Decomposition Study of Methyl α-D-Glucopyranoside (MGPα) and Lignin Model Compounds for better Glucose Yield during Sulfurous Acid Treatment

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From the perspective of bio-refinery, sulfurous acid (H_2SO_3) treatment of lignocellulosic biomass is attractive because of its ability to act both as an acid catalyst and as a sulfonation agent. Therefore, its capability to hydrolyze polysaccharides (especially glucan) into monosaccharides was compared with two other acids, hydrochloric and sulfuric acids. The decomposition of methyl α-D-glucopyranoside (MGPα) in these three acids, hydrochloric, sulfuric, and sulfurous acids were studied. In addition, *p*-creosol and vanillyl alcohol were introduced to check whether it is possible to convert polysaccharides (such as hemicelluloses) into monosaccharides during the sulfurous acid treatment. The results showed that the decomposition of MGPα is much slower in H_2SO_3 than in HCl and H_2SO_4. The ligninsulfonic acid resulting from the lignin sulfonation reaction can be expected to improve the efficiency of hydrolysis of polysaccharides into monosaccharides during sulfurous acid treatment. Moreover, a higher actual yield of glucose was obtained in this case than in the other two acids.
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Keywords: Methyl α-D-glucopyranoside (MGPα); Sulfurous acid hydrolysis; Monosaccharide; Hemicelluloses; Cresol; Vanillyl alcohol

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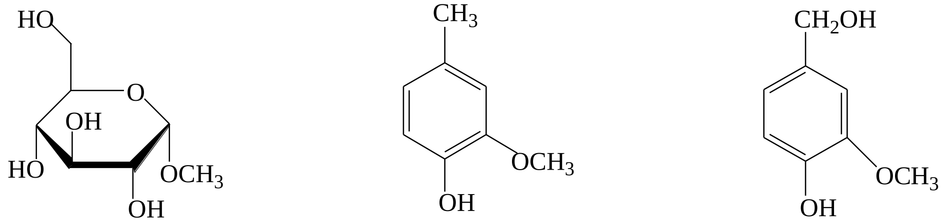
INTRODUCTION

The acid hydrolysis of glycosidic bonds during the conversion of wood polysaccharides into monosaccharides is of importance in many technical processes (Sjostrom 1993). The efficiency of the hydrolysis reaction is closely related to the recovery yield of each monosaccharide. It has been reported that hemicelluloses in lignocellulosic biomass are first hydrolyzed by acids such as HCl, H_2SO_4, or H_2SO_3 into monosaccharides (Taherzadeh and Karimi 2007). Subsequently the fermentation process was employed to convert the monosaccharides into useful compounds, such as ethanol and others (Tanifuji *et al.* 2011; Gaur *et al.* 2016; Kapoor *et al.* 2016). As catalyst, acid catalyzes not only the hydrolysis of polysaccharides into monosaccharides, but also further degradation reactions of monosaccharides into furans and carboxylic acid. Thus the yield of monosaccharides from hemicelluloses during acid treatment can be affected by both the efficiency of polysaccharide hydrolysis and the preservation of monosaccharides produced. Many researchers have shown that sulfurous acid treatment seems to be beneficial to bio-refinery for hemicelluloses processing (Zhu *et al.* 2009; Yu *et al.* 2010; Shi *et al.* 2012; Shi 2014). In a way, sulfurous acid treatment of lignocellulosic biomass is attractive because it not only removes lignin effectively but also hydrolyzes hemicelluloses to monosaccharides (Takahashi *et al.* 2010; Shi *et al.* 2013). Usually the temperatures used in acid hydrolysis of lignocellulosic materials are between 110 °C to 160 °C. Previous results by the authors suggested that sulfurous acid is a weak acid to hydrolyze hemicelluloses compared with H_2SO_4 and HCl, but the hydrolysis of cellulose in sulfurous acid treatment at 140 °C is not more severe than that in H_2SO_4 and HCl treatments at 120 °C (Shi *et al.* 2012), indicating that the use of sulfurous acid retains the yield of cellulose, while the hemicelluloses of wood are equally well hydrolyzed to monosaccharides in comparable yield with strong mineral acids such as HCl and H_2SO_4. Such sulfurous acid treatment is well suited to biorefinery operation of an existing sulfite-based pulping facility for the recovery of hemicelluloses-derived monosaccharides, especially xylose and glucose. Therefore, in order to understand the difference in abilities to hydrolyze polysaccharides (especially glucan) into monosaccharides using H_2SO_3 and two other acids, MGPα was selected as the carbohydrate model compound (Bragd *et al.* 2000; Ciriminna *et al.* 2000; Nakagawa *et al.* 2015). Creosol and vanillyl alcohol as lignin model compounds (Carvalho *et al.* 1998; Nie *et al.* 2014) were employed to check their effect on glucose yield during acid hydrolysis. The molecular structures of three models are shown in Fig. 1.



