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Enzymatic degradation of polysaccharides in Chinese vinegar residue to produce alcohol and xylose

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Abstract: Vinegar residue is a key secondary waste in the brewing industry that is often disposed irresponsibly, due to its large quantity and lack of reasonably effective use, causing environmental pollution issues. NaOH was used to pretreat Chinese vinegar residue, and the reaction products were consumed by the enzyme complex and *Saccharomyces cerevisiae* 1300 during the stage of simultaneous saccharification and fermentation (SSF). The results show that the optimal retreatment conditions for Chinese vinegar residue were solid-to-liquid ratio of 1 : 11 (w/v), NaOH concentration of 2.2%, pretreatment temperature of 63 °C, pretreatment time of 80 min, and amount of 4.9 IU g^{-1} xylanase. While these optimal conditions allowed more effective enzymatic degradation of the dried vinegar residue and resulted in the total sugar yield of 66.1%. Subsequently, dried vinegar residue and enzyme complex were added into the SSF process four times, and SSF reacted in a shaker at 120 r min^{-1} and 37 °C for 120 h, the yields of ethanol and xylose were 31.4% and 18.5%, respectively. Therefore, the method of Chinese vinegar residue for alcohol and xylose production by SSF was proved.

Keywords: NaOH; pretreatment; enzymolysis; simultaneous saccharification and fermentation; xylanase; enzyme complex

Vinegar is consumed worldwide as a food condiment and preservative (Li et al. 2014). Vinegar can increase appetite, and its long-term use is believed to relieve fatigue, protect the gastric mucosa, prevent osteoporosis, lower blood pressure and blood lipids, and possess anticancer effects (Kishi et al. 1999; Kondo et al. 2001; Johnston and Buller 2005). Meanwhile, Chinese vinegar has a history of more than 3 000 years, traditional Chinese vinegar is made from different sorts of cereals, especially glutinous rice, and its solid-state fermentation process comprises four successive stages, namely koji preparation, saccharification of starch and alcohol fermentation, acetic-acid fermentation and maturation (Liu et al. 2004). There are at least 14 main types on the market, including Sichuan bran vinegar, Shanxi old mature vinegar and Zhenjiang aromatic vinegar, whereas all consist of water, acetic acid, polysaccharides, soluble salt-free solids content, and other constituents (Chen et al. 2012).

However, due to the characteristics of the traditional solid-state fermentation process, a large amount

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of vinegar residue is produced. At present, China produces approximately 2.5 million tons of vinegar and 2 million tons of vinegar residue (Zhang et al. 2019). Due to its large quantity and lack of a reasonably effective reusing method, vinegar residue is often disposed irresponsibly. Moreover, this may affect the healthy development of the brewing industry. Currently, the search for multi-purpose reuse of vinegar residue is a hot topic in the field. Nowadays, the main channels to reuse vinegar residue is the feed production and for use as cultivation substrates and bioenergy (Song et al. 2013; Tian et al. 2017). The application of vinegar residue in feed production can improve the nutritive value. However, as this process still produces by-products, the environmental pollution issue has not been completely solved (Yu et al. 2009; Song 2012; Song et al. 2013; Wang et al. 2018). As cultivation substrates, vinegar residue can reduce cultivation and production costs, which in turn increase economic efficiency. However, the use of vinegar residue as cultivation substrates is mainly for inexpensive fungi or landscape plants (Wang et al. 2011; Yang et al. 2014). As bioenergy, vinegar residue is mainly used for biogas production, pyrolysis and anaerobic fermentation (Smirnova 2000; Jing and Tian 2009; Zhou et al. 2017). The research on vinegar residue as bioenergy is limited to the mechanistic and procedural aspects, whereas the applicability in actual energy production still requires a further technological breakthrough.

