial
1	0.0044	0.013	0.0085	0.0057
2	0.0050	0.0059	0.0058	0.0052
5	0.0051	0.0042	0.0042	0.0036
Average	0.0048	0.0077	0.0062	0.0048

At neutral pH, the degradation rate constants for xylose, Table 1, were higher for glass vials than steel reactors, and the variation in constants with xylose concentration was more apparent as well. However, at lower pH this variation in glass vials was less prominent, and the rate constants for xylose degradation were

higher in steel reactors than glass vials. Based on these results, we decided to use glass vials for all subsequent experiments because we thought that steel might catalyze degradation. Furthermore, to avoid the impact of water quality on the results, deionized water and water adjusted to different pH's with sulfuric acid was kept refrigerated for all subsequent experiments.

3.2. Xylooligomers disappearance

To see the impact of dilute sulfuric acid on xylooligomer degradation, experiments were performed in glass vials over a pH range from neutral down to 1.45 at 160 °C, the pH was adjusted with sulfuric acid. At higher pH and for a given time, oligomers with DP of 4 and 5 gave lower xylose yields than xylobiose or xylotriose as illustrated in Figure 2 for a pH of 4.75. However, the yield and the selectivity of xylose formation increased appreciably as acid was added for all the oligomers studied as shown in Figure 3. However, the degradation of xylose becomes appreciable if too much acid was added to the reaction media.[17] Furthermore, yields seemed to drop with increasing DP at any acid concentration for these five oligomers, as also shown in Figure 3.

3.3. Kinetic study

3.3.1. Xylose monomer degradation. As discussed in Section 3.2, the rate of xylose degradation seemed to depend inversely on its concentration at higher pH values, especially in glass vials. On the other hand, most previous kinetic studies were performed at highly acidic conditions where these effects were not seen, leading to the conclusion that xylose degradation is first order in xylose concentration and depends only on acid concentration.[13,21,22] Yet, appreciable differences have been observed for degradation studies between batch and flow systems.[12–14] In the current study, the degradation rate constants did not change appreciably at xylose concentrations of 2 mg/mL (13.3 mM) until a pH of 2.75 was reached and then became higher, consistent with trends observed in a previous study at our laboratory as shown in Table 2.[17] However, compared to the studies performed at our laboratory, the ratio of degradation constants, calculated using models reported in the literature, was quite high over the pH range, as Table 2 shows. In addition, the ratio of degradation constants at pH 1.45 to those at neutral condition was very high, as predicted by models in the literature. Figure 4 shows the variation of xylose degradation constants with pH and comparisons with the literature values.

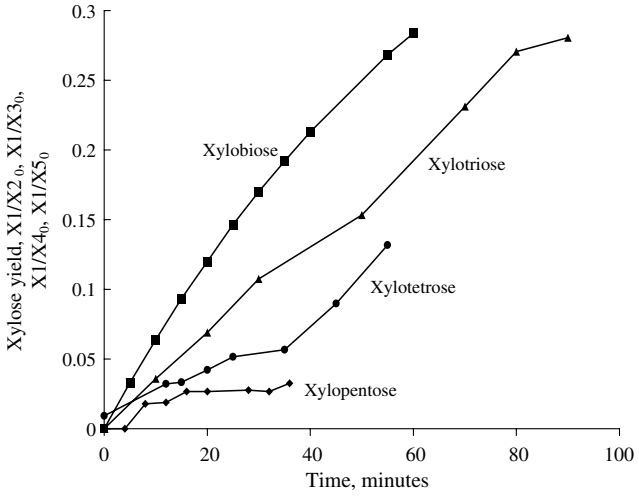


Figure 2. Fractional xylose yields from xylobiose up to xylopentose for reaction at 160 °C and a pH of 4.75.

3.3.2. Xylooligomer disappearance. Overall, disappearance rate constants for xylooligomers were determined using the Solver Routine in Excel to minimize the differences between experimental and predicted residual concentrations of xylose and xylooligomers for the following kinetic expressions for all DP oligomers:

$$\frac{dX_n}{dt} = -k_n * X_n$$

$$\frac{X_n}{X_{n_0}} = \exp(-k_n * t)$$

in which $n = 1$–5; and k_n = rate constants to describe the overall disappearance of each xylooligomer. The rate constants for overall disappearance of different xylooligomers increased with DP as shown in Figure 5. The ratios of xylooligomer disappearance rate constants to xylose degradation rate constant were greater than 2 for all the oligomers, and this ratio increased with decreasing pH, as shown in Table 3, contrary to the prediction by the Kamiyama and Sakai model[15] (developed for lower temperatures and higher acid molar concentration) shown in Table 3 in parenthesis.