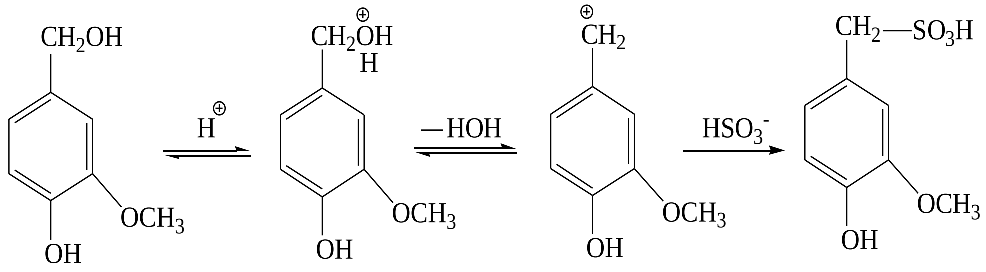
Methyl *α*-D-glucopyranoside (MGP*α*)

2-Methoxy-4-methylphenol (2-Methoxy-*p*-cresol)

3-Methoxy-4-hydroxybenzyl alcohol (Vanillyl alcohol)

Fig. 1. Molecular structures of three model compounds

It was presumed that one of the lignin model compounds, the creosol, would not react with HSO_3^- ion but vanillyl alcohol would react with HSO_3^- to form vanillyl alcohol sulfonic acid *via* the carbonium ion intermediate in acidic solution as shown in Fig. 2.



Vanillyl alcohol

Carbonium ion intermediate

Vanillyl alcohol sulfonic acid

Fig. 2. Reaction of vanillyl alcohol in sulfurous acid

The adducts thus produced from the reaction with HSO_3^- ion may affect the glucose yield from hemicelluloses or cellulose of wood after sulfurous acid treatment.

EXPERIMENTAL

Materials

All monosaccharides including D-glucose, D-xylose, L-arabinose, D-mannose, and D-galactose, sodium borohydride ($NaBH_4$), sodium bisulfite ($NaHSO_3$), and other reagents were obtained from Wako Pure Chemical Industries Ltd. (Japan). The monosaccharides were used without further purification. Methyl α-D-glucopyranoside (MGPα) was purchased from Tokyo Chemical Industry Co. Ltd. (>98.0%). 2-Methoxy-4-methylphenol (Cresol (98%)) was supplied by Hubei Xin Ming Tai Chemical Co. Ltd. (China).

Experimental Instruments

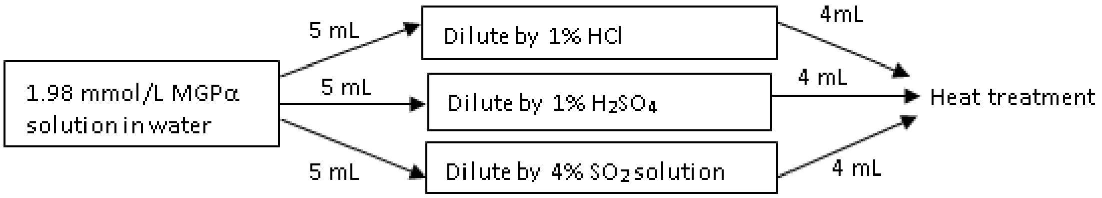
The following instruments were used for performing the experiments and analysis:

1. Teflon inner tube sealed container (TAF-SR type; total volume of Teflon inner tube: 50 mL; maximum working pressure: 10 MPa; maximum working temperature: 180 °C; and seal O-ring: Fluorinated gum) from Taiatsu Techno Corp., Japan)
2. Oil bath with shaker RSO-200TE (Riko, Japan)
3. Gas chromatograph (GC-14B, Shimadzu, Japan)

