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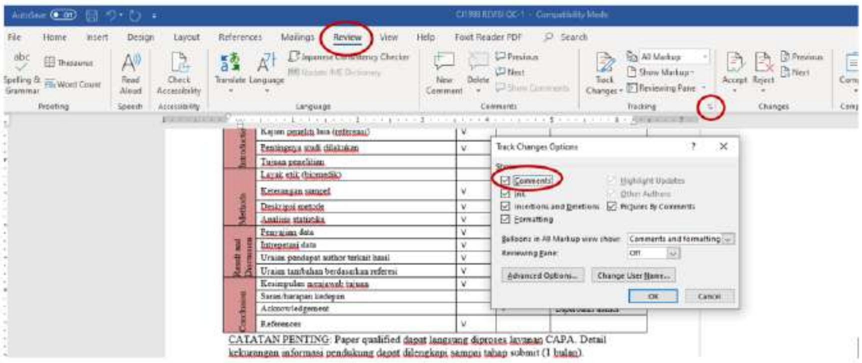

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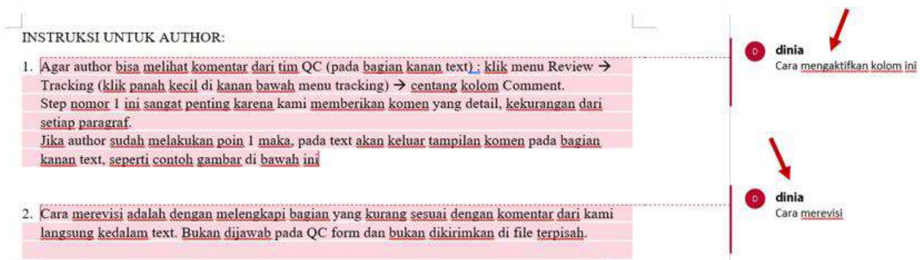

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Proces Fermentation of Filtrate Bamboo with Saccharomyces Cerevisiae and Zymomonas Mobilis

Ni Ketut Sari[1*], [2]Sofi Bachtiar, [3]Wahyuningtiyas, [4]Intan Purbasari Yuniar

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Abstract. Fermentation is the process of the formation of ethanol from glucose by using enzymes. Bamboo is one of the materials containing glucose is high enough, that is previously done hydrolysis in advance. Bamboo used when the hydrolysis process of bamboo that does not include lignin and the pentose done process of pretreatment and not lignification. The purpose of this research is to produce ethanol as a raw material substitution of bioethanol, knowing pentose and dirt left in the bamboo. Therefore, need to be studied in the future, with the best process, that we used biological processes that can optimize the production of ethanol. The use of the enzyme (Saccharomyces Cerevisiae and Zymomonas Mobilis) is also significant because of the optimum enzyme conditions. Temperature, pH, and the yeast with optimal conditions when it can raise the level of his work. The fermentation process at temperature 25 C and 45 C, the filtrate is 500 ml solution of bamboo and the stirring speed of 200 rpm. The variable composed enzyme with a ratio (v/v) of 0.25 to 0.75. Resulting from the fermentation processed can produce ethanol with a yield 30.5% and 36% of the weight of the bamboo. The result of the process of fermentation obtained bioethanol with low ethanol yield of 10-15%, which requires the flash distillation process to obtain yield bioethanol technical 90-95%.

Introduction

~~Biomass from plants has been declared as an alternative raw material for gasoline fuel substitution in the form of bioethanol, bioethanol obtained from biomass and bioenergy crops has proclaimed as one of the feasible alternatives as gasoline fuel~~ Bioethanol has been proclaimed as one of the feasible alternatives for gasoline fuel substitution. Bioethanol is mostly obtained from biomass, especially from plant [1]. Sustainable bioethanol from rice straw Bioetanol bisa diperoleh dari bahan baku batang padi [2]. ~~Ethanol~~ The production of ethanol is usually from lignocellulose by chemical hydrolysis process ~~chemically~~ or enzymatically. ~~,~~ The first step is ~~conducted the process of~~ pre-treatment and ~~,~~ the next process is the ~~process of~~ fermentation and distillation process. In pre-treatment processes, the most important is to remove the lignin and pentosane, which can dissuade the lignin and pentosane. Various approaches have been performed earlier as pre-treatment in acids, bases, ammonia, sodium chlorite, and biological [3].

Thee research was conducted to evaluate acid pretreatment from hydroxide paper waste as material for bioethanol production by~~,~~ optimization of~~zed~~ sulfuric acid hydrolysis, while the fermentation process of hydroxide acid of paper waste by using Pichia Stipites. The ethanol content was obtained at 77.54%. By one more distillation process, the ethanol content received at the level of 95-96% [4]. Chemical pretreatment of lignocellulose biomass ~~with~~ using s~~S~~ulphur (H_2SO_4) and phosphorus (H_3PO_4) acids are commonly used since they are relatively cheap and efficient in hydrolyzing lignocellulose, though the letter gives a milder effect and is more benign to the environment penggunaan s~~S~~ulphur (H_2SO_4) and phosphorus (H_3PO_4) lebih ramah lingkungan. Hydrochloric acid is more volatile and more natural to recover and attacks biomass better than H_2SO_4 [5]. Similarly, nitric acid (HNO_3) possesses good cellulose to sugar conversion rates [6]. However, both acids are expensive compared to s~~S~~ulphur acid kestabilan atau jumlah H+ pada H_2SO_4 lebih banyak dibanding HCl dan HNO_3 pada konsentrasi yang sama. Selain itu, H_2SO_4 mempunyai pengaruh anion dan faktor disosiasi asam yang lebih tinggi dibandingkan HCl dan HNO_3.