Exploiting the high-value use of fermentation secondary waste in the vinegar industry, transforming waste into valuable resources, achieving zero emission of vinegar residue, as well as allowing the fermentation industry to achieve reasonable industrial ecology with the efficient use of secondary waste as resources, are of great significance to China's resource development and environmental protection. These are also important topics for the future of the brewing industry. This study primarily explored the stepwise degradation of vinegar residue into alcohol and xylose by enzyme complex and microorganisms, and has provided a breakthrough for the effective recycling of vinegar residue.

MATERIAL AND METHODS

Material. Fresh vinegar residue (68.0% water and 0.7% acetic acid, unprocessed wet residues after vinegar extraction, stored at 4 °C), cottonseed hulls and bran were both provided by Sichuan Baoning Vinegar Co., Ltd (China); Glucose kit, Changchun Huili Biotech Co., Ltd. (China); dextran amylase NS22035 (enzyme activity was 21 IU mL^{-1}, 1 IU of dextran amylase is defined as the enzymatic loading that releases 1 μmol glucose min^{-1} from starch at 37 °C and pH 6.8), xylanase NS220839 (enzyme activity was 17 IU mL^{-1}, 1 IU of xylanase is defined as the enzymatic loading that releases 1 μmol xylose min^{-1} from xylan at 50 °C and pH 4.8), cellulase NS22086 [enzyme activity was 25 IU mL^{-1}, 1 IU of cellulase is defined as the enzymatic loading that releases 1 mg glucose min^{-1} from Whatman 1 filter paper (United Kingdom)], and enzyme complex NS22002 (enzyme activity of the main β-glucosidase was 7 IU mL^{-1}, 1 IU of β-glucosidase is defined as the enzymatic loading that releases 1 μmol glucose min^{-1} from salicin at 37 °C and pH 5.0) were from Novozymes Biotechnology Ltd. (China); *Saccharomyces cerevisiae* 1300 was purchased from Sichuan Microbial Resources Infrastructure and Culture Collection Center (China); xylose, phloroglucinol, sodium hydroxide were of analytical grade.

Test flow. The test flow is shown in Figure 1. Firstly, crude starch was recovered from fresh vinegar residue. Secondly, NaOH was used to pretreat the dried vinegar residue after crude starch recovery. Thirdly, NaOH-pretreated dried vinegar residue was treated by xylanase. Fourthly, cellulase and enzyme complex were added to xylanase-degraded dried vinegar residue for simultaneous saccharification and fermentation (SSF).

Crude starch recovery from fresh vinegar residue. In a 500-mL glass flask, 100 g of fresh vinegar residue was mixed with 100 mL of 0.05 mol L^{-1} citrate buffer solution (pH was 6.0), and the pH was adjusted to 5 with NaOH solution (0.1 mol L^{-1}). Various amounts of dextran amylase were added. The flasks were placed in an HZO-X100 air bath shaker (Taicang, China) for 72 h at 50 °C and 150 r min^{-1}. After the reaction, vacuum filtration (SHZ-III vacuum pump; Shanghai Yarong Biochemical Instrument Factory, China) was used for solid-liquid separation. The solid was dried at 60 °C (101C-3B drying oven; Shanghai Chongming Test Instrument Factory, China) and used for follow-up tests. The recovered liquid was used as a supplemental sugar solution to SSF.

NaOH pretreatment of dried vinegar residue after crude starch recovery. Dried vinegar residue obtained after residual starch was crushed and mixed homogeneously (CS-2000 crusher; Wuyi Haina Electric Appliance Co., Ltd., China) with NaOH solution at different conditions: NaOH concentrations were