Analytical Procedures

Acid hydrolysis of methyl- α-D-glucopyranoside (MGPα)

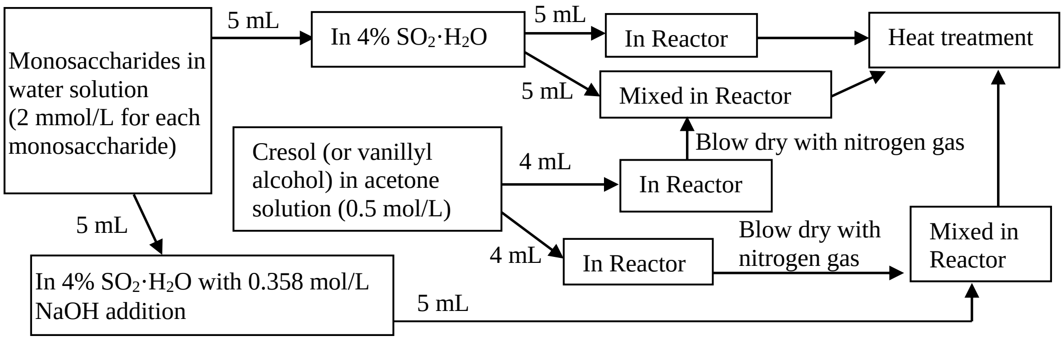
Three acid solutions of the same pH containing the same amount of MGPα were prepared, as shown in Scheme 1. It was assumed that commercially available SO_2 solution is 5% with pH = 0.93 (25.9 °C). Therefore, before heat treatment in oil bather shaker, the initial pH of the reaction mixture was determined to be more than 0.93. Similarly HCl, pH = 1.04 (25.7 °C); H_2SO_4, pH = 1.02 (25.9 °C); and 4% solution of SO_2, pH = 1.01, respectively. Approximately 4 mL of the diluted solution was put in a reactor and subjected to heat treatment at 140 °C. Acetylation reaction was then conducted with the addition of sodium acetate (CH_3COONa) and acetic acid at 120 °C for 3 h.



Scheme 1. Preparation of MGPα in three acids with the same pH

Preparation of monosaccharides in sulfurous acid containing lignin model compound

The experimental procedure is shown in Scheme 2. To begin with, 0.5 mol/L acetone solutions of cresol and vanillyl alcohol were prepared. About 4 mL of this solution was taken in the reactor and blown dried with nitrogen gas. Then 5 mL of monosaccharides solution in sulfurous was added, and subjected to heat treatment at 140 °C.



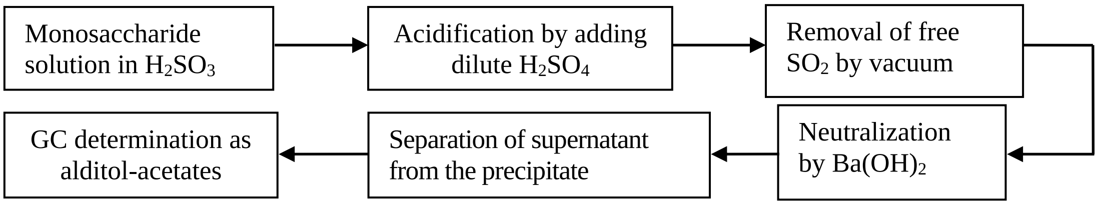
Scheme 2. Preparation of monosaccharides in sulfurous acid with and without addition of base

GC-Analysis of monosaccharide yield by alditol-acetate method (Borchardt & Piper 1970)

The resultant reaction mixture from heat treatment was allowed cool. An internal standard (inositol, 3 mmol/L), was then added. The reaction mixtures from acid treatments was neutralized to pH 5.5 with NaOH solution for HCl treatment and $Ba(OH)_2$ solution for sulfuric acid treatment (for the neutralization of H_2SO_3, see next section). The supernatant liquid of neutralized samples were reduced by $NaBH_4$ for 24 h. The excess $NaBH_4$ present in the reaction medium was quenched by the addition of acetic acid, and the borate was then removed by repeated methanol addition and followed by evaporation. The alditols thus produced were converted into acetates by the addition of acetic anhydride. The gas chromatographic analysis was performed (GC-14B, Shimadzu, Japan) under the following conditions: Column used - TC-17, 30 m × 0.25 mm; Column temperature, 220 °C; Injection temperature, 220 °C; Detector temperature, 230 °C.