3.3.3. Xylooligomer hydrolysis versus degradation. The disappearance rate constants account for the loss of the oligomers but not the selectivity of xylose formation versus loss to degradation products. As discussed earlier, previous research by our group showed that the selectivity of xylose formation from oligomers was low near neutral pH values. In addition, calculated rate constants based on following just xylooligomer degradation could not adequately describe the formation of xylose monomer or degradation products from soluble xylooligomers based on first-order degradation of xylooligomers in series.[16] From this observation, it was hypothesized that direct degradation of oligomers also occurs in parallel with the formation of xylose and lower DP oligomers as follows:

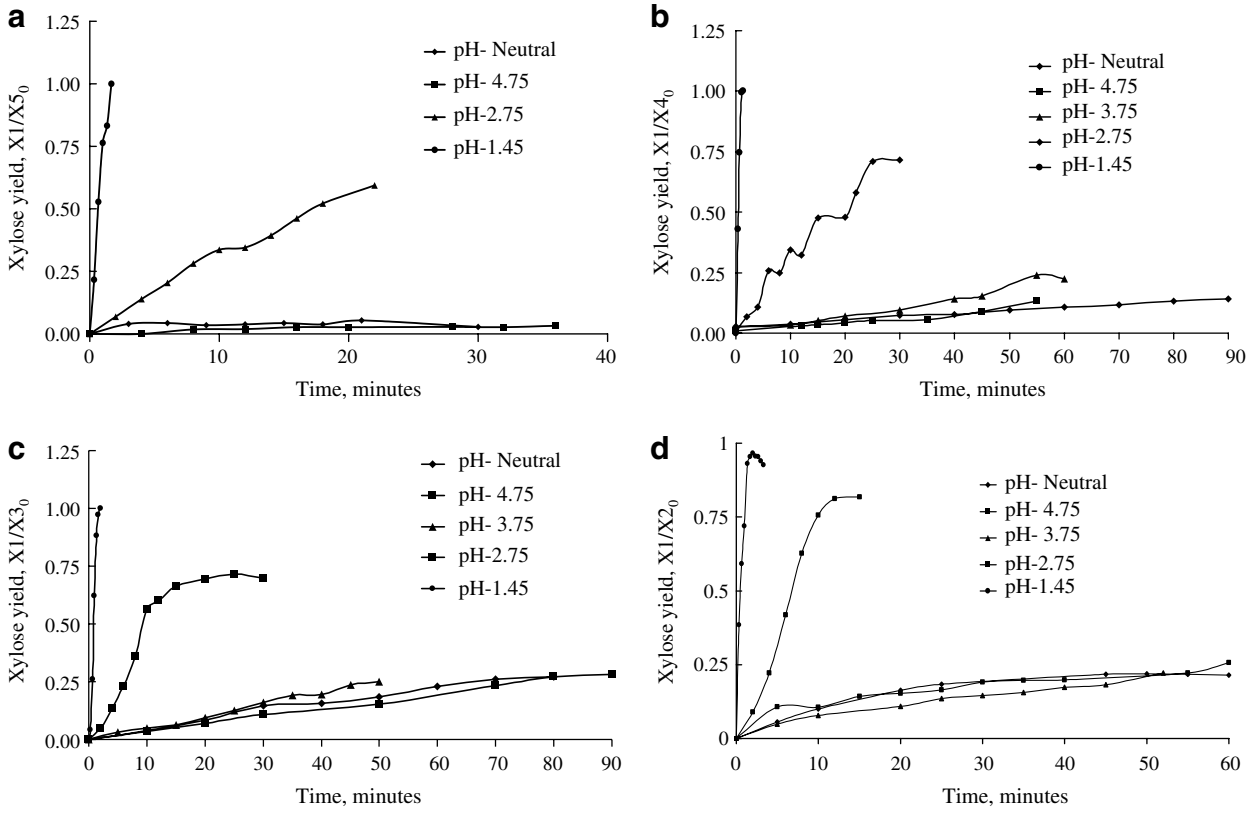
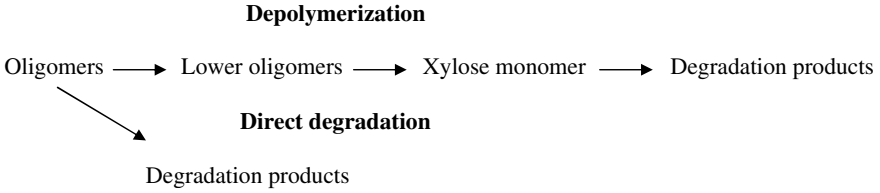


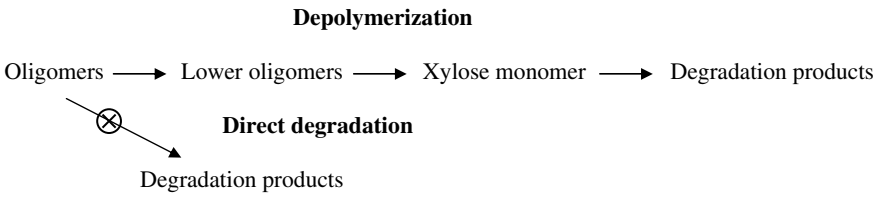
Figure 3. Fractional xylose yields at 160 °C and various pH values for (a) xylopentose, (b) xylotetrose, (c) xylotriose, and (d) xylobiose.