. ~~Pretreatment of lignocellulose has received considerable research globally due to its affluence on the technical, economic, and environmental sustainability of cellulose ethanol production. This paper reviews know, and emerging chemical pretreatment methods, the combination of chemical pretreatment with other ways to improve carbohydrate preservation reduce formation to degradation product, achieve high sugar yield at mild reaction conditions, reduce solvent loads and enzyme dose, reduce waste generation~~ Pretreatment of lignocellulose has attracted many researchers due to its influence on the technical, economic, and environmental sustainability of cellulose ethanol production. This paper reviews knowledge and chemical pretreatment method and also the combination of chemical pretreatment to escalate the carbohydrate preservation, reduce the degradation product, obtain a high yield of sugar in a mild condition of the reaction, reduce the solvent loaded and enzyme dosage, and also reduce the waste. [7]. Initiatives of the future for lignin in biomass to bioethanol, pretreatment technology to separate the main tree biopolymers (cellulose, hemicellulose, and lignin) Selanjutnya untuk memperoleh bioethanol dari bahan baku biomassa, dilakukan proses pretreatmen untuk menghilangkan lignin serta memisahkan biopolimer (selulosa, hemicellulose, dan lignin) dari bahan baku biomassa [9]. ~~Pretreatment for hydrogen and bioethanol production from olive oil~~

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Author dapat menggunakan alasan kestabilan atau jumlah H+ pada H2SO4 lebih banyak disbanding HCl dan HNO3 pada konsentrasi yang sama. Selain itu, pengaruh anion atau bisa juga dari faktor disosiasi asam.

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~~waste products was ethanol yield 5.4 % treatment with 1.75 w/v Sulphur acid and heated it at 140 ºC for 10 min, and was ethanol yield 5.0 % no pretreatment~~ Pretreatment for production of bioethanol from olive oil waste that used sulfuric acid and heating at 140°C for 10 min result in ethanol 5.4 %. This yield was higher than ethanol obtained without pretreatment, about 5.0 % [10]. Pretreatment followed with simultaneous scarification and fermentation on bioconversion of microcrystalline cellulose for bioethanol production, the yield value of 67 % bioethanol bioconversion apabila pretreatment dilanjutkan dengan proses skarifikasi dan fermentasi maka akan menghasilkan rendemen 67% [11]. A sustainable feedstock bioethanol production, cellulose hydrolysis was microwave irradiation using hydrochloric acid as catalyst, fermentation with yeast (*Saccharomyces cerevisiae*), modest reaction conditions (2.38 M acid concentration), irradiation time 7 min, and yield of 0~~,~~.67 g glucose /~~-~~g cellulose [12]. Elements contained in the lignocellulose biomass of the plants are usually used lignocellulose biomass, a potential for bioethanol production globally Tanaman yang termasuk biomassa lignocellulose berpotensi menjadi bioetanol. Agriculture (softwood), forestry (pretreatment method obtained ethanol content below 16% Bahan baku dari Pertanian (kayu lunak), kehutanan (kayu keras) berpotensi menghasilkan ethanol dibawah 16%, dengan melakukan proses pretreatment. Pretreatment with dilute acid (sulfuric acid) ~~eliminating~~ aims to eliminate the hemicellulose components and increase the sugar. The pretreatment method of sulfuric acid has ~~long~~ been recognized as an important step towards eliminating the hemicellulose fraction of lignocellulose substrates and saving~~e~~ the conversion of cellulosic biomass [20]. The research conducted by [21] about ethanol production from sago pith waste (SPW) using microwave hydrothermal hydrolysis catalyzed by carbon dioxide~~,~~ resulted in a higher energy saving compared to previous techniques in the absence of enzymes, acid, and base catalyst. They obtained ethanol content below 15.6%. The production of bioethanol from lingo-cellulosic biomass through ~~process~~ several steps, such as pretreatment, hydrolysis, fermentation, distillation [26]. Ethanol production from lingo-cellulosic technologies is largely determined by hydrolysis and pretreatment, ~~whether~~ either chemical or biological [27].

Ethanol from the liquid waste of rising flour using fermentation by Saccharomyces obtained~~,~~ a maximum of 23.8% glucose and 40.5% ethanol yield.~~,~~ ~~t~~This~~e~~ developed technique for a liquid waste of rising flour resulted in higher energy saving compared to the previous method in the absence of enzymes, acid or base catalyst [28]. Bioethanol dari bahan baku biomassa (hardwood), and industrial waste are significant lignocellulose biomass for bioethanol production. The lignocellulose biomass is one of the potential main sources for economic bioethanol production globally. Agricultural, forestry (soft and hardwoods), and industrial wastes are the major lignocellulose biomasses [13]. The bioethanol production from lignocellulose biomass by using ~~process~~ pretreatment, hydrolysis, fermentation, and recovery of ethanol~~, was obtained~~resulted in ~~by~~ ethanol ~~under~~ less than 16% v/v, meanwhile~~,~~ ~~with~~ by the distillation process ~~will again be derived~~would obtain ethanol 95-96% v/v. The research conducted bioethanol production from lignocellulose biomass by using the pretreatment process, hydrolysis, fermentation, and ethanol recovery. Therefore, ethanol content obtained in the level below 16%, and by one more distillation process the ethanol content would receive of 95-

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apabila pretreatmemn dilanjutkan dengan proses skarifikasi dan fermentasi maka akan menghasilkan rendemen 67%

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96% v/v Maksudnya disini (hanya 1 kali distilasi), proses fermentasi menghasilkan kadar ethanol dibawah 16%, pada proses distilasi menghasilkan kadar ethanol 95-96% [18]. The research conducted by [19] about bioethanol production from agricultural waste using PROFER Cellulosic or second-generation (SG) bioethanol produced from lingo-cellulosic biomass (LB) ~~in~~ involved three main steps: pretreatment, hydrolysis, and fermentation. Pretreatment involves the use of physical processes, chemical ~~methods~~process, physico-chemical processes, biological ~~methods~~process, and several combinations thereof to fractionate the lignocellulose into its components. It ~~results in~~will lead to the disruption of lignin seal to increase enzyme access to cellulose [29, 30], reduction of cellulose crystallinity [31, 32], an increase in the surface area [33, 34] and porosity [35, 36] of pretreated substrates, resulting in increased hydrolysis rate. In hydrolysis, cellulose and hemicelluloses are broken down into monomeric sugars via the addition of acids or enzymes such as cellulose. Enzymatic hydrolysis offers advantages over acids such as low energy consumption due to the mild process requirement, high sugar yield, and no unwanted wastes. Enzymatic hydrolysis of cellulose affected by properties of the substrate such as porosity, cellulose fiber crystallinity, and degree of polymerization, as well as lignin and hemicellulose content [37, 38], optimum mixing [39], substrate and end-product concentration, enzyme activity, reaction conditions such as pH and temperature [40, 41].