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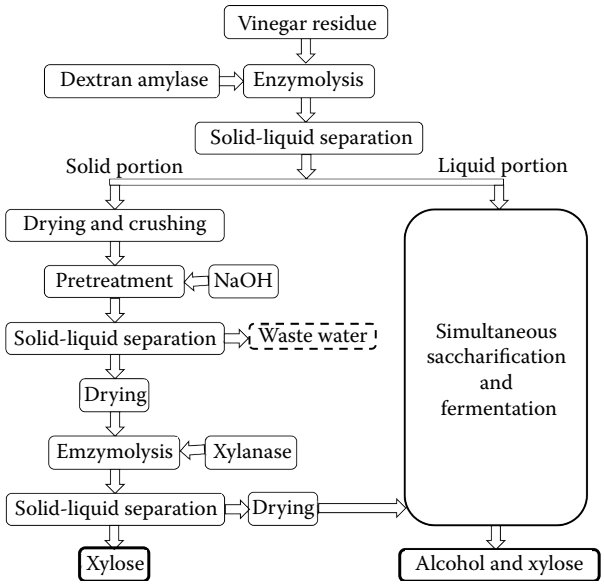

Figure 1. The test flow of this research

between 0.5% and 4%, solid-to-liquid ratios were between 1 : 6 and 1 : 14 (w/v), pretreatment times were between 30 min and 90 min, and pretreatment temperatures were between 45 °C and 85 °C. After the pretreatment, solid-liquid separation was performed. The residue was washed with warm water until neutralised, and it was dried at 60 °C (101C-3B drying oven; Shanghai Chongming Test Instrument Factory, China) to constant weight.

Xylanase treatment of NaOH-pretreated dried vinegar residue. In a 500-mL flask, dried vinegar residue, xylanase (the ratio of xylanase and dried vinegar was 0.1 : 1), and 200 mL of 0.05 mol L^{-1} citrate buffer solution were mixed. The pH of the solution was adjusted to 5, and the solution was placed in an air bath shaker (HZO-X100; Taicang, China) for 96 h at 60 °C and 150 r min^{-1}. Solid-liquid separation was performed after the reaction, while the solid was dried at 60 °C to constant weight, and the liquid was recycled.

Uniform experimental design. With reference (Liu et al. 2011) to the test results of the enzymatic hydrolysis of sugar from the residues of white spirit production, 5 factors were found to affect saccharification and degradation: x_1, x_2, x_3, x_4, and x_5 (Liu et al. 2011) while x_1 was solid-to-liquid ratio [(1 : 9~1 : 16 (w/v)]; x_2 was NaOH concentration (0.5%~4.0%); x_3 was pretreatment temperature (60~95 °C); x_4 was pretreatment time (60~95 min); x_5 was amount of xylanase (0.85~6.80 IU g^{-1}). The total sugar yield was used as a reference index, and data processing system (DPS) was used to design the uniform design table U_8 (8^5) (Wang et al. 2014). The levels of different factors are shown in Table 1, and the uniform design table U_8 (8^5) is shown in Table 2.

Simultaneous saccharification and fermentation. Cellulase and enzyme complex were added to xylanase-degraded dried vinegar residue for SSF. Firstly, 96 mL sugar solution obtained from the residual starch recovery step was mixed with 5 g dried vinegar residue, and 10 mL *S. cerevisiae* 1300 seed solution (1.1×10^7 CFU mL^{-1}) in a 250-mL flask. Meanwhile, 1 mL cellulase [5 IU g^{-1} (enzyme/solid)] and 0.25 mL enzyme complex [0.35 IU g^{-1} (enzyme/solid)] were added to the flask. The flask was covered with the plastic balloon to capture the CO_2 produced during fermentation, and was placed in the air bath shaker set at 100–140 r min^{-1} and 31–37 °C for 24 h (HZO-X100; Taicang, China). Secondly, 5 g dried vinegar residue, 1.0 mL cellulase, and 0.25 mL en-

Table 1. Levels of factors for pretreatment

Serial number	Factors				
	x_1 (w/v)	x_2 (%)	x_3 (°C)	x_4 (min)	x_5 (IU g^{-1})
1	1 : 9	0.5	50	60	0.85
2	1 : 10	1.0	55	65	1.70
3	1 : 11	1.5	60	70	2.55
4	1 : 12	2.0	65	75	3.40
5	1 : 13	2.5	70	80	4.25
6	1 : 14	3.0	75	85	5.10
7	1 : 15	3.5	80	90	5.95
8	1 : 16	4.0	85	95	6.80

x_1 – solid-to-liquid ratio; x_2 – NaOH concentration; x_3 – pretreatment temperature; x_4 – pretreatment time; x_5 – amount of xylanase