Without acid:



However, it was assumed that direct degradation of oligomers is reduced or eliminated by adding acid either by accelerating hydrolysis reactions or reducing degradation as follows:

With acid:



Consistent with this hypothesis, another study by our group showed that sulfuric acid dramatically increased the xylobiose depolymerization rate constant compared to the overall disappearance rate constant, shifting the ratio closer to one as the pH dropped.[17] Thus, we kinetically modeled xylooligomer disappearance based on this mechanism assuming the degradation of oligomers to shorter chain oligomers and degradation products followed first-order kinetics.

3.3.4. Xylobiose hydrolysis. As shown in Figure 6, experimental and modeled xylose concentrations, using the k_2 determined for xylobiose, were not the same if only the reaction of xylobiose to xylose was considered, while incorporation of direct degradation of xylobiose

Table 2. Xylose degradation rate constants at different pH and their comparison to the literature values[a]

n	pH	k_d Ref. 21 (ratio k_{dn}/k_{dn-1})	k_d Ref. 12 (ratio k_{dn}/k_{dn-1})	k_d Ref. 16 (ratio k_{dn}/k_{dn-1})	k_d Current study (ratio k_{dn}/k_{dn-1})
1	Neutral	8.47E−06	3.90E−08	1.88E−03	5.9E−03
2	4.75	1.26E−04	1.91E−06	1.88E−03	6.6E−03
		(14.9)	(50)	(1)	(1.1)
3	3.75	6.18E−04	1.91E−05	1.89E−03	5.2E−03
		(4.9)	(10)	(1)	(0.8)
4	2.75	3.0E−03	1.91E−04	2.0E−03	5.5E−03
		(4.9)	(10)	(1.06)	(1.06)
5	1.45	2.39E−02	3.80E−03	4.34E−03	9.8E−03
		(7.9)	(20)	(2.16)	(1.80)
$k_{d\,1.45}/k_{d\,neutral}$		2.8E2	10 E5	2.30	1.60

[a] Values are degradation rate constants reported by references noted, while there in ( ) are ratio of the value of k_d shown to that for previous at higher pH value.

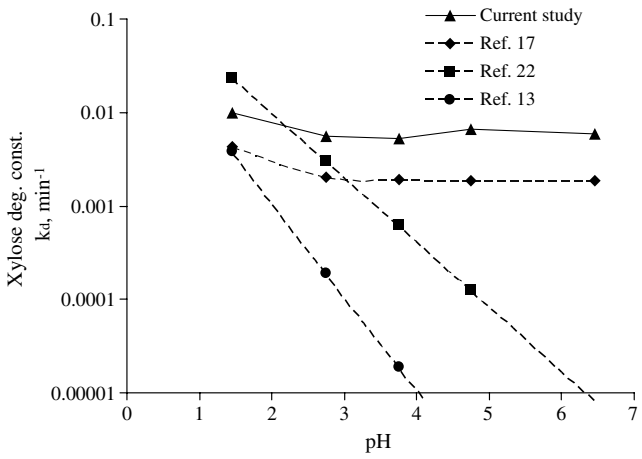
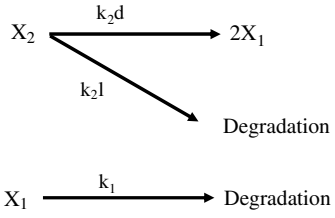


Figure 4. Xylose degradation rate constants and comparison to the literature values at different pH levels and 160 °C.

into the model resulted in predicted concentrations based on disappearance rate constants that closely agreed with observed xylose concentrations. As shown in Table 4 for xylobiose, the ratio of depolymerization constants to the overall disappearance constant was found to increase with acid concentration and approach one at lower pH, for the following kinetic expressions:



$$\frac{dX_2}{dt} = -(k_2d + k_2l) * X_2$$

$$\frac{dX_1}{dt} = k_2d * X_2 - k_1 * X_1$$

in which k_2l is the xylobiose loss constant; k_2d, the xylobiose depolymerization constant; $k_2 = (k_2l + k_2d)$, the xylobiose overall disappearance constant; and k_1, the xylose degradation constant of 13.3 mM xylose.

Parameters were obtained by minimizing the difference between experimentally measured and predicted concentrations of xylose monomer and oligomers using Excel Solver. As Table 4 shows, the rates of both xylobiose depolymerization to xylose monomer and degradation to unknown products increased with acid concentration. However, the ratio of depolymerization rate constants to degradation rate constants increased and the ratio of degradation constants to overall disappearance constant decreased with acid concentration.

