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The Development of Cement and Concrete Additive

Based on Xylonic Acid Derived Via Bioconversion of Xylose‡

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Abstract

The present work attempted to utilize xylose by converting it to an aldonic acid. In the present study, xylose was converted to xylonic acid by using commercial glucose oxidase enzyme, palladium catalysis, and microbial bioconversion. The enzyme conversion was successfully done using a commercial glucose oxidase. The microbial conversion with *Gluconobactor oxydans* proceeded even with the presence of a large amount of lignosulfonate. Thus obtained xylonic acid products were evaluated as a cement dispersing agent in cement and concrete tests. It was found that xylonic acid is approximately twice as effective as lignosulfonate. Xylonic acid can be effectively utilized in concrete water reducer application.

Index Entries: Xylose utilization; biochemical oxidation; xylonic acid; cement; concrete.

Introduction

Concrete based on Portland cement is probably one of the most important construction and building materials to support human activity. Important urban infrastructures, such as bridges, highways, and buildings cannot be built without concrete. Various admixture chemicals are used in concrete today to improve its physical properties. One of the most important classes of concrete admixture is called water reducer. The water reducer is fundamentally a cement-dispersing chemical that deflocculates cement grains so that the cement paste becomes more fluid. The fluidity translates into the improved placement ability of the concrete. Alternatively, the action

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of water reducer allows less amounts of water in concrete without sacrificing workability or placement ability, which results in the greater strength and better durability of the concrete. The most widely used water-reducing chemical is lignosulfonate contained in spent sulfite liquor (SSL) from a sulfite pulp mill. Meanwhile, aldonic acid, particularly gluconic acid, is known to be a superior inorganic-dispersing chemical. Sodium gluconate is used in cement and concrete industries as a cement-dispersing and a retarding agent today, but its use as a general water-reducing chemical is somewhat limited as a result of its cement set retarding tendency.

In the present study, the authors attempted to convert xylose to xylonic acid and evaluate its potential as a water-reducing chemical in concrete. Xylose was selected as a starting aldose because, although it is abundant in various agricultural waste streams, the pentose is significantly underutilized because of its poor fermentatability. Xylonic acid is expected to be a cement dispersant because of its similarity to gluconic acid. However, little or no research has been done on evaluating xylonic acid as a dispersant.

Materials and Methods

Bioconversion of Xylose With Commercial Glucose Oxidase

Glucose oxidase enzymes used are OxyGo® 1500 (Genencor International, Rochester, NY) and Novozym 771® (Novozymes North America, Franklinton, NC) and catalase used was Fermacolase® 1000 (Genencor International). The enzymes were kindly supplied by the manufacturers. The enzyme conversion experiments were done according to Lantero et al. *(1)* except that 1 atm back-pressure was not applied. A preliminary experiment showed that the OxyGo sustained its activity longer than Novozym, especially at the elevated temperature; therefore, most of the experiments were done with OxyGo. Typical reaction conditions are as follows: OxyGo (2.25 mL) and Fermcolase (1.55 mL) were added to 125 g of a sugar (10–30 wt% concentration). Air was bubbled at 3 SCFH rate. The pH controller was set at 5.0, and the temperature was kept at either 40 or 60°C. A 0.1 *N* NaOH solution was used for pH adjustment. The conversion rate was measured primarily by the alkaline consumption. The aldonic acid formation was confirmed by high-performance liquid chromatography (HPLC) method described later.

Catalytic Oxidation of Xylose

The catalytic oxidation was performed using 5% palladium on activated carbon (Aldrich, Milwaukee, WI). Typical conditions of aldose conversion with palladium catalyst is described elsewhere *(2)*. Twenty grams of sugar were dissolved in 170 mL of water, and 4 g of wet 5% palladium on carbon catalyst (2 g dry powder) were added. Air was bubbled through sintered glass at 3 SCFH. Temperature was held at 35 or 50°C. The

pH was adjusted at 10.0 with using 25 wt% NaOH solution. The consumption of the sodium hydroxide solution was monitored to measure the conversion rate.

Microbial Oxidation of Xylose in SSL

Microbial oxidation of xylose in SSL was carried out using *Gluconobactor oxydans suboxydans* (ATCC 621). Substrate used was concentrated hardwood (birch) SSL from Fraser Paper (Park Falls, WI), which contains approx 30 wt% of xylose on dry solid basis.