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Table 2. Uniform design U_8 (8^5) for pretreatment

Serial number	Factors					Total sugar yield (%)
	x_1	x_2	x_3	x_4	x_5	
1	1	5	4	6	8	48.2
2	4	1	3	7	2	43.8
3	3	6	5	2	4	64.0
4	6	7	7	8	5	47.5
5	2	3	8	4	3	63.5
6	7	8	2	3	1	20.5
7	5	2	6	1	7	52.2
8	8	4	1	5	6	44.3

x_1 – solid-to-liquid ratio; x_2 – NaOH concentration; x_3 – pretreatment temperature; x_4 – pretreatment time; x_5 – amount of xylanase; $U_8(8^5)$ – among the 8^5 tests, 8 tests were selected as representatives according to the uniform design principle

Table 3. Main components of dried vinegar residue before and after crude starch recovery

Component (%)	Before crude starch recovery	After crude starch recovery
Crude starch	16.12 ± 0.28	11.97 ± 0.16
Cellulose	26.47 ± 0.73	28.13 ± 0.68
Hemicellulose	24.19 ± 0.89	25.77 ± 0.95
Lignin	17.05 ± 0.52	18.42 ± 0.57
Ash	9.67 ± 0.53	9.95 ± 0.43
Other	6.50 ± 0.21	5.76 ± 0.24

zyme complex were incubated at 31–37 °C for 24 h in a shaker at 100–140 r min^{-1}. For the third step, 5 g dried vinegar residue, 1.0 mL cellulase, and 0.25 mL enzyme complex were incubated at 31–37 °C for 24 h in a shaker at 100–140 r min^{-1}. For the fourth step, the rest of dried vinegar residue, 0.3 mL cellulase, and 0.1 mL enzyme complex were incubated at 31–37 °C for 24–72 h in a shaker at 100–140 r min^{-1}.

Ethanol yield calculation. During the test, the yields of total sugar, glucose, xylose, and alcohol were calculated as follows (Equations 1 and 2):

$$SY = \frac{W_S \times 0.9}{W_{VRCS}} \times 100\% \quad (1)$$

$$EP = \frac{W_{AX}}{W_{VRED}} \times 100\% \quad (2)$$

where: SY – the yield of total sugar, glucose or xylose; W_S – the weight of total sugar, glucose or xylose after degradation; W_{VRCS} – the weight of the vinegar residue after crude starch recovery; EP – the yield of alcohol or xylose; W_{AX} – the weight of alcohol or xylose after SSF; W_{VRED} – the weight of the vinegar residue after enzymatic degradation.

Mass balance calculation. Mass balance of matter was calculated using the Equations 3 and 4 (Nwobi et al. 2015; Awedem et al. 2019).

Analytical Method. The cellulose, hemicellulose and lignin of dried vinegar residue were measured by the improved National Renewable Energy Laboratory (NREL) method (Sluiter et al. 2012). Total sugar was measured by dinitrosalicylic acid (DNS) method, glucose was measured by glucose assay kit, xylose was measured by phlo-

$$\text{Solid fraction recovery (\%)} = \frac{\text{Solid dry mass out (g)}}{\text{Solid dry mass in (g)}} \times 100\% \quad (3)$$

$$\text{Liquid fraction recovery (\%)} = \frac{\text{Liquid mass out (g)}}{\text{Liquid mass in (g)}} \times 100\% \quad (4)$$

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roglucinol chromatography, and alcohol was measured by TRACE GC DSQ II gas chromatography (Thermo, United States) using isopropanol as internal standard (Li and Sun 2002; Chen et al. 2007; Wang et al. 2010). The data were three repeats, and the main components in dried vinegar residue are shown in Table 3.

