

BIOETHANOL PRODUCTION FROM RICE BRAN BY *SACCHAROMYCES CEREVISIAE*

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ABSTRACT

Carbohydrate biomass is considered as the future feedstock for bioethanol production because of its low cost and its huge availability. One of the major carbohydrate materials found in great quantities to be considered, especially in tropical countries, is rice bran. The effect of different pH and temperature with enzymatic saccharification treatment on conversion of rice bran was studied. The produced glucose was fermented to bioethanol, using *Saccharomyces cerevisiae* yeast and the amount of produced bioethanol was measured by gas chromatography. Enzyme treatment at 60°C and pH 5 is an effective treatment method for converting rice bran to glucose. Up to 22.75% glucose v/v could be achieved after enzyme treatment. Fermentation of treated rice bran shown that glucose after 3 days fermentation the maximum bioethanol of 17.31% (v/v) was attained. This process is expected to be useful for the bioethanol production from rice bran as a source of carbohydrate renewable biomass from abundant agricultural by product.

Keywords: Rice bran, enzymatic hydrolysis, fermentation, bioethanol

1. INTRODUCTION

Currently, efforts have increased towards commercial production of bioethanol, considered the most promising from biomass, and it is well known that a low cost feedstock is a very important factor in establishing a cost-effective technology (Rabelo *et al.*, 2009). Rice bran (*Oryza sativa*) material as agricultural residue is one of the most interesting substrates, potential source of sugars for bioethanol production. In recent years, a large number of studies regarding the utilization of biomass as a feed stock for producing bioethanol are being carried out. A number of processes based on the application of enzymes for bioethanol production from rice washing drainage and rice bran biomass have been proposed (Watanabe *et al.*, 2009). Great attention is focused on renewable sources in bioethanol production.

Rice bran (rice milling by-product) production in Indonesia is 50.7 million tons/year and consist of a 8-10% of total paddy weight. Due to its low lignin content compared to other biological waste products, milled and defatted rice bran (27% w/w cellulose, 37 % w/w hemicellulose, 5 % w/w lignin) is an interesting representative of the group of lignocellulose biomass (Hernandez, 2005), defatted rice bran is mainly used as nutritional additive for cattle feed.

According to Reddy and Reddy (2005, 2006) *Saccharomyces cerevisiae* is one of bioethanol producing organisms used in industrial processes. Under very high gravity conditions if appropriate environment and all required nutrients in adequate amounts were provided, *S. cerevisiae* could ferment increased amount of sugars in the medium. Thomas *et al* (1996) and Bafnrcová *et al* (1999) reported by that under appropriate environment and nutritional condition, *S. cerevisiae* could produce and tolerate high ethanol concentrations.

This research aimed to find out the effect of pH and temperature with enzymatic saccharification treatment on conversion of rice bran to glucose and fermentation of glucose to bioethanol *S. cerevisiae* yeast.

2. MATERIALS AND METHODS

2.1. Material

Rice bran has been furnished by public huller in Surakarta. Pretreatment method contained milling (particle size <180 μ m). Glucose was obtained from hydrolysis of rice bran from Surakarta regency. After the hydrolysis was kept at 18°C until use (Khongsay, *et. al.*, 2010).

2.2. Methods

2.2.1 Enzyme pre-treatment

150 g rice bran was suspended in 500 mL H₂O in ratio of 3:10 (w/v) rice bran and added of 0.1 mL of α -amylase enzyme. The pH of sample was adjusted at pH 5, 5.5, and 6. The sample was incubated in water bath 100°C for 30 minutes, after that the mixture was applied for second enzymatic treatment (0.2 ml of glucoamylase). Finally, hydrolysate was pressed through cheese cloth. The amount of reducing sugar in juice was measured.

2.2.2 Fermentation

The pretreated samples from 2.2.1 was carried out for fermentation experiments. The yeast *S. cerevisiae* was used for fermentation (1.5g, 3.0g, and 4.5g). After 3 fermentation days the ethanol content was measured by gas chromatography. All the measurements were duplicated and the data reported are average of two replications.

3. RESULTS AND DISCUSSION

3.1. Effect of enzyme pre-treatment methods on glucose content of rice bran

Table 1. Effect of pH on glucose content of rice bran

| pH | Temperature (°C) | Glucose (%) |
|-----|------------------|-------------|
| 5 | 60 | 22.75 |
| 5.5 | 60 | 21.80 |
| 6 | 60 | 21.00 |
| 5 | 70 | 21.70 |
| 5.5 | 70 | 20.69 |
| 6 | 70 | 19.72 |

In the Figure 1 the relation between glucose concentration and pH is depicted. Increasing pH showed reverse effect on glucose concentration in sample. This is expected because of conversion of carbohydrate to glucose (Yoswathana *et al.*, 2009). The highest glucose up to 22.75 % glucose on the rice bran basis could be obtained on pH 5.