Determination of monosaccharides from sulfurous acid treatment

The resultant reaction mixture was exposed to the same conditions outlined in Scheme 3 for estimating the monosaccharide from sulfurous acid treatment. As described earlier, after the addition of the inositol (internal standard), the reaction mixture was further acidified to pH 1 by the addition of dilute sulfuric acid solution, and then kept under reduced pressure for about 1 h to eliminate SO_2 from the medium. After this step, the reaction mixture was neutralized to pH 5.5 with $Ba(OH)_2$ solution. The supernatant liquid was converted into alditol acetates in the same manner as described previously and subjected to GC analysis.



Scheme 3. Sample preparation to determine monosaccharides after sulfurous acid treatment

RESULTS AND DISCUSSION

Calibration Curve of α-D-Glucopyranoside (MGPα)

A standard solution containing 50 mmol/L of MGPα and 3 mmol/L of inositol was prepared for calibration. The calibration curve for α-D-glucopyranoside is shown in Fig. 3. Thereafter, the actual glucose yield resulted from the decomposition of MGPα at 140 °C was determined by GC analysis, and specific calculation methods are as follows:

$$\text{Actual yield of glucose} = \frac{W_{\text{actual glucose}}}{(W_{\text{original MGP}\alpha}/194.18)*180} *100\% \quad (1)$$

$$\text{Remaining MGP}\alpha = \frac{W_{\text{MGP}\alpha}}{W_{\text{original MGP}\alpha}} *100\% \quad (2)$$

$$\text{Theoretical D-glucose yield} = \text{degraded MGP}\alpha = (1\text{- remaining MGP}\alpha *100\% \quad (3)$$

where, $W_{\text{actual glucose}}$ is the weight of actual glucose from decomposition of MGPα, calculated based on calibration curve y_{glucose}; $W_{\text{MGP}\alpha}$ is the weight of remained MGPα, calculated based on calibration curve $y_{\text{MGP}\alpha}$; $W_{\text{original MGP}\alpha}$ is the weight of MGPα added to Teflon inner tube sealed container; $y_{\text{glucose}} = 1.0042\, x_{\text{glucose}} + 0.0503$ (here, x is the area ratio of glucose to inositol, y is the Molar ratio of glucose to inositol), $y_{\text{MGP}\alpha} = 1.1171\, x_{\text{MGP}\alpha}$ (here, x is area ratio of MGPα to inositol, y is the Molar ratio of MGPα to inositol)

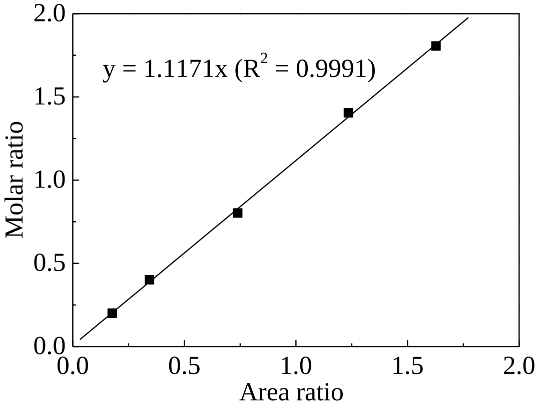


Fig. 3. Calibration curve prepared for α- D-glucopyranoside (MGPα)

Decomposition of Methyl α-D-Glucopyranoside (MGPα) in HCl, H_2SO_4, and H_2SO_3 Solutions

Decomposition of MGPα was investigated at 140 °C, and the results showed that the decomposition of MGPα was much slower in SO_2 aqueous solution than in HCl and H_2SO_4 (as shown in Fig. 4) solutions. The yield of glucose formed in SO_2 aqueous solution was almost the same as that formed in HCl and H_2SO_4 solutions, nevertheless at the expense of treatment time. At short treatment times (less than 15 min), the yield of glucose formed in SO_2 aqueous solution was about the half of that formed in HCl and in H_2SO_4 solutions. Although the hydrolyzing efficiency of MGPα to form glucose was lower in H_2SO_3 than by the other two acids, the preservation of glucose that are produced seemed to be better in H_2SO_3 than in the other two acids after full degradation of MGPα.

