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Optimization of Glucose from *Saccaromycess cerevicae* Liquid Waste Using the Acid Hydrolysis Process

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*Corresponding author: E-mail:	ABSTRACT
ketutsari.tk@upnjatim.ac.id	Selection of <i>Saccharomyces Cereviceae</i> (SC) wastewater as a component produced by PT. Enero comes from a fermentation process that uses SC yeast, it's availability is great and it has not been used as a raw material for glucose, which until now has been industrial waste. Efforts have been made to convert SC waste water into liquid fertilizer and biodiesel feedstock. The recovery of glucose from SC waste water in the chemical hydrolysis process using the reaction surface method (RSM), the hydrolysis process was carried out with a digital stirrer at a rotation speed of 200 rpm and the addition of 5 ml of 10% NaOH. The variables used were 50–250 mL SC waste water and 10–30 mL HCl volume. The optimization results using RSM of the chemical hydrolysis process are shown in three-dimensional graphs, pH and SC waste water ratio and HCl volume, glucose content and SC waste water ratio and HCl volume. The best results with SC waste volume of 8.58 mL and HCl volume of 34.14 mL achieved a pH of 4.38. In order to achieve a pH of 4.5, a hydrolysis process is carried out because the pH. The result of glucose from the hydrolysis process is 17.96%, the required volume of waste water SC of 137.14 ml, the addition of 34.14 mL of HCl as a raw material for the fermentation process. Based on the optimization results using RSM and chemical hydrolysis method, the glucose content is 17.96% and the pH is 4.38 with the SC waste waste volume of 137.14 mL and the HCl volume of 34.14 mL.
	Keywords: Acid hydrolysis, glucose, waste water, optimization, Saccaromycess cereviceae

Introduction

Saccharomyces Cereviceae waste water is a byproduct of the fermentation process, whereas in the fermentation process using SC yeast, Yeast movements were performed monthly to produce SC waste water, availability of liquid waste SC and PT. Power is limited abundant and unused, which is a waste of industry, effort was designed to convert SC wastewater into liquid fertilizer and biodiesel raw material, glucose content is 15.5%, protein 1.15%, starch 10.28%, carbohydrates 42.4%, crude fiber 7.73% and impurities 14.3% (N, P, K, Ca, mg, S) (PT. Enero, 2023). A number of steps are taken bioethanol production process, cellulose pretreatment process using sulfur the acid process and delignification use strong points that can damage lignin level of 24.7% and produce a cellulose level of 49% at a temperature of 100 0C for minutes (90-120), the continuation of the hydrolysis process using cellulose. enzymes at a temperature of 45 0C, and 200 rpm for 25 minutes to produce a glucose level of 9% (Kuhad et al., 2010). Hydrolysis process Liquid rice flour using HCl, the glucose level was obtained at 9.98%, From elephant grass, a glucose level of 37.9% is obtained and from bamboo, glucose levels were found to be 23.6% (Sari et al., 2017). Hydrolysis The process of organic bamboo uses cellulose enzymes that contain a temperature microcontroller, time 137 minutes and temperature 97 0C,

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obtained Glucose content is 23.6%, optimization of results using Proportional Integral Derive (PID) in the Delphi programming language (Sari et al., 2018), using Matlab language and object-oriented programming language (Sari et al., 2013).

The first treatment can increase blood sugar after it is done Through the process of enzymatic saccharification, which can increase the efficiency throughout the process, the conversion of glucose to xylose and ethanol requires new color technology for the production of bioethanol from agricultural products waste (Sarkar et al., 2012). Bioethanol production from waste sago pith uses microwave hydrothermal hydrolysis, maximum of 43.8% theoretical glucose and 40.5% ethanol production, a process developed to save energy without using enzymes, but use of acid or alkali (Saravana et al., 2014). ferment acid from waste leaves containing Pichia stipitis led to the production of ethanol 3.73 \pm 0.16 g/l and 77.54 \pm 4.47% (Alok et al., 2012). Bioethanol studies production from rice flour, in the hydrolysis process using bacilli produces 5-10% glucose, fermentation process using *Saccharomyces cereviceae* producing 11 to 16% ethanol (Sari et al., 2016).

The production of glucose from SC wastewater using acid hydrolysis method, the variables used are the volume of SC wastewater and the amount of HCl, NaOH was used first, and the hydrolysis process with HCl reversed. starch into glucose. Development of digital tools for hydrolysis processes and data processing improvement using Minitab 18 and RSM method.

Material and Methods

Starch preparation in the hydrolysis process

Starch is a more complex component than disaccharides, before the fermentation process is carried out, starch must be broken down using the amylase enzyme into a disaccharide component in the form of Maltose, using the Maltase enzyme, and the maltose will be hydrolyzed into glucose. The hydrolysis process is influenced by several factors, including the amount of content. The hydrolysis process is influenced by several factors, including the amount of starch content in the raw material, the degree of acidity (pH), the acid concentration used, the hydrolysis time, the hydrolysis temperature, and the catalyst (Sari et al., 2016).