Origin Pro 10.2 software was used for data mapping, and PASW Statistics 18 software was used for statistical analysis.

RESULTS AND DISCUSSION

Analysis of crude starch recovery from fresh vinegar residue. The effects of dextran amylase on the yield of crude starch recovered from fresh vinegar residue were studied. The total sugar was measured after the experiment, and the solid and liquid fractions were recovered. As shown in Figure 2, when the amylase was at 0.16 mL 100 g^{-1}, 0.18 mL 100 g^{-1} and 0.20 mL 100 g^{-1}, the total sugar concentrations were 19.3 g L^{-1}, 19.9 g L^{-1} and 20.2 g L^{-1}, respectively. The yield of total sugar increased along with the amount of amylase used. The increase in the yield of total sugar was less obvious when more than 0.16 mL 100 g^{-1} of enzymes was used ($P < 0.05$). Considering the cost of enzymes, 0.16 mL 100 g^{-1} of enzymes should be used for recovering crude starch from fresh vinegar residue. Upon calculation, the amount of recovered liquid was 96 mL, the concentration of total sugar was 19.3 g L^{-1}. The percentage of crude starch recovered from fresh vinegar residue was 35.9%, the amount of recovered solid was 27.1 g, solid fraction recovery was 84.7%, and the percentage content of crude starch was 12.0% (Table 3).

Effects of NaOH concentration on enzymatic degradation of vinegar residue. The effects of different NaOH concentrations on enzymatic degradation of dried vinegar residue obtained after starch recovery were studied under the following conditions: a solid-to-liquid ratio of 1 : 10 (w/v), temperature of 65 °C, and reaction time of 60 min. The results are shown in Figure 3. As NaOH concentrations increased, the recovery rate of solid decreased, indicating that higher concentrations of NaOH had higher capabilities to dissolve the lignin in the raw materials. In addition, the increase in NaOH concentrations also led to losses of other materials, such as cellulose, in the vinegar residue. The concentrations of total sugar, glucose, and xylose in the enzymatic hydrolysate gradually increased. When the NaOH concentrations were 2%, 3%, and 4%, the solid fraction recovery was 64.2%, 62.9%, and 61.3%, respectively. Considering factors such as solid recovery rate and sugar concentration, 2% NaOH was chosen for the pretreatment of dried vinegar residue obtained after starch recovery. In such conditions, the concentrations of total sugar, glucose, and xylose in the enzymatic hydrolysates reached 56.7 g L^{-1}, 28.0 g L^{-1}, and 15.9 g L^{-1}, respectively.

Effects of solid-to-liquid ratio on enzymatic degradation of dried vinegar residue. The effects of the solid-to-liquid ratio on enzymatic hydrolysis and saccharification were evaluated with 2% NaOH, temperature of 65 °C, and reaction time of 60 min. The results are shown in Figure 3. As the solid-to-liquid ratio decreased, the solid recovery rate also gradually decreased. This might be due to the fact that a higher concentration of NaOH increased the contact surface of the raw materials. The higher solid-to-liquid ratio

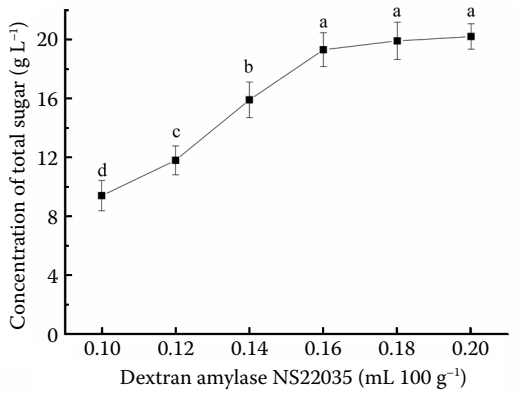

Figure 2. Effects of the amount of amylase (0.10 mL 100 g^{-1} to 0.20 mL 100 g^{-1}) on crude starch recovery from fresh vinegar residue carried out at 150 r min^{-1} and 50 °C for 72 h