The research that will be carried out is the development [42] on the manufacture of bioethanol from wheat flour liquid waste, wherein this study ~~on hydrolysis process~~ used H_2SO_4 catalysts in hydrolysis process and Turbo Yeast 48 in the process of fermentation ~~of yeast~~ that has a better quality~~, namely.~~. ~~Turbo Yeast 48 so that~~Thus, it is expected to get higher ethanol content results. Turbo yeast is a blend of dry wine yeast (*Saccharomyces* ~~C~~c*erevisiae*) and nutrients optimized to provide the ~~right~~ best combination of nitrogen, vitamins, and trace minerals that yeast needs in different stages of alcohol fermentation. In this study, bioethanol levels from wheat flour liquid waste and optimum time in the fermentation process were~~,~~ examined, as well as the influence of optimum Turbo Yeast levels on ethanol levels produced.

Penelitian ini untuk memperoleh kadar bioethanol yang optimum dengan proses hidrolisis menggunakan katalis H_2SO_4 dan proses fermentasi menggunakan Turbo Yeast 48. Tujuan penelitian ini untuk mencari kadar bioethanol optimum terhadap pengaruh kadar Turbo Yeast 48 dan waktu fermentasi dengan *response surface methodology*. Urgensi penelitian pertama adalah dalam proses fermentasi selama ini diperoleh kadar bioethanol dibawah 16%, dengan menggunakan turbo yeast 48 memungkinkan diperoleh kadar bioethanol 25-40%. Urgensi penelitian kedua adalah optimalisasi kadar bioethanol terhadap pengaruh 2 variabel (kadar Turbo Yeast 48 dan waktu fermentasi) menggunakan grafik 3 dimensi, dengan metode *response surface methodology*. Batasan penelitian pertama pada hasil eksperimen dengan proses hidrolisis menggunakan katalis H_2SO_4 dengan kadar 5 %v/v, menghasilkan filtrate glucose dengan kadar 11 %. Batasan penelitian kedua pada hasil eksperimen dengan proses fermentasi menggunakan Turbo Yeast 48 dan waktu fermentasi menghasilkan bioethanol dengan kadar 37 %. Batasan penelitian ketiga, dari data yang diperoleh pada proses fermentasi, dibuat grafik tiga dimensi, yang terdiri dari : sumbu x (kadar turbo yeast 48), sumbu y (kadar bioethanol)

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one more distillation di sini maksudnya re-distilasi atau hanya 1 kali distilasi?

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dan sumbu z (waktu fermentasi). Kebaruan penelitian

Literature review

The hydrolysis process changes starch to monosaccharides (glucose) with ~~the help of~~ H_2SO_4 catalysts through the hydrolysis process. can be separated from the mixture with the addition of alkali such as calcium, so that it can be deposited in the form of calcium sulfate. The rate of hydrolysis process will be increased by the high concentration of acids.~~, using~~ H_2SO_4 catalysts provide greater levels kadar asamnya than the use of hydrochloric acid ~~catalysts,~~ because H_2SO_4 ~~has several~~offers more H+ ions than HCl so the reaction ~~speed~~ rate ~~is increasing~~will increase and provide~~s~~ a greater product of hydrolysis results. ~~With~~ In the same concentration of different catalysts, both H_2SO_4 and hydrochloric acid have the same amount of water, but H_2SO_4 has more H+ ions than hydrochloric acid resulting in better ~~lasting~~ bond disconnection ~~better~~. T~~t~~he highest furfural yield ~~is~~ obtained ~~at~~in a 1% H_2SO_4 catalyst concentration, and hydrolysis time of one hour [43].

Several f~~F~~actors that influence the hydrolysis process are: pH is very influential on acid concentration and hydrolysis.~~,~~ I~~i~~f the acid concentration is high, then ~~the resulting~~ pH is low.~~,~~ ~~the pH is good~~The optimum pH for the hydrolysis process that is at pH 4.5 [44]. To speed up the course of the reaction, the hydrolytic reaction requires a catalyst~~almost all hydrolytic reactions require a catalyst~~. The catalyst used can be either enzymes or acids, because it works faster secara proses biologi menggunakan enzim (Bacillus) atau secara proses kimia (HCl). ~~Acids used range from hydrochloric acid, Factors that influence the hydrolysis process are: pH is very influential on acid concentration and hydrolysis, if the acid concentration is high, then the resulting pH is low, the pH is good for the hydrolysis process that is at pH 4.5. To speed up the course of the reaction, almost all hydrolytic reactions require a catalyst. The catalyst used can be either enzymes or acids, because it works faster. Acids used range from hydrochloric acid, Factors that influence the hydrolysis process are: pH is very influential on acid concentration and hydrolysis, if the acid concentration is high, then the resulting pH is low, the pH is good for the hydrolysis process that is at pH 4.5 [44]. To speed up the course of the reaction, almost all hydrolytic reactions require a catalyst. The catalyst used can be either enzymes or acids, because it works faster.~~ The a~~A~~cids used range from hydrochloric acid (HCl), sulfuric acid (H_2SO_4), to nitric acid (HNO_3). The concentration of ~~H ions~~the acids also affects the reaction speed~~, so it's not because of the type of acid.~~ Generally, a high concentration of acids is used.~~ used acid solution that has a higher acid concentration.~~ The effect of temperature on reaction ~~speed~~ rate follows Arrhenius' equation that is, the higher the temperature, the faster the reaction. The ~~speed~~ rate of hydrolysis reaction will increase almost 2 times for each temperature rise of 100 ^{0}C Tingkat reaksi hidrolisis akan meningkat hampir 2 kali lipat pada suhu 100 ^{0}C. For the reagents to collide with each other properly, it is necessary to stir. For batch processes, this can be achieved with the help of a stirrer or shaker [43]. The longer the hydrolysis time, the greater the concentration of glucose produced. At low levels of use, the balance will shift to the right ~~well~~. High suspension levels result in the increase of~~d~~ viscosity of the mixture, resulting in an increasing amount of insoluble starch particle content. This ~~results in~~lead to the