3.3.5. Xylotriose hydrolysis. The experimental and modeled xylose and xylobiose concentrations were not the same if only the reaction of xylotriose to xylobiose and the reaction of xylobiose to xylose were considered, as shown in Figure 7a, while incorporation of direct degradation of xylotriose and xylobiose into the model resulted in predicted concentrations based on the disappearance constants that closely agreed with those for the observed xylobiose and xylose concentrations, as Figure 7b shows. For parameter estimation, the least-square method of the Berkeley Madonna software was used with the reaction modeled as xylotriose breaking down to xylobiose and xylose and degradation products in parallel reactions. In addition, xylobiose decomposed to xylose and degradation products, and xylose further decomposed to degradation products. The kinetic parameters determined previously were used to describe xylotriose hydrolysis with $k_1 = 0.0059$, $k_2d = 0.0059$, $k_2l = 0.0062$, and $k_3 = 0.024\ min^{-1}$ at neutral pH. The stoichiometric equations and corresponding rate laws for xylotriose hydrolysis are as follows:

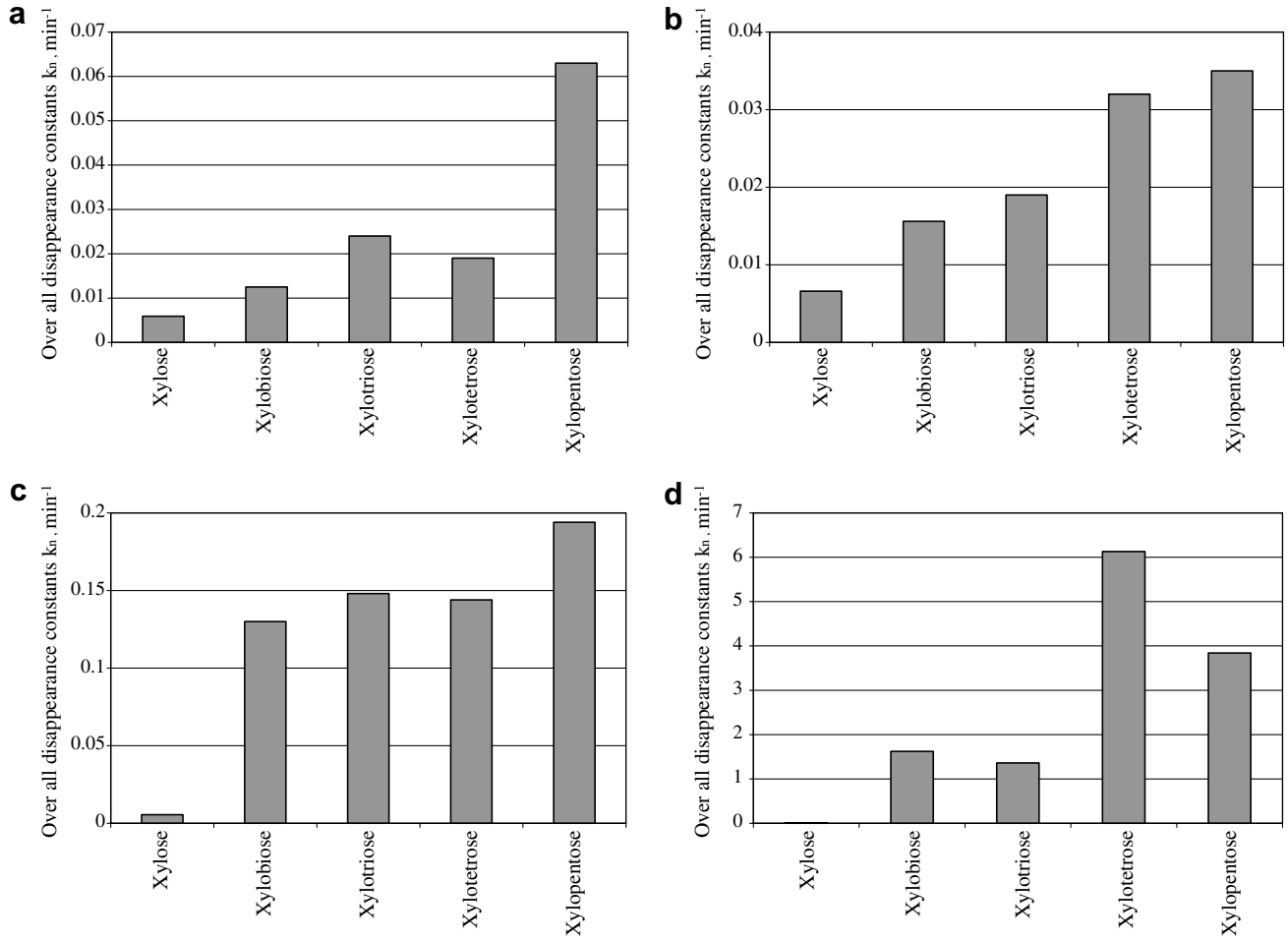
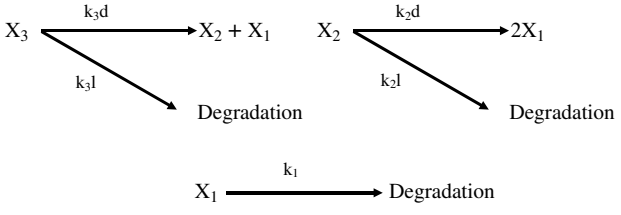


Figure 5. Overall disappearance rate constants for xylose and its oligomers up to xylopentose at 160 °C and pH (a) neutral, (b) 4.75, (c) 2.75, and (d) 1.45.



$$\frac{dX_3}{dt} = -(k_3d + k_3l) * X_3$$

$$\frac{dX_2}{dt} = 2/3 * k_3d * X_3 - (k_2d + k_2l) * X_2$$

$$\frac{dX_1}{dt} = 1/3 * k_3d * X_3 + k_2d * X_2 - k_1 * X_1$$

in which k_3l is the xylotriose loss constant; k_3d, the xylotriose depolymerization constant; and $k_3 = (k_3d + k_3l)$, the xylotriose disappearance rate constant.