Fermentation experiments were done based on the studies by Buchert et al. *(3,4)*. Inoculums was prepared by a shake flask culture of *G. oxydans* using pure xylose basal media according to Buchert et al. *(3,4)*; the basal media was inoculated (10^5 cells/mL) and incubated (50 mL in 250 mL) at 25°C. The pH, OD (550 nm), and viable counts were checked for the cell growth. Typically after 5 d (over 10^7 cells/mL) of cultivation, although *G. oxydans* is under exponential growth phase, the cell was spun down and used for the bioconversion test. The bioconversion of xylose in SSL was performed with sterilized 500 mL fermentation setup. Four hundred milliliters of SSL substrate solution at 15 wt% was inoculated at the cell density of either 1.02 or 4.59 (g dc/L), which corresponded to the viable count of 2×10^7 and 6×10^7 (CFU). Filtered air was supplied to the reactor and pH was controlled constant at 5.5. Quantification of the xylonic acid as well as the xylose/xylonic acid ratio were measured by HPLC using an organic acid column (Aminex HPX-87H, Bio-Rad, Hercules, CA) in combination with both ultraviolet (UV) detector and refractive index (RI) detector. By using both UV and RI responses of pure xylose and xylonic acid standards, the calibration factors matrix could be established so that each concentration of xylose and xylonic acid in the mixture could be accurately determined even though the two peaks were overlapped.

Cement Paste Test

A cement paste test was conducted according to the procedure described elsewhere *(5,6)*. Xylonic acid was also tested in a cement heat calorimeter, 3114/3236 TAM Air Isothermal Calorimeter (Thermometric AB, Sweden), to study its setting time behavior. The initial set-time was determined by the onset of the main heat peak. Two commercial Type I/II ordinary Portland cements were used for this calorimetric testing, referred to as Cement B and C. Cement B had a lower soluble alkali content than Cement C.

Mortar Flow Test

Mortar flow test was performed based on JIS A 5201 and A 1173. Mortar was mixed using Hobart mixer. Admixture chemical was added 1 min after mortar was mixed with a mixing water ("delayed" addition). The delayed addition is known to represent actual field concrete performance

better than adding a chemical to the mixing water before the mortar preparation. Mortar mix proportions were either cement/sand/water ratio of 460/1350/235 g (w/c = 0.511) or 384/1350/230 g (w/c = 0.599). Cement used was Type I/II ordinary Portland cement. Sand was ISO standard EN-sand. Both mortar slump and flow were measured, and workability = (slump + flow) – 100, was calculated. No tests were made on hardened mortar specimens.

Concrete Test

Concrete testing was performed according to ASTM C192 (specimen preparation), C143 (slump test) and C39 (compressive strength) with the xylonic acid prepared above and compared with performance of the desugared calcium lignosulfonate (Fraser Paper, WI) and sodium gluconate. The starting material, D-xylose, was also tested for slump for a comparison. The concentrated SSL before desugaring process was also obtained from Fraser Paper. The SSL samples before and after the microbial conversion were tested in concrete along with the desugared SSL. The cement factors applied were either 564 lb/yd^3 concrete (334.6 kg/m^3) or 517 lb/yd^3 concrete (306.7 kg/m^3). Coarse aggregate amount was 1800 lb/yd^3 concrete (1068 kg/m^3). The water-to-cement ratios were between 0.567 and 0.617. Varied dosages of the samples were added to concrete and 9-min slump, air content, initial set-time, and 3, 7, and 28-d compressive strength were measured according to ASTM.

Results and Discussion

Bioconversion of Xylose With Commercial Glucose Oxidase

Figure 1 shows the initial enzymatic conversion rate of glucose and xylose at 40°C. As seen in the figure, xylose was successfully converted to xylonic acid although the conversion rate was approx 1/5 of glucose. Interestingly, when the temperature raised to 60°C with all the other variables constant, the conversion rate of xylose increased dramatically and became comparable to that of glucose at 40°C (Fig. 2). The results imply that the oxygen supply might become the rate-determining step for glucose conversion, although the other step was rate determining in the case of xylose conversion *(7)*.

Table 1 summarizes the initial conversion rate of xylose compared to glucose. Under the present conditions, the impact on the xylose conversion rate with commercial GO*x* appears the following order: temperature > enzyme dosage = substrate concentration > catalase dosage. It should be noted that OxyGo already contains some catalase. It seems that the enzymatic oxidation of xylose may be practical if the temperature tolerance of the GO*x* enzyme is further improved.