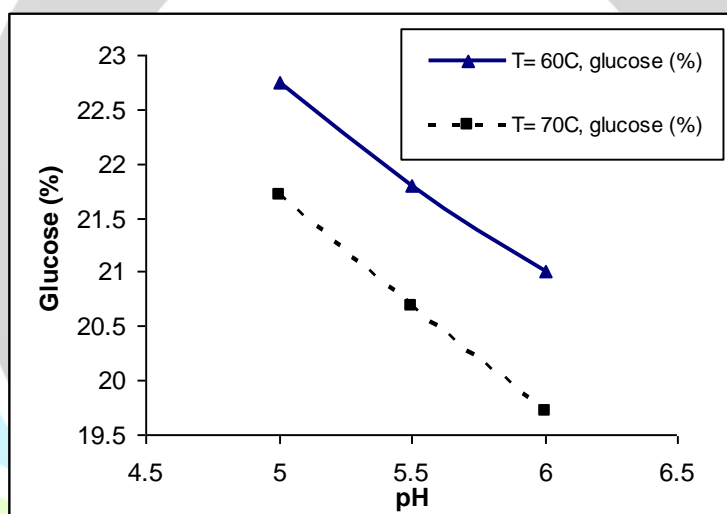


Figure 1. Glucose content in enzyme pretreated rice bran

3.2. Fermentation

Table 2. Production of bioethanol from rice bran

| pH | T (°C) | Rice Bran (g) | Sugar (%) | <i>S.cerevisiae</i> (g) | Ethanol (%) |
|----|--------|---------------|-----------|-------------------------|-------------|
| 5 | 60 | 150 | 22.75 | 1.5 | 10.05 |
| 5 | 60 | 150 | 22.75 | 3 | 15.92 |
| 5 | 60 | 150 | 22.75 | 4.5 | 17.31 |

| | | | | | |
|-----|----|-----|-------|-----|-------|
| 5.5 | 60 | 150 | 21.80 | 1.5 | 8.52 |
| 5.5 | 60 | 150 | 21.80 | 3 | 10.48 |
| 5.5 | 60 | 150 | 21.80 | 4.5 | 13.57 |
| 6 | 60 | 150 | 21 | 1.5 | 10.53 |
| 6 | 60 | 150 | 21 | 3 | 11.62 |
| 6 | 60 | 150 | 21 | 4.5 | 11.93 |

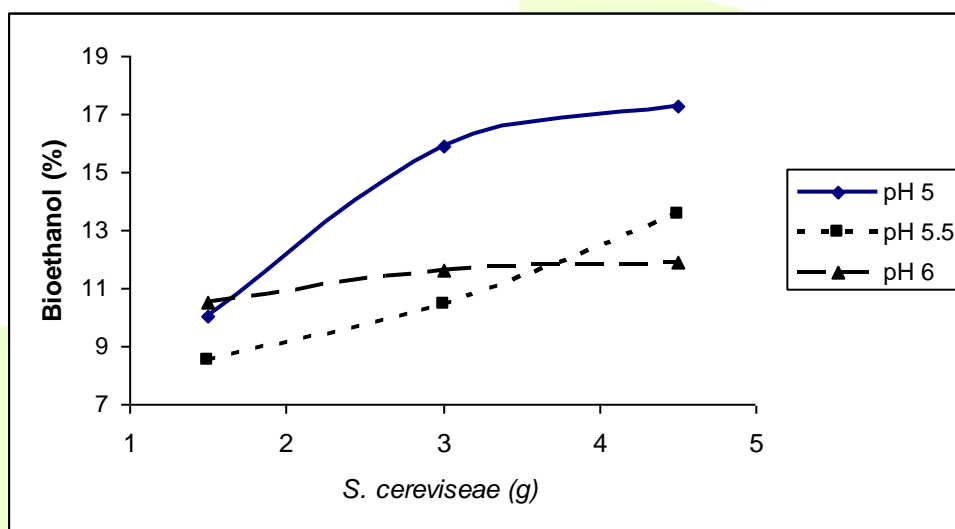


Figure 2. Bioethanol contain in *S. cerevisiae* at temperature 60°C

Bioethanol was produced by using pH and *S. cerevisiae* concentration treatment. These were 1.5g, 3.0g, and 4.5g. It can be seen in Figure 2 that pH 5.0 tended to give higher ethanol concentration than that in other pH, therefore, pH 5.0 was chosen for ethanol fermentation. This was supported by Thuesombat *et al.* (2007). A look at the results of rice bran, production of bioethanol in Table 2 shows that pH 5 at temperature 60°C had higher quantities of bioethanol produced and bioethanol content of 17.31% (v/v). This confirm that the higher sugar content in the juice, the more bioethanol can be produce (Woods, 2001 and Yamba *et al.*, 2007).

It was observed that the sugar content was directly proportional to the quantities of the ethanol collected. Therefore, the higher the sugar content, the more the ethanol can be produced (Yamba *et al.*, 2007).

4. CONCLUSIONS

It can be concluded that it is possible to successively use rice bran for bioethanol. Enzyme treatment at 60°C and pH 5 is an effective treatment method for converting rice bran to glucose. Up to 22.75% glucose v/v could be achieved after enzyme treatment. Fermentation of treated rice bran shown that glucose

after 3 days fermentation the maximum bioethanol of 17.31% (v/v) was attained. Production of bioethanol has been considered to be one of economical way for the utilization of rice bran. So the manufacture of glucose could easily be undertaken as an additional source of income.

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