Modeling of xylotriose hydrolysis and parameter estimation were performed for experimental data at other pH values as well, with the model parameters and statistical coefficients R^2 shown in Table 5. The ratio of k_3d to k_3 and k_3d to k_3l were found to increase with increasing acid concentration. In addition, losses were observed to decline with acid concentration.

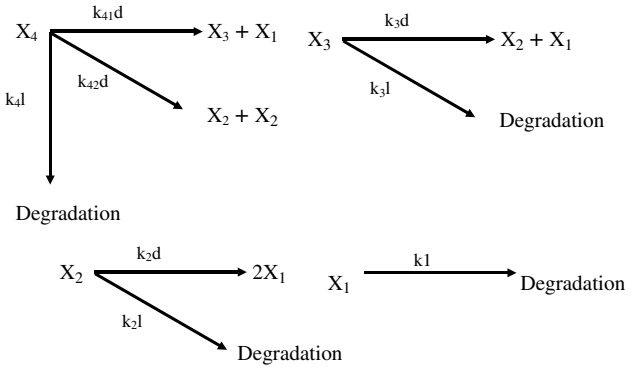
Table 3. Ratio of overall degradation rate constant (k_n) of oligomer to xylose degradation rate constant (k_1) at various pH[a]

pH	X_2	X_3	X_4	X_5
Neutral	2.1 (2.3)[a]	4.1 (3.4)	3.1 (4.7)	10.7 (6.0)
4.75	2.4 (2.2)	2.9 (3.5)	4.8 (4.7)	5.3 (5.9)
2.75	23.6 (2.3)	26.9 (3.6)	26.2 (4.8)	35.3 (5.9)
1.45	165.3 (2.2)	138.8 (3.5)	625.5 (4.7)	391.8 (5.9)

[a] Values in parenthesis are ratios for degradation rate constants predicted by the Kamiyama and Sakai model.[15]

3.3.6. Xylotetrose hydrolysis. Assuming first-order reactions, xylotetrose could depolymerize to shorter oligomers by two possible mechanisms and also decompose directly to degradation products. Xylotetrose disappearance could lead to the formation of xylotriose and xylose and two xylobiose molecules simultaneously. Further, xylotriose, xylobiose, and xylose could all

degrade as per the mechanisms described earlier. The parameters determined at different pH values for xylotetrose, xylotriose, and xylobiose disappearance and xylose degradation were used in the xylotetrose degradation model, and the new parameters estimated by the least-square method. Thus $k_4 = 0.1438$, $k_3 = 0.148$, $k_3d = 0.1394$, $k_3l = 9.0\text{E}{-3}$, $k_2 = 0.128$, $k_2d = 0.116$, $k_2l = 0.012$, and $k_1 = 0.0066\ \text{min}^{-1}$ for a pH value of 2.75. The kinetic expressions and corresponding rate laws are shown below



$$\frac{dX_4}{dt} = -(k_{41}d + k_{42}d + k_4l) * X_4$$

$$\frac{dX_3}{dt} = 3/4k_{41}d * X_4 - (k_3d + k_3l) * X_3$$

$$\frac{dX_2}{dt} = k_{42}d * X_4 + 2/3 * k_3d * X_3 - (k_2d + k_2l) * X_2$$

$$\frac{dX_1}{dt} = 1/4k_{41}d * X_4 + 1/3 * k_3d * X_3 + k_2d * X_2 - k_1 * X_1$$

in which $k_{41}d$ and $k_{42}d$ are the xylotetrose depolymerization constants; k_4l is the xylotetrose loss const.; and $k_4 = (k_{41}d + k_{42}d + k_4l)$ is the xylotetrose overall disappearance constant.

The experimental and predicted concentrations are shown in Figure 8 at 3.75 pH. The values of the parameters and the residual sum of square (RSS) determined at different pH values are shown in Table 6. The sum of the parameters $k_{41}d$, $k_{42}d$, and k_4l was constrained to be equal to k_4, and the difference between oligomers and xylose concentration was minimized using the least-square method. The experimental and predicted concentration of oligomers and xylose monomer were

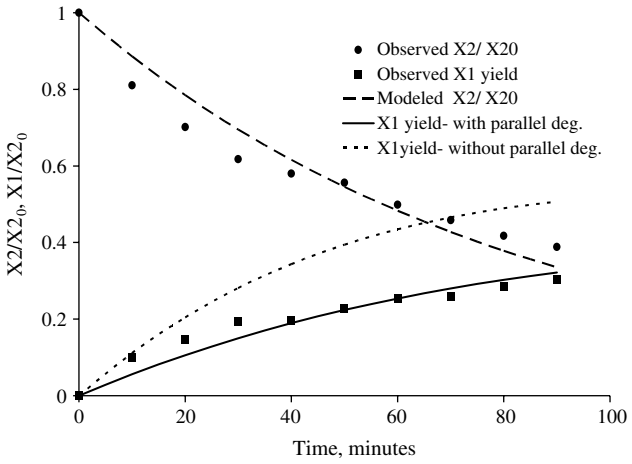


Figure 6. Xylobiose disappearance and xylose formation at 160 °C and neutral pH.