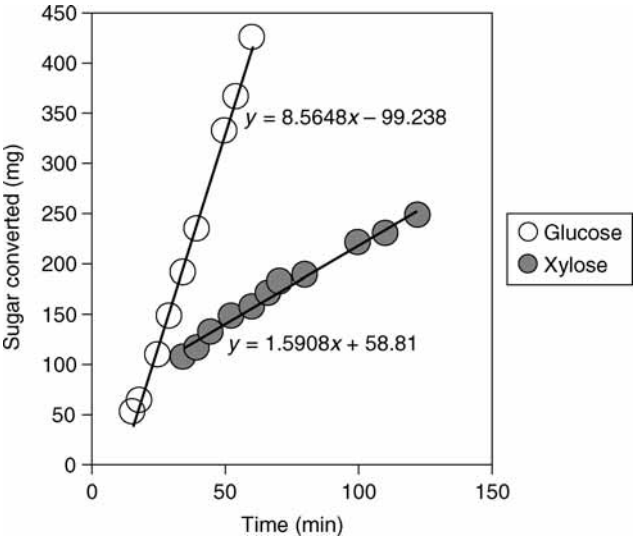

Fig. 1. Initial conversion rate of glucose and xylose using glucose oxidase (OxyGo 1500 at 2.25 mL/125 g substrate) and catalase (fermacolase at 1.55 mL/125 g substrate) at 40°C. Substrate concentration is 25%.

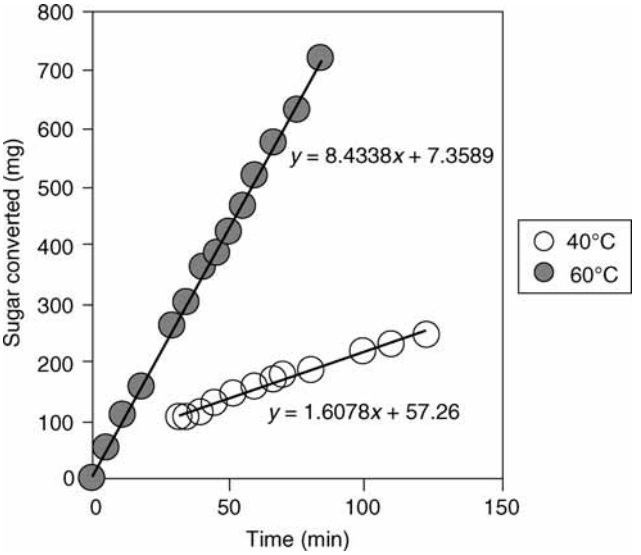

Fig. 2. Initial conversion rate of xylose using glucose oxidase (OxyGo 1500 at 2.25 mL/125 g substrate) and catalase (Fermacolase at 1.55 mL/125 g substrate) at 40°C and 60°C. Substrate concentration is 25%.

Catalytic Oxidation of Xylose

Palladium catalyst conversions was done in a similar way to the enzymatic conversion except that the pH was adjusted to 10.0. Figure 3 shows the catalytic conversion results of glucose and xylose at 35°C with

Table 1
Initial Conversion Rate of Xylose and Glucose Using Glucose Oxidase (Oxygo 1500) and Catalase (Fermacolase)

Substrate (125 g)	Substrate concentration (%)	Temperature (°C)	OxyGo (mL)	Fermcolase (mL)	Initial rate (mg/min)
Glucose	25	40	2.25	1.55	9.1
Glucose	10	60	2.25	1.55	11.8
Xylose	25	40	2.25	1.55	1.6
Xylose	25	60	2.25	1.55	8.4
Xylose	10	60	2.25	1.55	5.1
Xylose	10	60	2.25	0.8	3.7
Xylose	10	60	4.5	1.55	8.7
Xylose	10	60	4.5	0.8	9.5
Xylose	10	60	4.5	3.2	9.6
Xylose	10	63.5	2.25	0	3.4
Xylose	10	63.5	2.25	0.4	3.1
Xylose	10	63.5	1.15	0.4	1.2
Xylose	10	63.5	1.15	0.2	1.7

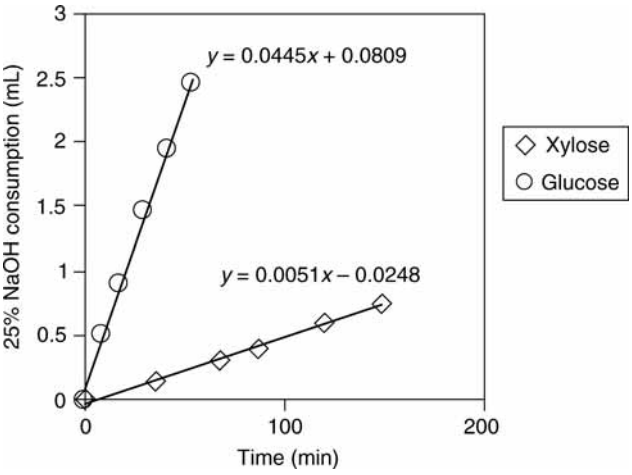

Fig. 3. Catalytic (Pd/C) oxidation rates of xylose and glucose at 35°C .