Different letters mean significant difference ($P < 0.05$)

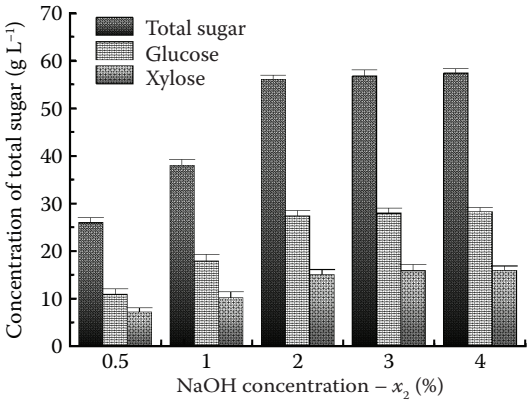

Figure 3. Effects of NaOH concentration (0.5–4.0%) on enzymatic degradation of dried vinegar residue evaluated with 1 : 10 (w/v) solid-to-liquid ratio at 65 °C for 60 min

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also resulted in a gradual increase in the concentrations of total sugar, glucose, and xylose in the enzymatic hydrolysate. As shown in Figure 4, the optimal overall effect was achieved when the solid-to-liquid ratio was 1 : 12 (w/v). The solid fraction recovery of this ratio was 65.3%, and the concentrations of total sugar, glucose, and xylose in the enzymatic hydrolysate reached 63.6 g L^{-1}, 38.2 g L^{-1}, and 13.4 g L^{-1}, respectively.

Effects of pretreatment time on enzymatic degradation of dried vinegar. The effects of pretreatment time on enzymatic hydrolysis and saccharification were evaluated with 2% NaOH, solid-to-liquid ratio of 1 : 12, and temperature of 65 °C (Figure 5). The prolongation of pretreatment time resulted in a gradual decrease in the solid recovery rate. This might be due to the better dissolutions of the raw materials with longer pretreatment time. As shown in Figure 5, the overall effect was better seen at a pretreatment time of 75 min: the solid fraction recovery was 66.7%, and the concentrations of total sugar, glucose, and xylose in the enzymatic hydrolysate reached 83.4 g L^{-1}, 47.8 g L^{-1}, and 16.5 g L^{-1}, respectively.

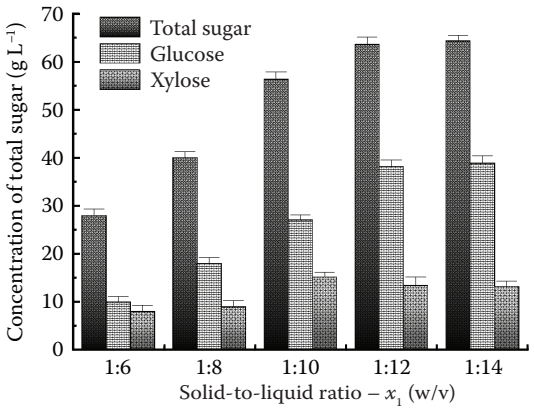

Figure 4. Effects of solid-to-liquid ratio [1 : 6–1 : 14 (w/v)] on enzymatic degradation of dried vinegar residue evaluated with 2% NaOH at 65 °C for 60 min

Effects of pretreatment temperature on enzymatic degradation of dried vinegar. The effects of pretreatment temperature on enzymatic hydrolysis and saccharification were evaluated with 2% NaOH, solid-to-liquid ratio of 1 : 12, and reaction time of 75 min (Figure 6). The increase in NaOH pretreatment temperature resulted in a gradual decrease in the solid recovery rate, indicating that a higher pretreatment temperature led to a better dissolution of lignin in the raw materials. The concentrations of total sugar, glucose, and xylose first increased, followed by a decrease. The overall effect was better seen at a pretreatment temperature of 65 °C: the solid fraction recovery was 67.4%, and the concentrations of total sugar, glucose, and xylose in the enzymatic hydrolysate reached 84.6 g L^{-1}, 48.2 g L^{-1}, and 16.6 g L^{-1}, respectively.