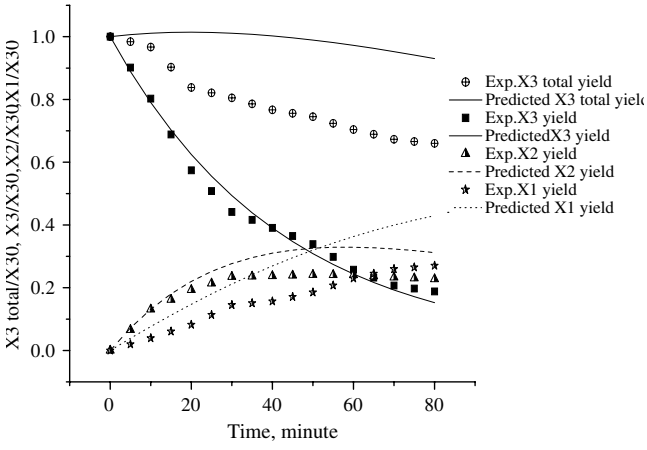


Figure 7a. Xylotriose degradation at 160 °C and neutral pH without incorporating direct losses of xylobiose and xylotriose.

similar, except at pH 4.75, with and without considering direct loss of xylotetrose. Thus, direct loss of xylotetrose at higher pH was unimportant.

3.3.7. Xylopentose hydrolysis. Xylopentose depolymerization could take place via three routes assuming all the reactions are of first order: xylopentose may break down to xylotetrose oligomer and xylose monomer; xylotriose and xylobiose oligomers; and directly to degradation products, as shown below. The parameters estimated individually for lower oligomers and xylose monomer

Table 4. Xylobiose overall disappearance (k_2), depolymerization (k_2d) rate, and loss (k_2l) rate constants and their ratios at different pH values

pH	k_2	k_2d	k_2l[a]	k_2d/k_2l	k_2d/k_2	k_2l/k_2
Neutral	0.0121	0.0059	0.0062	0.95	0.49	0.51
4.75	0.0156	0.0068	0.0088	0.77	0.44	0.56
3.75	0.0092	0.0056	0.0036	1.56	0.61	0.39
2.75	0.128	0.116	0.012	9.67	0.91	0.094
1.45	1.62	1.5	0.12	12.5	0.93	0.074

[a] $k_2l = k_2 - k_2d$.

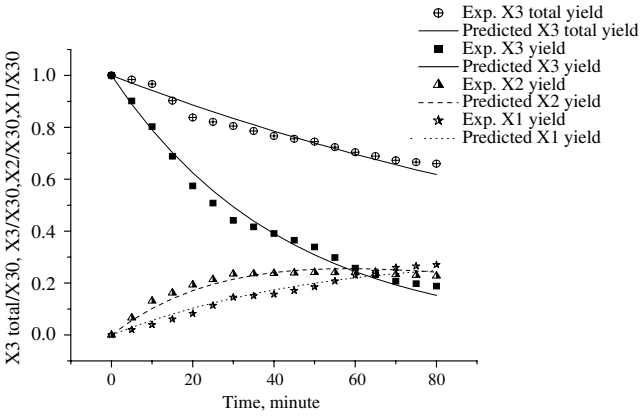
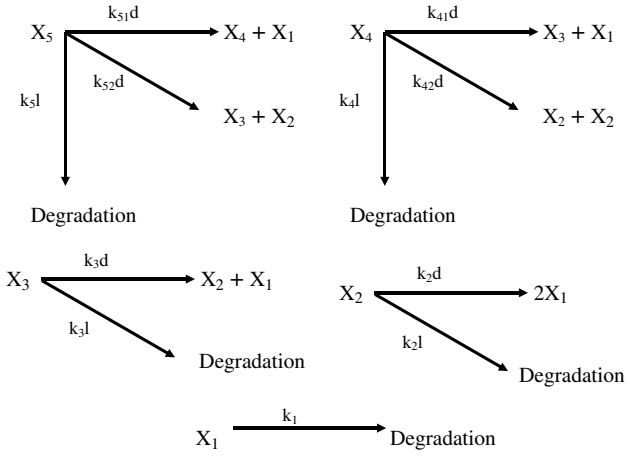


Figure 7b. Xylotriose degradation at 160 °C and neutral pH including the direct losses of xylobiose and xylotriose.