25 wt% substrate concentration. The conversion rate of xylose turns out to be approx 1/8 of glucose. When the temperature raised to 50°C, the conversion rate of xylose became comparable with glucose at 35°C (Fig. 4). The observed temperature effect is similar to the enzymatic conversion case. Mannose and galactose were even slower than xylose (Fig. 4).

Microbial Oxidation of Xylose in SSL

During the bioconversion, viable count remained relatively constant. The microbe was not growing but held its viability and enzyme activity.

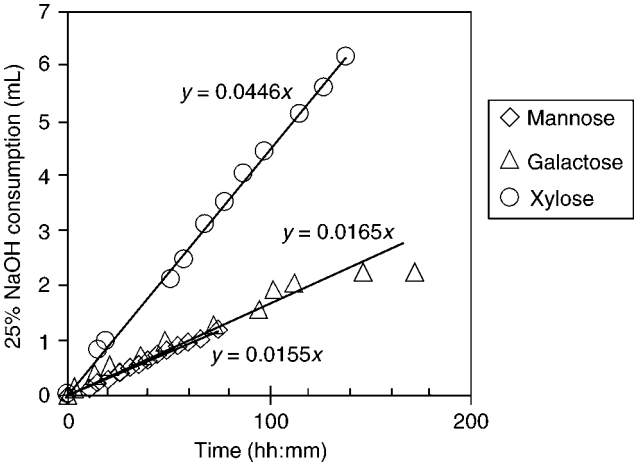

Fig. 4. Catalytic (Pd/C) oxidation rates of xylose, galactose, and mannose at 50°C.

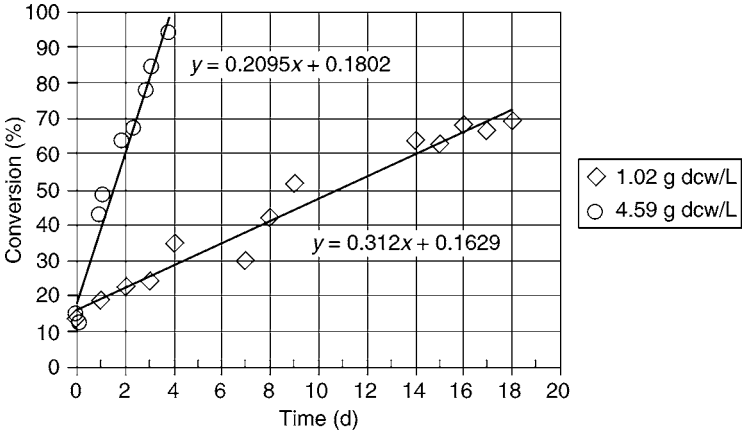

Fig. 5. Microbial bioconversion of xylose in hardwood (birch) spent sulfite liquor to xylonic acid using *Gluconobactor oxydans suboxydans* (ATCC 621).

Figure 5 shows the two bioconversion tests results with different amount of cell density. The conversion rate appears to be linear to the cell density in this range. With the higher cell density, the conversion was completed within 4 d. The high-cell density result, 4 d turn around, implies that the conversion could potentially be done without sterilized condition. Thus obtained product was tested in cement paste, mortar, and concrete to estimate the performance improvement by the bioconversion.

Cement Paste Calorimetric Test

Figure 6A and B shows the cement paste calorimety results of xylonic acid and sodium gluconate in two different cements. Both cements showed considerable set-time retardation with gluconate, although xylonic acid increases the set-time in a more linear manner. The tendency is generally