DPS analysis of NaOH pretreatment on enzymatic degradation of dried vinegar residue. Quadratic regression analyses were performed to determine the significance of the effects of each factor on the total sugar yield after enzymatic degradation of vinegar residue using results from the uniform design (Table 2) and DPS. The equation for the quadratic regression is shown below (Liu et al. 2011) in Equation 5:

$$y = 609.3 - 14.5x_1 + 3862.1x_4 + \\ + 25.2x_3 \times x_3 - 153.1x_4 \times x_4 - \\ - 453.9x_5 \times x_5 + 157.2x_2 \times x_5 \quad (5)$$

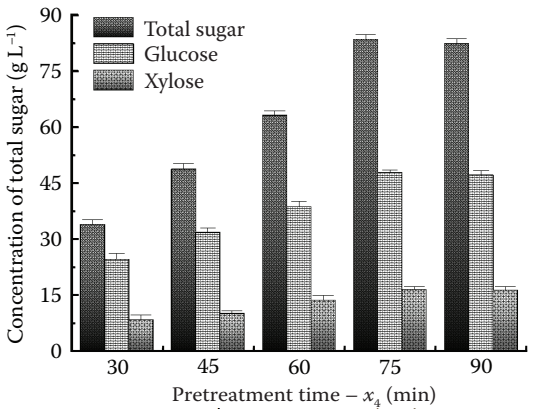

Figure 5. Effects of pretreatment time (30–90 min) on enzymatic degradation of dried vinegar residue evaluated with 2% NaOH and 1 : 12 (w/v) solid-to-liquid ratio at 65 °C

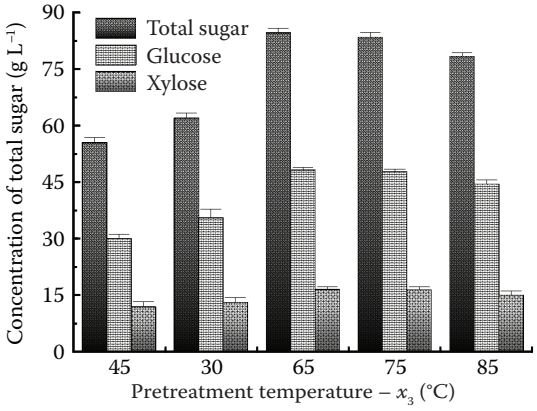

Figure 6. Effects of pretreatment temperature (45–85 °C) on enzymatic degradation of dried vinegar residue evaluated with 2% NaOH and 1 : 12 (w/v) solid-to-liquid ratio for 75 min

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where: x_1 – solid-to-liquid ratio; x_2 – NaOH concentration; x_3 – pretreatment temperature; x_4 – pretreatment time; x_5 – amount of xylanase.

The complex correlation coefficient R was 0.9947 and F statistic was 58.1341, with the significance value P of 0.0082 and standard deviation of 0.0092. The larger the values of the complex correlation coefficients R and F, the smaller the standard deviations, indicating a better fit between the data and the equation. The interactions between factor 2 and factor 5, i.e., interactions between NaOH concentrations and pretreatment time, can be seen from the regression equation. With higher NaOH concentrations, pretreatment time can be reduced. The most direct effect on total sugar yield was exerted by factor 5 – the amount of xylanase: a higher amount of enzymes leads to higher sugar yield. Table 4 shows the best combination of factors for the enzymatic degradation of vinegar residue based on the regression equation and DPS analyses. Under the condition of a solid-to-liquid ratio of 1 : 11, NaOH concentration of 2.2%, pretreatment temperature of 63 °C, pretreatment time of 80 min, and amount of 4.9 IU g^{-1} xylanase, the NaOH-pretreated, enzyme-degraded dried vinegar residue resulted in the concentrations of total sugar, glucose, and xylose reaching 89.6 g L^{-1}, 51.8 g L^{-1} and 15.3 g L^{-1}, respectively. Because the volume of the reaction solution was 200 mL and the weight of the vinegar residue after crude starch recovery was 27.1 g, the yield of total sugar, glucose and xylose was 66.1%, 38.2%, and 11.3%, respectively. After enzymatic degradation, the weight of the recovered solid was 16.2 g, the solid fraction recovery was 59.8%. In addition, 136 mL xylose solution was obtained, and the liquid fraction recovery was 68.0%; thus, the weight of xylose was 2.1 g.