Table 5. Depolymerization (k_3d), loss (k_3l), and overall disappearance (k_3) rate constants and their ratios for xylotriose at different pH values

Parameter	pH				
	Neutral	4.75	3.75	2.75	1.45
k_3	0.024	0.020	0.029	0.148	1.362
k_3d	0.018	0.017	0.020	0.1394	1.362
k_3l	0.005	0.003	0.009	0.009	0.00001
k_3d/k_3	0.77	0.87	0.68	0.941	0.99
k_3l/k_3	0.22	0.13	0.32	0.059	1E−5
k_3d/k_3l	3.46	6.84	2.13	15.9	
RSS	0.0632	0.066	0.065	0.162	0.3244

were used in modeling xylopentose hydrolysis, for example, at neutral pH $k_5 = 0.0629$, $k_4 = 0.0180$, $k_{41}d = 0.0148$, $k_{42}d = 0.0032$, $k_3 = 0.024$, $k_3d = 0.0183$, $k_2 = 0.0121$, $k_2d = 0.0059$, and $k_1 = 0.0059\ \text{min}^{-1}$. Three kinetic parameters (k_{51}d, k_{52}d, and k_5l) known to account for xylopentose degradation were estimated by constraining their sums to equal the overall disappearance rate constant (k_5) and minimizing the difference between experimental and modeled concentrations of oligomers and xylose by using the least-square method of Berkeley Madonna software for the data at all pH values. The stoichiometric relationships and rate laws are as follows:



$$\frac{dX_5}{dt} = -(k_{51}d + k_{52}d + k_5l) * X_5$$

$$\frac{dX_4}{dt} = 4/5k_{51}d * X_5 - (k_{41}d + k_{42}d + k_4l) * X_4$$

$$\frac{dX_3}{dt} = 3/5k_{52}d * X_5 + 3/4k_{41}d * X_4 - (k_3d + k_3l) * X_3$$

$$\frac{dX_2}{dt} = 2/5k_{52}d * X_5 + k_{42}d * X_4 + 2/3 * k_3d * X_3 - (k_2d + k_2l) * X_2$$

$$\frac{dX_1}{dt} = 1/5k_{51}d * X_5 + 1/4k_{41}d * X_4 + 1/3 * k_3d * X_3 + k_2d * X_2 - k_1 * X_1$$

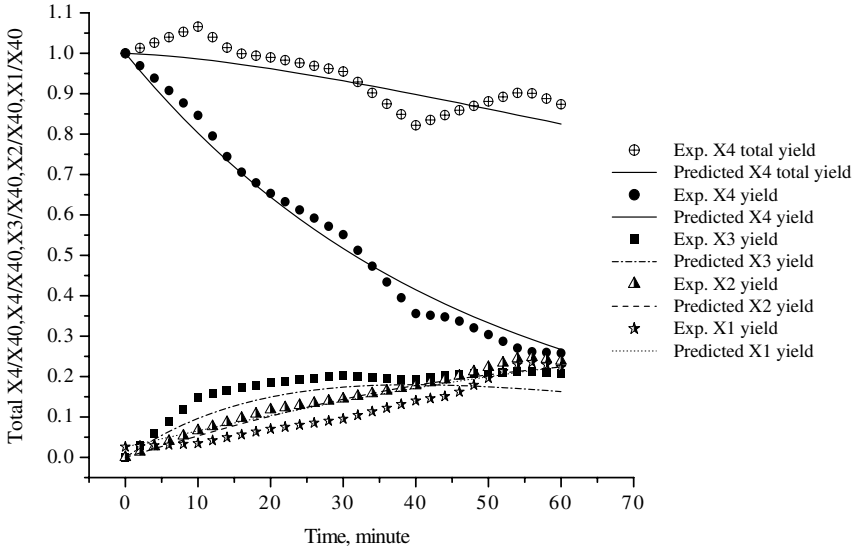


Figure 8. Xylotetrose disappearance at 160 °C and pH 3.75.

Table 6. Depolymerization ($k_{41}d$ and $k_{42}d$), loss (k_4l), and overall disappearance (k_4) rate constants and their ratios for xylotetrose at different pH values

Parameter	pH				
	Neutral	4.75	3.75	2.75	1.45
k_4	0.0184	0.0323	0.022	0.1438	6.13
$k_{41}d$	0.0148	0.0166	0.016562	0.080454	1.7800
$k_{42}d$	0.0032	0.0051	0.0054	0.0633	4.3510
k_4l	0.00032	0.003	0.009	0.009	0.00001
k_4d/k_4	1.0	0.67	1.0	1.0	1.0
k_4l/k_4	0.00	0.33	0.00	0.00	0.00
RSS	0.136	0.172	0.18	0.426	0.55

Table 7. Depolymerization ($k_{51}d$ and $k_{52}d$), loss (k_5l), and overall disappearance (k_5) rate constants and their ratios for xylopentose at different pH values

Parameter	pH			
	Neutral	4.75	2.75	1.45
k_5	0.0629	0.0350	0.19465	3.8386
$k_{51}d$	0.04186	0.0280	0.110791	1.9140
$k_{52}d$	0.0210	0.0071	0.0839	1.9246
k_5l	0.0000	0.0000	0.0000	0.0000
k_5d/k_5	1.0	1.0	1.0	1.0
RSS	0.334	0.649	0.85	0.55

in which $k_{51}d$ and $k_{52}d$ are the xylopentose depolymerization constants; k_5l is the xylopentose loss rate constant; $k_5 = (k_{51}d + k_{52}d + k_5l)$ is the xylopentose overall disappearance constant.