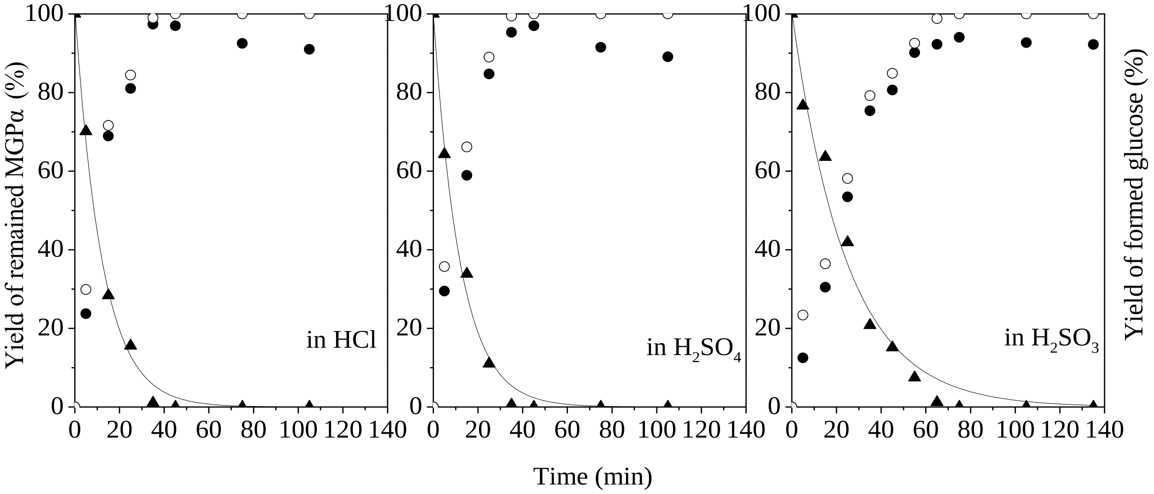


Fig. 4. Disappearance of MGPα and formation of glucose in the acid hydrolysis (▲ remained MGPα; ○ theoretical yield of glucose; ● actual yield of glucose)

When woody biomass was hydrolyzed by H_2SO_3, the ligninsulfonic acid formed from lignin sulfonation reaction can be expected to improve the hydrolyzing efficiency of polysaccharides into monosaccharides in H_2SO_3. The decomposition of MGPα in H_2SO_3 with vanillyl alcohol at 140 °C, 15 min was also investigated.

As shown in Table 1, in the presence of vanillyl alcohol in H_2SO_3, the hydrolyzing efficiency of MGPα into glucose was considerably improved, moreover with higher actual yield of glucose than in other two acids. However, the presence of cresol in H_2SO_3, the hydrolyzing efficiency of MGPα into glucose was not distinctly enhanced but there is a slight increase in actual yield of glucose.

Table 1. Comparison of Decomposition of MGPα and Formation of Glucose at 15 min in Various Acidic Systems

	Remained MGPα	D-glucose yield		Actual D-glucose yield /degraded MGPα
		actual	theoretical	
In HCl	28.36	68.95	71.64	0.96
In H_2SO_4	33.83	58.92	66.17	0.89
In H_2SO_3	63.55	30.49	36.45	0.84
In H_2SO_3, with vanillyl alcohol	24.34	70.15	75.66	0.93
In H_2SO_3, with cresol	62.85	31.43	37.15	0.85

Effect of Formation of α-Sulfonic Acid on Degradation of Monosaccharides in Sulfurous Acid

Introduction of base to SO_2 aqueous solution leads to the formation of more HSO_3^- ions. In the presence of *p*-vanillyl alcohol (or cresol), the degradation experiments of monosaccharides in sulfurous with and without base were investigated at 140 °C. Para-Vanillyl alcohol as lignin model compound reacted with HSO_3^- ions to produce vanillyl alcohol sulfonic acid.