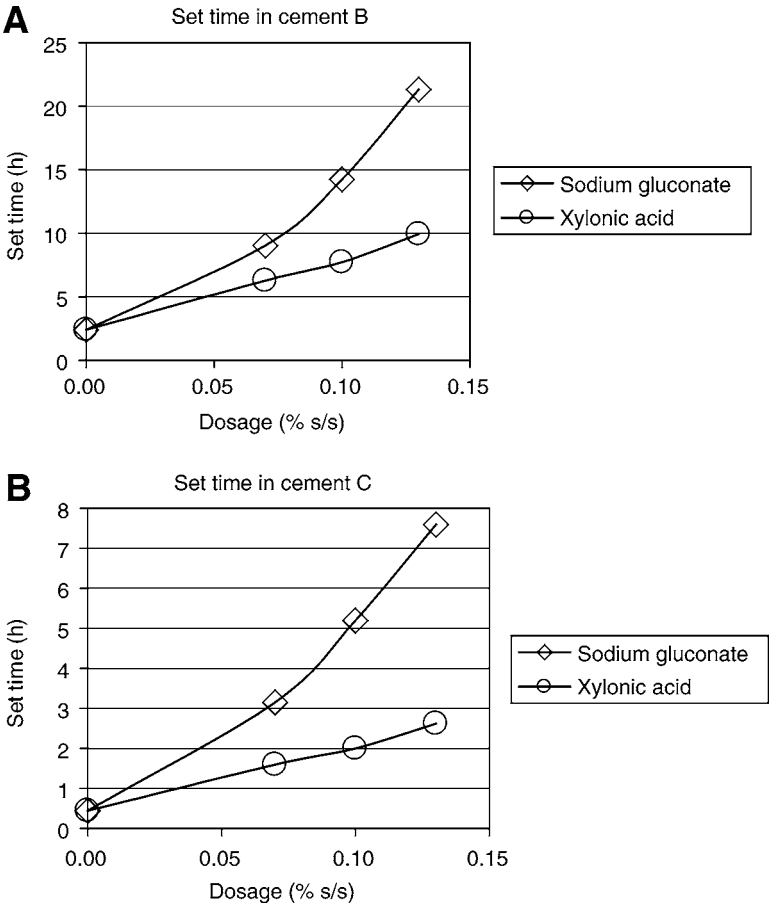

Fig. 6. Cement paste calorimetry results of xylonic acid and sodium gluconate with **(A)** regular soluble alkali cement, **(B)** higher soluble alkali cement.

favorable for water-reducing chemical application because the set-time is more predictable. The cement retardation mechanism of gluconic acid is still unclear, but, in general, it is believed that the precipitation of gluconate on the hydrating cement surface prohibits further hydration of cement. This tendency can be seen in many compounds having α-hydroxy carbonyl functionality *(8)* although there are several exceptions, such as lactic acid *(9)*.

Mortar and Concrete Test Results of Xylonic Acid

Table 2 shows the mortar test results on xylonic acid. The workability result confirmed that xylose becomes an active cement dispersant by converting it to the corresponding aldonic acid. The dispersion effect was comparable to that of gluconate while the retardation was not as large as gluconate.

Figures 7 and 8 show the concrete slump and set-time test results of xylonic acid as a function of applied dosage (wt% on cement weight), respectively. The concrete test results also demonstrate that the xylonic

Table 2
Mortar Test Results of Xylonic Acid

	Workability index (mm)	Setting time (hh:mm)
Blank	83	4:21
Sodium gluconate	182	13:53
Xylose	55	7:04
Xylonic acid	174	8:52

The cement/sand/water ratio was 460/1350/235 (w/c = 0.51). Chemicals' dosage fixed at 0.1 wt% on cement.

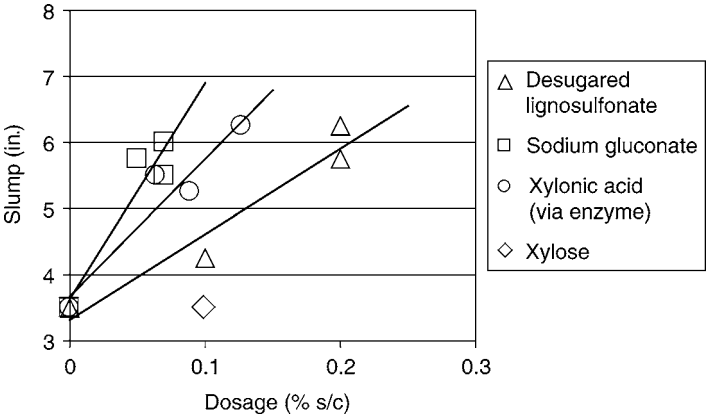

Fig. 7. Concrete slump test results of xylonic acid, sodium gluconate, and desugared lignosulfonate, cement factor = 564 lb/yd^3 (334.6 kg/m^3), coarse aggregate = 1800 lb/yd^3 (1068 kg/m^3), and w/c = 0.567.