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hydrolysis process ~~not being able to~~cannot run properly or perfectly. The more starch suspension levels are hydrolyzed, the longer the processing time needed to hydrolyze~~sis~~ the starch [45].

Batch fermentation is a closed system, no addition of new media, no addition (O_2), antifoam, acid or base is carried out through using pH control. Batch fermentation is widely used in the industrial world to produce ethanol due to the ease in the sterilization and control process of tools. But there are some drawbacks of batch fermentation that is, obstacles due to high sugar content, the limited concentration of ethanol yield (12%), and low productivity. However, batch fermentation is still an option because the yield obtained is higher compared to other methods. In the ethanol production with batch process, microorganisms work in high concentrations of substrates at first and high concentrations of products eventually. Generally, batch fermentation is characterized by low productivity. Microbes that can be used are *Saccharomyces* ~~C~~*cerevisiae*, ~~which~~ that are the best microbes for ethanol fermentation because they are relatively more efficient at converting sugar into ethanol and are more tolerant to ethanol [46]. *Saccharomyces* c~~C~~*erevisiae* microorganisms have several brands circulating in the community including Turbo Yeast 48, red star, baker's yeast, all tech, and strand. Based on the results of research, several types of brands that obtained the highest ethanol yields in a row are the red star, Turbo Yeast 48, strand, baker's yeast, and All tech. In this study Turbo Yeast 48 was used to convert glucose into ethanol. The yeast used to inoculate fermentation meets the following criteria: m~~M~~ust be in a healthy and active state to minimize the length of the lag phase in the next fermentation. It should be available in large volumes ~~large enough~~ to provide an inoculum ~~of~~ in optimal size. It should be in the form of appropriate morphology. It m~~M~~ust be contamination-free. It must maintain the ability to form its products.

The process adopted to produce an inoculum that meets these criteria is called inoculum development or starter inoculation. The formation of products in the starter culture is not a goal in the development of the inoculum so that the starter media can have a different composition of the production media. Fermentation is minimized by growing culture in 'final-type' media. Inoculum development media should be quite similar to production media to minimize the period of adaptation of culture to production media, thereby reducing lag phase and fermentation time. Inoculum which is usually used is between 3 and 10% 3-10% of medium volume. A relatively large volume of inoculum is used to minimize the length of the lag phase and produce maximum biomass in the production fermentation in ~~the~~ a shortest ~~possible~~ time, thereby increasing vessel productivity [47]. Factors that influence the fermentation process are: for *Saccharomyces* c~~C~~*erevisiae* used in wine fermentation, the manufacture of alcohol using low initial sugar content causes a short yeast growth time resulting in a low alcohol content. This is because the number of microbes formed is greater when compared to the amount of food available at the beginning of fermentation [48]. ***Saccharomyces cerevisiae*** can grow well in the pH range 3-6, if the pH is smaller than 3.5 range nya dari 3.5-6.0 then the fermentation process will be reduced in speed. The most optimum pH for the fermentation process is in the range of 4.5-5. At higher pH, yeast adaptation is lower and fermentation activity also increases, but there are also other influences on the formation of by-products, for example, if the pH is high it will increase the concentration of glycerin. In general, a good temperature for the fermentation process is between 20-30 ^{0}C. The

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lower the fermentation temperature, the higher the alcohol content produced. This condition occurs because, at low fermentation temperatures, fermentation will be more perfect and lose alcohol. After all, because it is carried by less carbon dioxide [43].

Fermentation time is generally about 7 days depending on sugar content, temperature, and others. For example, for materials containing 10% glucose ~~10%~~ ,~~of~~ the optimum fermentation time is 4-5 days, while for substances containing 18% glucose ~~18% of~~ , the optimum time of fermentation is 5-6 days. The yeast content used is very influential on the fermentation time as well as the alcohol content produced. High concentrations of the ingredients provide higher inhibitor concentrations and greater concentrations of yeast can make detoxification faster and the resulting alcohol content greater. Within the *Saccharomyces cerevisiae* species, many different strains have very different performance characteristics and produce diverse flavor congeners. For example, yeast strains used in Turbo Yeast 48 products have the ~~can~~ ability to ferment up to a very high percentage of alcohol (20 %) while producing a very low volatile compound. Conversely the usual yeast strain *Saccharomyces cerevisiae* can produce maximum CO_2 ~~to cause~~that cause the dough to expand, but usually dies at a much lower alcohol content when producing high volatile levels.