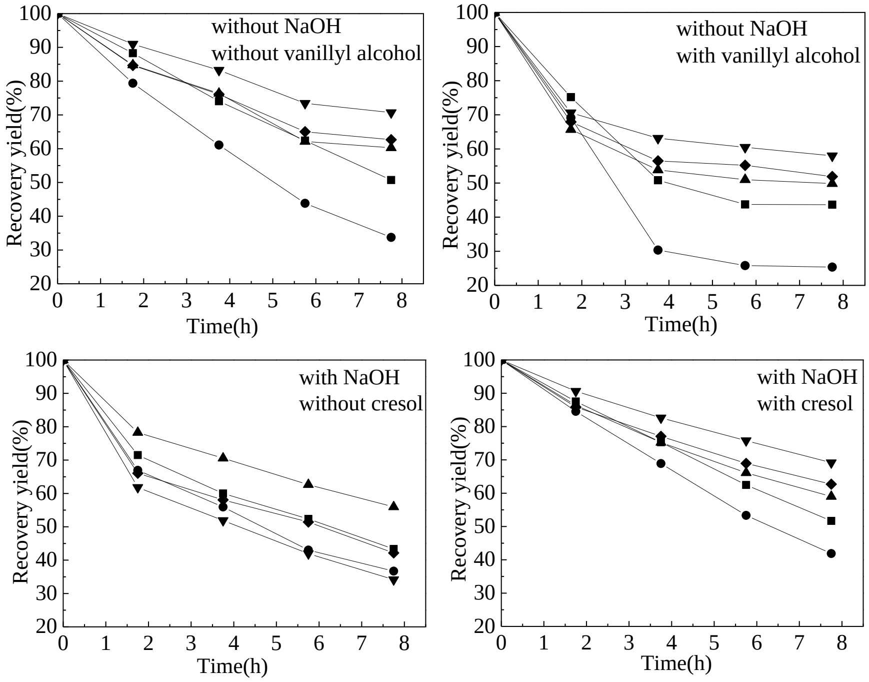


Fig. 5. Effect of lignin model compound on degradation of monosaccharides in 4% SO_2 solution with and without base at 140 °C (■ L-arabinose; ●D-xylose; ▲D-mannose ;▼D-glucose; ◆d- galactose)

As shown in Fig. 5, the degradation of each monosaccharide in sulfurous system without base and with base was lower in the absence of *p*-vanillyl alcohol than in the presence of *p*-vanillyl alcohol. Because in acidic condition, the decrease in recovery yield may be due to the formation of vanillyl alcohol-sulfonic acid, which lowered the pH of system and accelerated the degradation of each monosaccharide in sulfurous system because of the reaction between benzyl cation and hydroxyl group in monosaccharides. However, when base was added, sometimes the recovery yield of xylose and mannose were higher in the sulfurous system with base and with vanillyl alcohol than in the sulfurous system without base and without vanillyl alcohol. The addition of base seemed to prevent the degradation of arabinose, xylose, and mannose. Differently, the degradation of glucose and galactose was faster in sulfurous system with base than in sulfurous system without base. In the presence of *p*-vanillyl alcohol, mannose was most stable in sulfurous system with base. This showed that the decomposition of bisulfite adducts was slower from mannose than from other monosaccharides, suggesting that the binding power of SO_2 to the monosaccharide was the highest. This result was in agreement with the findings by other researchers (Voegele *et al.* 2002).

CONCLUSIONS

1. Decomposition of MGPα was much slower in SO_2 aqueous solution than in HCl and H_2SO_4 solutions. The yield of glucose formed in SO_2 aqueous solution was almost the same as that formed in HCl and H_2SO_4 solutions, nevertheless at the expenses of treatment time. Monosaccharides degraded at different rates in different acids even when the molar concentrations of the various acids were kept constant.
2. Lignin-sulfonic acid produced in H_2SO_3 improved the hydrolyzing efficiency of polysaccharides into monosaccharides in H_2SO_3.
3. Although the hydrolyzing efficiency of MGPα into glucose was lower in H_2SO_3 than in the other two acids, the preservation of glucose seemed to be better in H_2SO_3 than in the other two acids after the full degradation of MGPα during H_2SO_3 treatment.

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