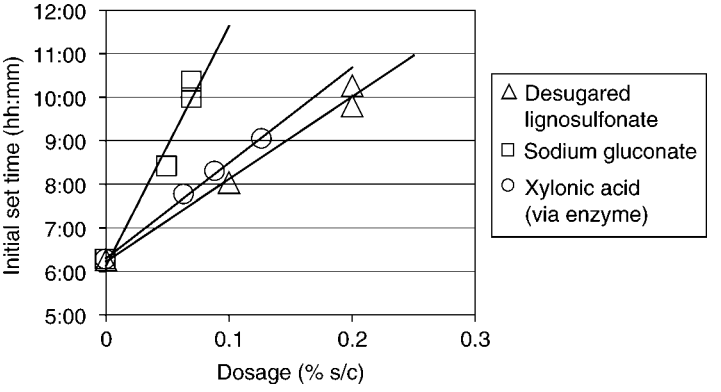

Fig. 8. Concrete initial set-time results of xylonic acid, sodium gluconate, and desugared lignosulfonate, cement factor = 564 lb/yd^3 (334.6 kg/m^3), coarse aggregate = 1800 lb/yd^3 (1068 kg/m^3), and w/c = 0.567.

acid effectiveness as a cement dispersant. The results indicates that the xylonic acid is twice as effective as lignosulfonate and slightly inferior to gluconate. The concrete set-time result also in agreement with the paste

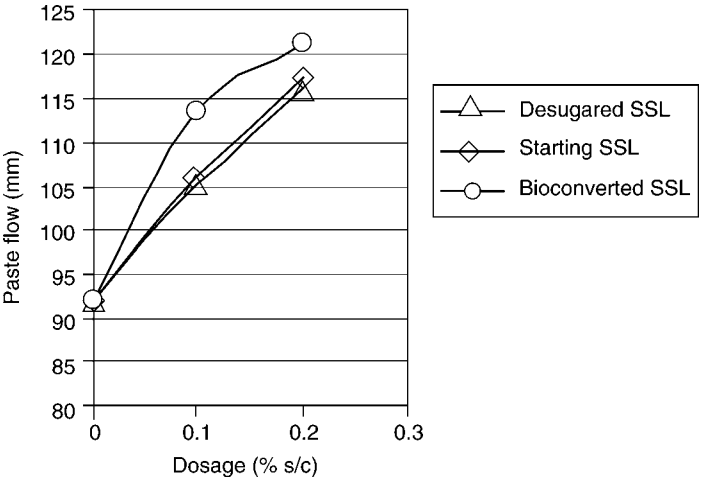

Fig. 9. Cement paste flow results of the bioconverted hardwood (birch) spent sulfite liquor (SSL) along with the starting SSL and the corresponding desugared SSL. The water-to-cement ratio is 0.5. The data points at 0.1% dosage was average of duplicate tests.

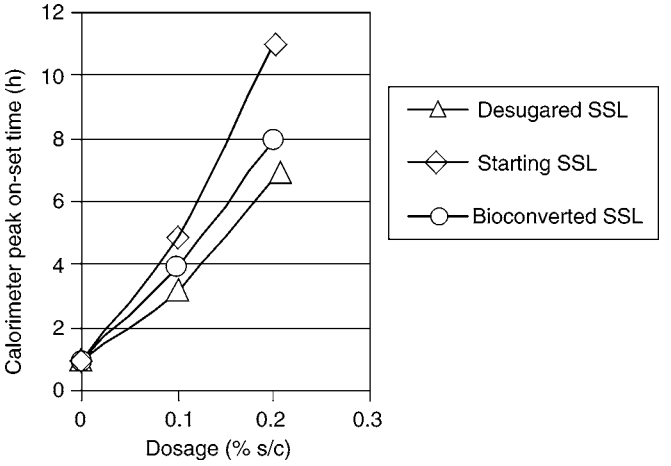

Fig. 10. Cement paste set-time results of the bioconverted hardwood (birch) spent sulfite liquor (SSL) along with the starting SSL and the corresponding desugared SSL. The water-to-cement ratio is 0.5. The data points at 0.1% dosage was average of duplicate tests. Set-time was determined by calorimetry.

and mortar results. It should be noted that the significantly shorter set-time of xylonic acid than gluconate may be somewhat exaggerated by residual chloride salt originated from the enzyme solution because chloride is a known set acceleration chemical.