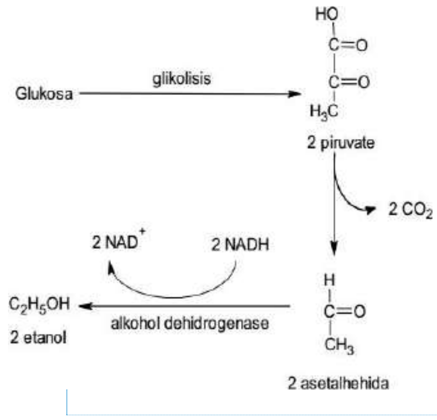

Fig. 1. ~~Reaction to the~~The mechanism of fermentation process. Glukosa mengalami glikolisis membentuk 2 piruvate, bereaksi membentuk 2 asetaldehida dan 2 CO_2, dengan dehidrogenasi 2 asetaldehida menghasilkan 2 etanol dengan enzim 2 NADH atau 2 NAD^+

Turbo Yeast 48 contains Ethanol TT yeast which is very actively mixed with Nutrient Turbo Yeast 48 for high alcohol fermentation. Turbo Yeast is commonly used for the manufacture of alcohol from fruits, sugar cane drops, as well as grains. Turbo yeast is a mixture of dry wine yeast (*Saccharomyces cCerevisiae*) and nutrients. The special strains used (there are many different strains of *Saccharomyces cCerevisiae*) ~~were~~ should be chosen because of their ability to produce alcohol and their nutrients are optimized to provide the right combination of nitrogen, vitamins, and trace minerals that yeast needs in different stages of fermented alcohol. It also contains a pH adjustment because the pH of the sugar/water mixture is far from optimal. The advantage of Turbo Yeast is ~~that~~ it produces a higher alcohol content ~~of~~ up to 20%, works in a

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~~relatively~~ fast time, and withstands in fairly high temperatures [49].

~~Acid hydrolysis uses acid as its catalyst, usually used is a strong acid~~Acid hydrolysis usually uses a strong acid as a catalyst. ~~A commonly used acid in hydrolysis is Hydrochloric acid~~ Commonly, the acid used is HCl or H_2SO_4 in the concentration range of 2-5%. ~~The speed of the hydrolysis reaction is influenced by the presence of H+ ions in the solution, so the greater the number of H+ ions, the higher the reaction speed and the greater the product of hydrolysis results. With the same concentration of different catalysts, both H_2SO_4 and hydrochloric acid have the same amount of water, but H_2SO_4 has more H+ ions than hydrochloric acid resulting in better bond disconnection [43].~~ Starch ~~is~~ has a more complex component than disaccharides. Starch consists of 2 fractions that can be separated by hot water~~,~~. ~~T~~The dissolved fraction is called amylose and the insoluble fraction is called amylopectin. Amylose has a straight structure with a bond of α (1~~,~~-4) D-glucose, while amylopectin has branches with a bond of α (1~~,~~-4) (1.4)D-glucose as much as 4-5% of the total weight. Before undergoing the fermentation process, starch should be broken down with water using the ~~help of~~ amylase enzymes. Amylase is an enzyme that serves to break down starch and can be grouped into three enzyme groups namely α-amylase, β-amylase, glucose amylase. Enzyme α – amylase works by breaking the α-1,4-glycosidic bonds in the straight amylase to produce glucose in alpha, maltose, and dextrin configurations [50]. Maltose is hydrolyzed into glucose using the enzyme maltase.

The formation of bioethanol from glucose through the fermentation process takes place through two stages, namely the glycolysis stage and the alcohol fermentation stage. In the early stages, carbohydrates will be broken down first into simple sugars ~~that are by~~ (hydrolysis of starch into glucose units). In the first stage of glucose, fermentation ~~is~~ always form~~ed~~s pyruvate acid. Glycolysis is a series of chemical reactions decomposition of glucose (which has 6 C atoms) to pyruvate acid (which has 3 atoms C), NADH, and ATP. NADH (Nicotinamide Adenine Dinucleotide Hydrogen) is a coenzyme that binds ~~to~~ electrons (H), so-called high-energy electrons. ATP (Adenosine Triphosphate) is a high-energy compound. Each phosphate released produces energy. In the glycolysis process, each glucose molecule is converted into 2 molecules of pyruvate acid, 2 NADH, and 2 ATP. In the second stage of alcohol fermentation, pyruvate is converted into alcohol through two stages: ~~namely first,~~ first, pyruvate is decomposed into acetaldehyde by pyruvate decarboxylase by involving pyrophosphate thiamine, and the second stage of acetaldehyde by dehydrogenase alcohol is reduced with NADH2 to alcohol [51].

The suitable design of the first-order experiment ~~suitable~~ for the filter stage of the factor is the 2^k factorial design (Two-Level Factorial Design). Equations and optimization results ~~are~~ were obtained by using Minitab software. In this software, the optimal results will be shown by using graphs as well as calculation result numbers.

Methodology

4 L ~~L~~liquid waste of wheat flour that has been cleaned from impurities ~~as much as four liters is~~was put in the hydrolysis tank~~,~~. ~~T~~The hydrolysis process ~~with a solution of~~was conducted by adding 5% (v/v) H_2SO_4 ~~5 (% v/v) while~~ and stirring for 1 hour at ~~a speed of~~ 1500 rpm at a temperature of ± 30 ^{0}C. Hydrolysis results ~~are~~ were filtered ~~to take~~

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~~filtrate to be used~~and the filtrate was processed for the fermentation ~~process and~~ while residue can be used for compost. Glucose obtained ~~is~~ was then analyzed using a glucose refractometer, the maximum level of glucose used in the fermentation process ~~is~~ was less than 16%, if glucose levels ~~are~~ were more than 16%~~ carried out the dilution process,~~ it should be diluted. The pH of ~~F~~filtrate result of impurities-free hydrolysis process ~~is~~was regulated ~~pH solution ±4.5~~ by adding with ~~a solution of~~ H_2SO_4 98% until the pH reached 4.5. This step could be done if the solution obtained was in alkaline condition. In contrast, if ~~if the solution is alkaline, if~~ the solution ~~is~~ was in acidic condition, it should be ~~then~~ added with ~~sodium hydroxide 8N~~8 N NaOH.