The result of the fermentation process is usually a dilute alcohol solution because yeast cells will die if the ethanol content exceeds 12-15%. Dilute ethanol from the fermentation process is subjected to a distillation process to obtain purer ethanol, the ideal is 51.1% ethanol and 48.9% carbon dioxide, the optimum is 48.8% ethyl alcohol, 46.6% carbon dioxide, 3.3% glycerol, succinic acid 0.6%, cellulose and so on 1.2% (Sarkar et al., 2012). Other factors that influence the fermentation process include pH, a good pH for the fermentation process is between pH 4 - 5, because lactic acid is good for yeast growth, but the downside is that it can grow butyric acid bacteria, which can be detrimental to yeast growth (Sumiyati & Sutrisno, 2009). The time required for the fermentation process depends on the temperature and sugar concentration, generally, the time required is between 36-50 hours, generally a good temperature for the fermentation process is between 25-30 °C, at low temperatures the alcohol will be lost, carried away by carbon dioxide gas (Sari & Purbasari, 2022).

Within the species Saccharomyces cerevisiae there are many different strains, which have very different characteristics, and produce a variety of flavors. The Saccharomyces cerevisiae strain in the Alcotec 48 Turbo yeast product has the ability to ferment up to a very high alcohol percentage (20%), and has a tolerance for alcohol levels of 8-20% under optimum conditions. Alcotec 48 Turbo yeast is used to make alcohol from fruit, molasses, and grains. The composition of Alcotec 48 Turbo yeast is Alcotec TT dry yeast 54%, carbamide 23.5%, phosphates 15%, sulphates 4%, carbonates 2%, vitamins 1%, trace minerals 0.5%. The advantages of Alcotec 48 Turbo yeast produces alcohol levels higher than 20%, works relatively quickly, and can withstand quite high temperatures (Kumar et al., 2009).

Results and Discussion

Hydrolysis process and data processing optimization

The initial glucose content from SC waste (*Limbah* SC) is 15%, which can be used as raw material for the fermentation process. The initial substrate pH is around 3.5. To reach a pH of 4.5, a pretreatment process is carried out by adding NaOH. The larger the volume of SC liquid waste, the more NaOH is added. In a variable volume of 500 mL to 2500 mL of SC liquid waste and a variable volume of HCl of 5 mL to 25 mL. The greater the volume of NaOH added, the more significant the increase in pH. In Figure 1, the optimum glucose content from the hydrolysis process is 17.96%, the SC wastewater volume is 137.14 mL, and the HCl volume is 34.14 mL. The pH at the end of the process decreased due to the bleaching process with the addition of a small amount of powdered activated charcoal and also due to the filtration process.

The influence of pH in the pretreatment process greatly determines the results of glucose levels in the hydrolysis process. For the volume of liquid flour waste between 200-600 ml, it shows an optimum pH of pH 7.1. The effect of pH on the increase in the volume of Bacillus, where the greater the increase in the volume of Bacillus, the smaller the pH. Because the fermentation process requires a pH of 4.5, increasing the Bacillus volume by 7% is the closest, for varying waste volumes (Sari et al., 2014).



Figure 1 Contour plot of pH, HCl, and SC waste for 2-dimensional graphs, and pH, HCl, and SC waste for 3-dimensional graphs

The results of the pretreatment process are used as the basis for the starch hydrolysis process, the process of breaking down starch into a simpler glucose structure, which is carried out using an acid. In the starch hydrolysis process, there are several stages, namely gelatinization, liquefaction, and saccharification. At all stages, use heating with a water bath so that the hydrolysis process takes place evenly. Acid hydrolysis of starch aims to break down starch into its simpler constituent parts such as dextrin, isomaltose, and glucose. According to previous researchers, the starch used came from solid waste tapioca flour which has a starch content of 76.055% consisting of amylose of 15.8429% and amylopectin of 60.2121%. The gelatinase process for tapioca flour is 52-64 OC, and the temperature of the substrate solution reaches around 50 OC, or when the substrate begins to thicken. The temperature of the substrate solution should not be too high, because if the temperature is too high the substrate solution process difficult, the enzyme added will not work optimally because there is a structural change in the starch which causes the substrate solution to become very thick, and the water absorbed only reaches 30% (Sari et al., 2018).



Figure 2. Multiple response prediction from SC Waste, HCl, and pH

Figure 2, it shows the predicted response from the hydrolysis process, the pH is 4.381, the SC waste water volume is 85.7864 mL, the HCl volume is 29.1421 mL, and the standard deviation (d) is 0.78702, which is declared to meet the requirements for two and three dimensional contour plots for determining glucose levels in the hydrolysis process.

Regression Equation in Un-coded Units:

pH = 4.340 - 0.000790 waste SC - 0.0278 HCl + 0.000000 waste SC* waste SC + 0.001075 HCl*HCl + 0.000001 waste SC*HCl

The regression equation a 2nd order polynomial equation was obtained, with pH as the response, and SC waste and HCl as variables.

Conclusion

The optimum glucose content results from the hydrolysis process using RMS were 17.96%, a volume of SC waste water was required of 137.14 mL, and an additional volume of HCl of 34.14 mL. used as raw material in the fermentation process. From the optimization results using RSM in the chemical hydrolysis process, the glucose content was 17.96%, pH 4.38, SC waste water volume 137.14 mL, and HCl 34.14 mL. From SC waste the initial glucose content is 15% and after processing by hydrolysis the glucose content is 17.96%, which can be used as raw material for ethanol production using the fermentation process.

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