Cement Paste, Mortar, and Concrete Test Results of Bioconverted SSL

Figures 9 and 10 show the cement paste tests results of bioconverted SSL along with the starting SSL containing about 30 wt% of xylose and the

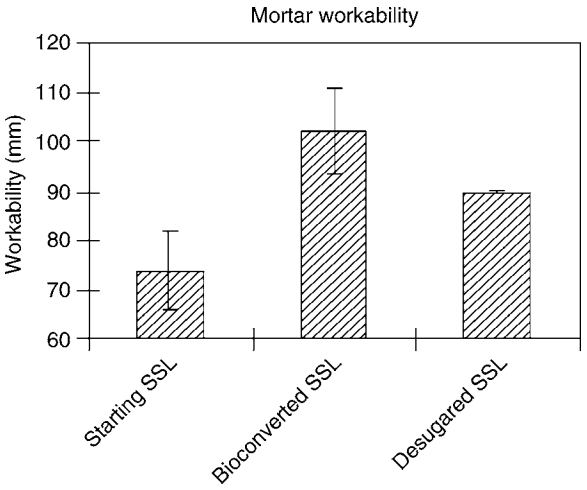

Fig. 11. Mortar workability test results of the bioconverted hardwood (birch) spent sulfite liquor (SSL) along with the starting SSL and the corresponding desugared SSL. The cement/sand/water ratio was 384/1350/230 (w/c = 0.598). Dosage was fixed at 0.1% on cement. Average of duplicate tests and the bars in the chart show the range of the data.

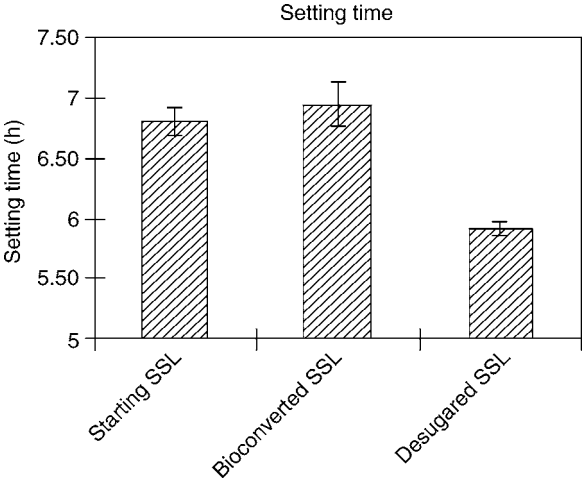

Fig. 12. Mortar set-time test results of the bioconverted hardwood (birch) spent sulfite liquor (SSL) along with the starting SSL and the corresponding desugared SSL. The cement/sand/water ratio was 384/1350/230 (w/c = 0.598). Dosage was fixed at 0.1% on cement. The set-time was determined by penetrometer method. Average of duplicate tests and the bars in the chart show the range of the data.

desugared lignosulfonate from the same source. It can be seen in Fig. 9 that the paste flow performance is improved by the bioconversion. Considering the xylose content in the SSL is about 30%, the improvement by the bioconverted seen in the result is in good agreement with the above straight xylonic acid results. Meanwhile the retardation of SSL was moderately increased by the bioconversion. Figures 11 and 12 shows the mortar results.

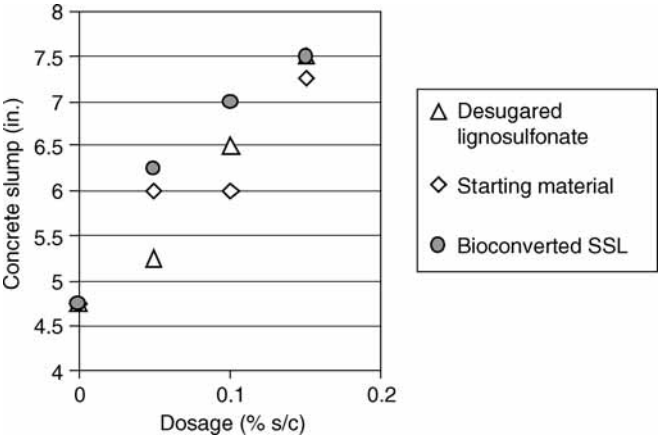

Fig. 13. Concrete slump test results of the bioconverted hardwood (birch) spent sulfite liquor (SSL) along with the starting SSL and the corresponding desugared SSL. Cement factor was 517 lb/yd^3 (306.7 kg/m^3). Coarse aggregate amount was 1800 lb/yd^3 (1068 kg/m^3). The water-to-cement ratios was 0.617.