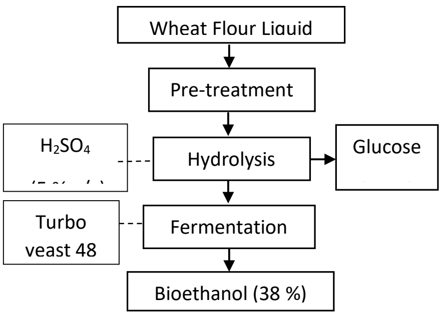

Fig. 2. The flow of ~~G~~glucose and bioethanol production ~~flow~~ used hydrolysis and fermentation process.

500 mL filtrate was taken and put into the bottle. This bottle then sterilized in incubators equipped with UV lamps for 15 minutes~~Filtrate is taken as much as 500 ml, put in each bottle. Bottles containing filtrate are sterilized in incubators equipped with UV lamps for 15 minutes~~. Then the lamp was turned off ~~Turn off the UV lamp,~~ to insert Turbo Yeast 48 ~~according to variables~~in a various concentration of 2, 4, 6, 8, 10 (g/L~~l~~) into each sterile bottle~~,~~. Then it was added with NPK and u~~U~~rea ~~each~~ in each concentration of 0.1 gram/100 m~~l~~L. Prepare the H_2O in a separate bottle to taste, close tightly both bottles, connect the connecting hose to bottle 1 and the H_2O bottle for the indicator of the presence of CO_2 and connect the sample hose to bottle 1 for sampling and addition of nutrients. After the bottle is tightly closed and all hoses have been arranged, the fermentation process ~~is~~ was carried out ~~with time according to variables~~in various time of 3, 4, 5, 6, 7 (days).

Results

The raw material used in the manufacture of bioethanol ~~is~~ was the liquid waste of wheat flour obtained from PT. Boga Sari Flour Mill. Wheat flour has a high starch content of around 70%, so it is expected that in the liquid waste there is still a high content of starch as well. According to laboratory analysis results, starch content in wheat flour liquid waste is on average 9.282 % v/v, and glucose content of 3.786 % v/v, so glucose levels can be obtained about 12 % v/v after the hydrolysis process. In the fermentation process, starch must be broken down first into glucose so that hydrolysis is necessary. In the process of making bioethanol based on wheat flour liquid waste, the material is hydrolyzed first using a solution of H2SO4 5% (v/ v) while stirring for 1 hour at a speed of 1500 rpm, then followed by a fermentation process using turbo yeast as a source of Saccharomyces Cerevisiae, urea and NPK 0.1g/100 ml as microorganism nutrition. The fermentation process lasts for 7 days with variable turbo yeast 2,

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4, 6, 8, 10 (g/l),(ya Bu Dian setuju) obtained the following data:

Table 1

Bioethanol Content Analysis in Fermentation Process

Content Bioethanol (% v/v)					
Content	Fermentation time (days)				
Turbo yeast (g/L~~l~~)	3	4	5	6	7
2	28	31	32	33	33
4	32	33	34	34	34
6	33	34	35	35	35
8	34	35	36	36	36
10	35	36	37	37	37

Based on previous research for fermentation of straw 80 g/L~~l~~ using Turbo Yeast obtained results that is optimum Turbo Yeast content of 6 g/l, and ethanol content of 18.6 % v/v [52]. Sebagai dasar penelitian menggunakan turbo yeast 6 g/l menghasilkan kadar etnanol 18,6%. The manufacture of bioethanol from jackfruit seeds with the process of liquidation and fermentation using *S~~s~~accharomyces cerevisiae* ~~obtained the results~~showed that the highest alcohol ~~content of~~was 40% with a fermentation time of 60 hours and T~~t~~urbo Y~~y~~east ~~content~~ used was 0.01 % v/v [49].

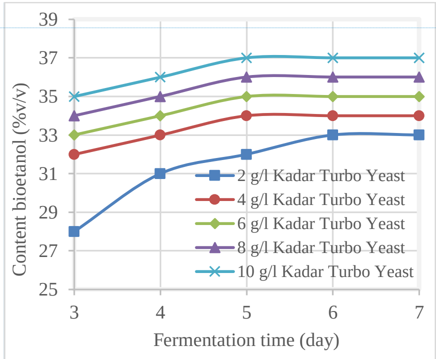

Fig. 3. ~~Fermentation time (days) to two-dimensional bioethanol content (% v/v).~~The dependence of the bioethanol (% v/v) on the fermentation time (days) and the concentration of Turbo Yeast

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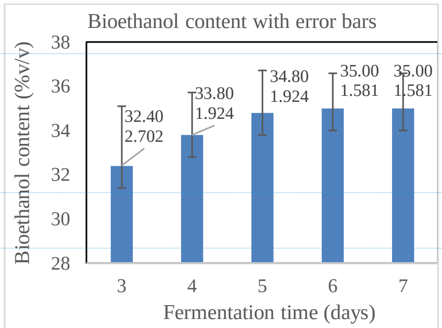

In previous study, a ~~B~~bioethanol production from liquid waste flour ~~with hydrolysis process using~~by using B~~b~~acillus in its hydrolysis process

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produced glucose levels of 5-10 (% v/v) and ~~with~~ when the fermentation process ~~using~~ used ~~s~~*Saccharomyces cerevisiae* for 6 days produced ethanol levels of 11-16 (% v/v). The ethanol leveled up to 95-96% when the process continued by ~~and continued with the~~

Based on Figs. 3 and 6 it can be ~~known~~ seen that this treatment ~~does~~ ~~not~~did not undergo an adaptation phase. ~~, this is~~ It is because on the 3rd-day microorganisms have reached the logarithmic phase so that bioethanol levels will increase over time and at some point, bioethanol levels will be constant~~,~~ ~~this is because~~due to microorganisms have undergone a stationary phase. However, the time to reach this stationary phase was varied~~ies~~ ~~with each treatment~~regarding the treatment. In a treatment with 2% T~~t~~urbo Y~~y~~east ~~treatment 2%~~ the stationary phase occured~~s~~ on day 6 to 7 ~~(days)~~. Meanwhile, ~~In~~ in a treatment by using 10% T~~t~~urbo Y~~y~~east, the ~~treatment 10%~~ stationary phase occured~~s~~ from day 5 to 7 ~~(days)~~. This suggests that ~~using too~~ high levels~~s~~ of turbo yeast can accelerate the stationary phase and death phase~~,~~ due to the ~~greater~~ increased number of microorganisms formed so it cannot be comparable with ~~compared to~~ the amount of food available~~, at the beginning of fermentation~~.