The modeled and experimental concentration values were found to agree closely, as shown by Figure 9 at pH 4.75. However, direct degradation of xylopentose was not evident with the degradation rate constant found to be being equal to zero at all pH values, as shown in Table 7.

4. Discussion

This study showed that xylose degradation depends on xylose concentration, especially in glass vial reactors at low concentrations of sulfuric acid. This finding will be important for pretreatments such as steam explosion, controlled pH, flowthrough, and dilute acid, where little if any acid is added.[23–25] However, based primarily on studies conducted at very acidic conditions in stainless steel reactors,[11,12,18] xylose degradation was shown to follow first-order kinetics, with the rate depending just on the amount of acid and temperature. The observation in this study is in agreement with Laboratory Analytical Procedure (LAP-02, paragraph-10.1.7, NREL, USA) and research done at NREL (Dr. Bonnie Hames, personal communication) that monomeric sugar degradation depends on their concentration and may be affected by aqueous media as well.

The observation that xylose degradation rate constants at lower pH are higher in steel reactors compared to glass vials could result from the combined catalytic effects of metal and acid. We have also observed that xylose degradation rate constants can vary significantly and sometimes by several fold when repeated on different days possibly due to changes in the amount of minerals and salts in the water.[26]

The selectivity of monomeric xylose formation from oligomers increased with acid concentration. However, this effect is observed until xylose concentrations build to the point that its degradation becomes significant, not determined in this study.[17] In addition, the overall selectivity of oligomer breakdown to monomeric xylose decreased with increasing DP at a given acid concentration.

The rate constants for xylooligomer disappearance increased with DP over the range of oligomers studied

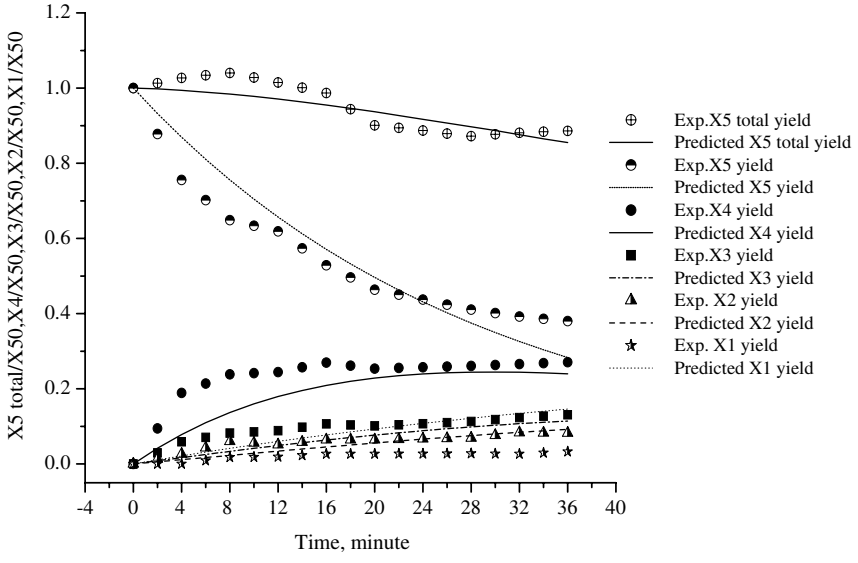


Figure 9. Xylopentose disappearance at 160 °C and pH 4.75.

from xylobiose to xylopentose. Xylooligomers degraded faster than monomeric xylose at all the pH values and the ratio of xylooligomer disappearance rate constants to xylose degradation rate constant increased with decreasing pH. On the other hand, in a study of xylooligomer degradation at 43 °C using alkaline medium, Sartori et al. showed the rate constants for xylooligomers degradation increased compared to the monosaccharide as the DP increased from 2 to 3 and then decreased slightly for oligomers studied of DP 4.[21]

The hypothesis[16,17] that oligomers directly degrade at low acid concentration was also tested for a range of oligomers from xylobiose to xylopentose. Observations with xylobiose and xylotriose strongly suggest that direct degradation of oligomers occurs along with depolymerization to lower oligomers and monomer and is minimized by the presence of acid. However, xylotetrose and xylopentose modeling contradicts the hypothesis, and direct degradation of these oligomers at low and high acid concentrations was found to be negligible. Thus, it may be interpreted that the direct degradation may only occur with lower oligomers such as xylobiose and xylotriose. Consistent with the finding in this research, Garrote et al. assumed for kinetic modeling of corn cob autohydrolysis that xylan breaks down to higher DP oligomers and that it then forms lower DP oligomers, which can degrade directly to furfural and form xylose.[27] However, this was an empirical fit in that the length of xyloligomers that directly degrade was not defined. Therefore, it can be assumed that oligomers of DP 2–3 are decomposed directly to degradation products at high pH while DP 4 and 5 do not. However, further validation is needed with higher oligomers to determine if this trend is followed by higher DP oligomers.

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