Analysis of enzymatic degradation and SSF of vinegar residue. The enzyme complex and *S. cerevisiae* 1300 were added to the dried vinegar residue that was pretreated with NaOH solution and degraded by xylanase for SSF. Following the orthogonal experiment as shown in Table 5, the highest alcohol concentration was 54.1 g L^{-1}, which was carried out at 120 r min^{-1} and 37 °C for 120 h. After fermentation, the dry weight was 6.3 g (solid fraction recovery was 38.9%), the volume of the fermentation broth was 94 mL (liquid fraction recovery was 85.3%). So the weight of alcohol was 5.1 g and the yield of alcohol was 31.4%, which was higher than the yield of 28.7% achieved in the Chinese liquor residue (Liu et al. 2011). Meanwhile, the concentra-

Table 4. Combination of factors for the highest total sugar yield

y (%)	x_1 (w/v)	x_2 (%)	x_3 (°C)	x_4 (min)	x_5 (IU g^{-1})
66.1	1 : 11	2.2	63	80	4.9

Table 5. Orthogonal test and analysis of simultaneous saccharification and fermentation

Level	Rotational speed (r min^{-1})	Reaction time (h)	Reaction temperature (°C)	Alcohol concentration (g L^{-1})
1	100	24	31	47.6
2	100	48	34	50.5
3	100	72	37	49.9
4	120	24	37	52.2
5	120	48	31	54.1
6	120	72	34	51.9
7	140	24	34	52.5
8	140	48	37	51.8
9	140	72	31	50.6
k_1	49.333	50.767	50.433	–
k_2	52.733	52.133	51.100	–
k_3	51.633	50.800	52.167	–
R_j	3.400	1.366	1.734	–

Level 1–9 – the experiment number; k – average of the same number of experimental results; $R_j = k_{max} - k_{min}$

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tion of xylose was 9.8 g L^{-1} and this portion of sugar solution can be mixed with xylose solution obtained from the enzyme-degraded vinegar residue, so that the weight of xylose was 3.0 g and the yield of xylose can reach 18.5%, which can be used for other purposes, such as xylitol production (Rao et al. 2006; Zhang et al. 2012; Zhang et al. 2013).

CONCLUSION

Uniform experimental designs were used to investigate each of the 5 factors involved in the NaOH pretreatment of dried vinegar residue, including solid-to-liquid ratio, NaOH concentration, pretreatment temperature, pretreatment time, and the amount of xylanase. Using DPS analyses, the optimal pretreatment conditions for vinegar residue were determined, which were solid-to-liquid ratio of 1 : 11, NaOH concentration of 2.2%, pretreatment temperature of 63 °C, pretreatment time of 80 min, and amount of 4.9 IU g^{-1} xylanase. These optimal conditions allowed more effective enzymatic degradation of the vinegar residue and resulted in a total sugar yield of 66.1%. Subsequently, dried vinegar residue, cellulase and enzyme complex were added into the SSF process four times, and SSF reacted in a shaker at 120 r min^{-1} and 37 °C for 120 h. Following SSF, alcohol and xylose were produced, the yields of ethanol and xylose were 31.4% and 18.5%, respectively. Therefore, the method of Chinese vinegar residue for alcohol and xylose production by SSF was proved.

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