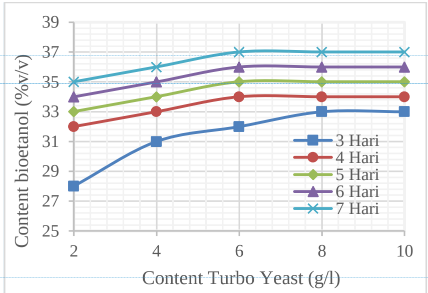

Fig. 6. Turbo yeast content (g/L~~l~~) to two-dimensional bioethanol content (% v/v).

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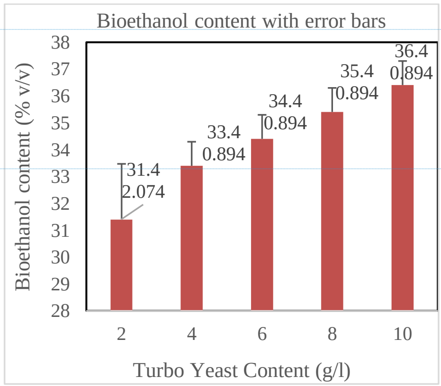

~~Based on~~ Figs. 4 and 7 show~~s~~ the ~~figure~~ model of optimization results. ~~The figure above shows the maximum optimum result.~~ The highest point in the picture above shows the highest point of result. Where if the red line is shifted following the shape

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Based on Figs. 5 and 8 after treatment with contour plot depiction and surface plot optimization using Surface Response Method in Minitab 17 software application can be done an optimization of desired results with certain parameters. The parameters of the optimization result are were set to produce the most optimum alcohol contentgain the optimum condition to produce alcohol with a high concentration. The optimization by using surface response method obtained Optimization results using surface response method has an optimum result at of turbo yeast content of 11.6569% and fermentation time of 5 days that resulted in alcohol content of 37.2073%.

Fig. 8. Surface characteristics of contour plot response between turbo yeast content and fermentation time to alcohol content

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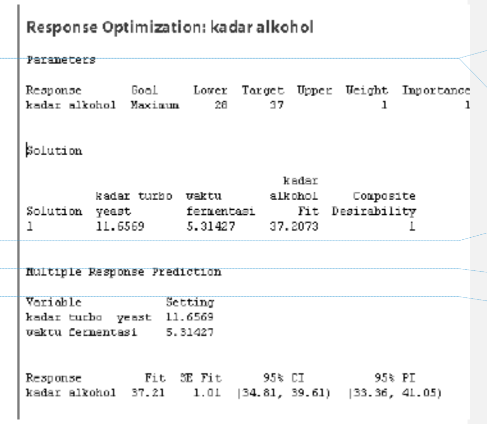

```
Response Optimization: kadar alkohol

Parameters

Response        Goal      Lower  Target  Upper  Weight  Importance
kadar alkohol   Maximum      28      37              1           1


Solution

                                        kadar
           kadar turbo  waktu           alkohol    Composite
Solution   yeast        fermentasi         Fit  Desirability
1          11.6569      5.31427        37.2073             1


Multiple Response Prediction

Variable              Setting
kadar turbo   yeast   11.6569
waktu fermentasi      5.31427


Response          Fit  SE Fit       95% CI          95% PI
kadar alkohol   37.21    1.01  (34.81, 39.61)  (33.36, 41.05)
```

Fig. 9. The output of Othe optimization rResult Output Graph withby using Minitab Software 17

As a result of our research, it hasThe results of this work provided the best bioethanol content ofhighest bioethanol obtained was 37% (v/v) in thewith a treatment using 11% (v/v) glucose levels of 11 % v/v, the addition of 10 g/L Tturbo Yyeast 10 g/l, 0.1 g/100 mL for each urea and NPK 0.1 g/100 ml, stirring 1500 rpm for 1 hour, and fermentation time of 5 days. This is due to the use of Turbo Yeast 48 is very active mixed with Nutrient Turbo Yeast 48 for high alcohol fermentation so that it can produce higher bioethanol levels than the use of ordinary Saccharomyces cerevisiae or the use of other microorganisms.

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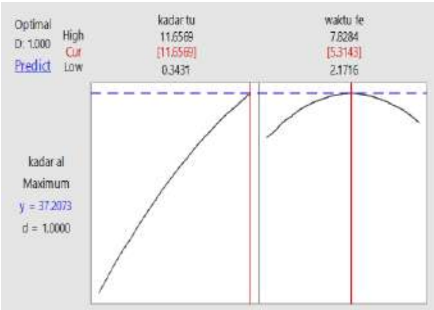