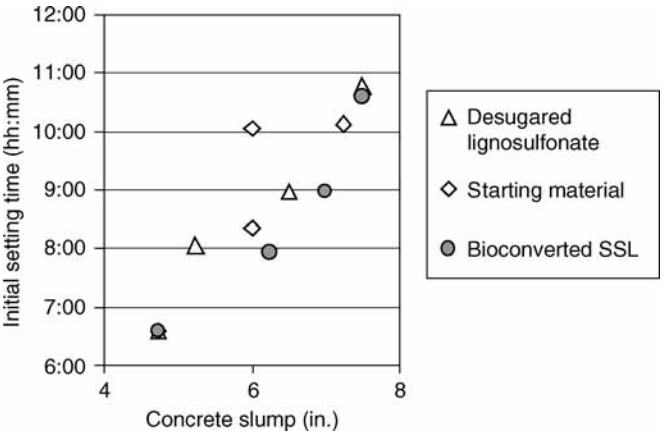

Fig. 14. Concrete initial set-time results of the bioconverted hardwood (birch) spent sulfite liquor (SSL) along with the starting SSL and the corresponding desugared SSL, plotted against respective slump value. Cement factor was 517 lb/yd^3 (306.7 kg/m^3). Coarse aggregate amount was 1800 lb/yd^3 (1068 kg/m^3). The water-to-cement ratios was 0.617.

The dosage is fixed at 0.1% solid on cement weight. The data are the average of two results. The mortar results are essentially the same as the above paste results. Figure 13 shows the concrete slump result of the sample. Although the data is rather scattered, the bioconverted SSL demonstrated superior slump performance against the benchmark desugared SSL. In Fig. 14, the setting times of the samples are plotted against the slump data so that set-time can be compared at the equivalent slump point. The plot shows that the bioconverted SSL has a shorter setting time at the same slump value, which is desirable for the water-reducing chemical application.

Table 3
Concrete Test Results (Two Sets) of Bioconverted SSL

	Dosage[a] (wt%)	Slump (cm)	Plastic air (%)	Set-time (hh:mm)		Strength (MPa)		
				Initial	Final	3 d	7 d	28 d
Blank	–	n/a[b]	2	n/a[b]	n/a[b]	21.7	25.6	31.6
Desugared lignosulfonate	0.15	17.1	2	6:21	7:58	21.9	28.6	33
Starting material	0.15	16.5	2.2	7:01	9:19	23.4	28.8	34.7
Bioconverted SSL	0.15	18.4	2.2	6:29	8:18	23.4	28.2	33.1
Blank	–	14	2.2	4:55	7:09	17	24.1	32.9
Desugared lignosulfonate	0.1	17.8	1.8	5:55	8:02	18.2	24.3	34.7
Starting material	0.1	18.4	1.7	6:30	8:25	18.5	25.1	36.9
Bioconverted SSL	0.1	19.1	1.7	6:25	8:34	17.4	23.8	34.5

[a]wt% of solid chemicals on cement weight.
[b]not available.
Cement factor was 517 lb/yd^3 (306.7 kg/m^3). Coarse aggregate amount was 1800 lb/yd^3 (1068 kg/m^3). The water-to-cement ratios were 0.603 and 0.600. Blank concrete has the same w/c ratio as concretes with the admixtures.

Table 3 shows the strength results of the bioconverted SSL. All the strength results of the admixture containing concrete are comparable and better than the blank concretes that have the same w/c ratio as the admixture concretes. Considering the better strength in addition to the clearly superior slump improvement of the bioconverted SSL, there is no doubt that the bioconverted SSL will be qualified as ASTM Type-A water reducer even without further formulation which is always done in a commercial water reducer products. Application and ASTM Type-F and/or Type-D may require some formulation for controlling setting time as well as potential air entrainment at higher dosage level.

All the performance test results of the bioconverted SSL indicate that the dosage performance of starting SSL was improved by approx 30%, which is in good agreement with the straight xylonic acid results; 30 wt% of nonactive xylose in the SSL was converted to the active xylonic acid that is twice as effective as lignosulfonate.

Conclusion

It was demonstrated that the xylonic acid can be derived either by enzymatic, microbial, or catalytic conversion. Enzymatic xylose conversion using a commercial glucose oxidase proceeded at the reasonable rate when higher temperature is applied. Microbial conversion seems to have an advantage if a large amount of impurities exist. Xylonic acid demonstrated that it can be effective cement dispersant having superior performance over lignosulfonate or sodium gluconate. In summary, xylose can

be effectively utilized by converting to its corresponding aldonic acid. As shown here, it cannot only replace gluconate in the existing applications, but can potentially replace other materials, such as lignosulfonate. It is expected that further investigation may provide more unique applications of xylonic acid.

Acknowledgments

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