Fig. 10. Graph of Alcohol Content Optimization Results

5. Conclusion

The hydrolysis process is known that the glucose content in the raw material of wheat flour liquid waste by 11 (% v/v). Limbah cair tepung terigu diproses hidrolisis menggunakan H2SO4 menghasilkan glukosa 11% dalam bentuk filtrat, Dari filtrat tersebut dilakukan proses fermentasi menggunakan variable turbo yeast 48 dan waktu fermentasi menghasilkan kadar bioethanol 37%. This work presents the optimum condition to obtain a high ethanol concentration (37% (v/v)) was 10 % turbo yeast with fermentation time until it reached a constant state was 5 days. ~~The best result in the fermentation process is at 10 % turbo yeast content and lasts on day 5 which produces bioethanol content of 37 (% v/v). Optimization results with~~ By using Response Surface Methodology (RSM) using Minitab 17 application, the optimum results acquired were 11.6569 (% v/v) turbo yeast and fermentation time of 5 days resulted in a bioethanol content of 37.2073 (% v/v).~~Response Surface Methodology (RSM) using Minitab 17 application obtained optimum results at turbo yeast content of 11.6569 (% v/v) and fermentation time of 5 days resulted in a bioethanol content of 37.2073 (% v/v).~~

Acknowledgment

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	Analisis statistika	-	-		
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Result and Discussion	Penyajian data	√			
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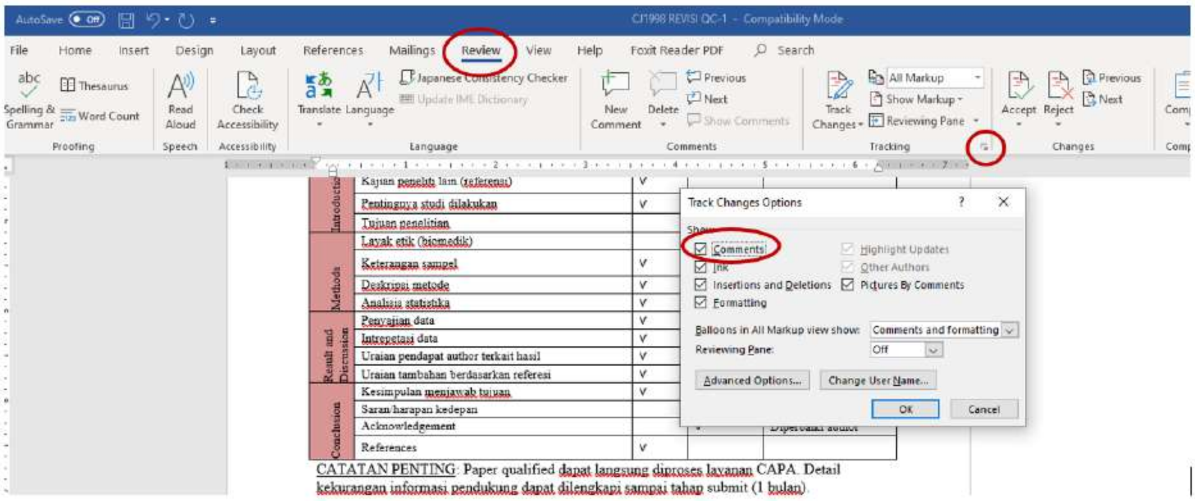
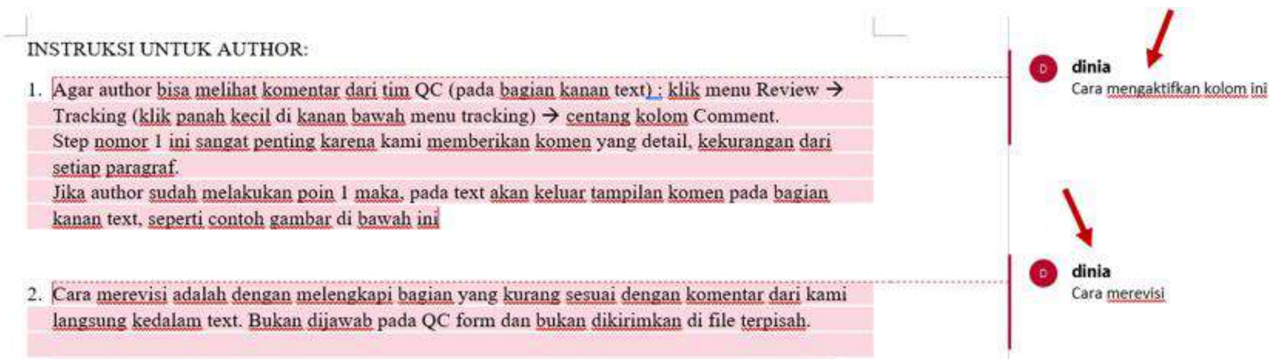
   

   Step nomor 1 ini sangat penting karena kami memberikan komen yang detail, kekurangan dari setiap paragraf.
   Jika author sudah melakukan poin 1 maka, pada text akan keluar tampilan komen pada bagian kanan text, seperti contoh gambar di bawah ini:

2. Cara merevisi adalah dengan melengkapi bagian yang kurang sesuai dengan komentar dari kami langsung kedalam text. Bukan dijawab pada QC form dan bukan dikirimkan di file terpisah.

3. Tandai bagian yang telah direvisi dengan warna merah seperti yang kami contohkan.

4. Jika ada bagian yang kurang jelas mohon segera diskusikan dengan tim kami.

Proces Fermentation of Filtrate Bamboo with Saccharomyces Cerevisiae and Zymomonas Mobilis

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Abstract. Fermentation is the process of the formation of ethanol from glucose by using enzymes. Bamboo is one of the materials containing glucose is high enough, that is previously done hydrolysis in advance. Bamboo used when the hydrolysis process of bamboo that does not include lignin and the pentose done process of pretreatment and not lignification. The purpose of this research is to produce ethanol as a raw material substitution of bioethanol, knowing pentose and dirt left in the bamboo. Therefore, need to be studied in the future, with the best process, that we used biological processes that can optimize the production of ethanol. The use of the enzyme (Saccharomyces Cerevisiae and Zymomonas Mobilis) is also significant because of the optimum enzyme conditions. Temperature, pH, and the yeast with optimal conditions when it can raise the level of his work. The fermentation process at temperature 25 C and 45 C, the filtrate is 500 ml solution of bamboo and the stirring speed of 200 rpm. The variable composed enzyme with a ratio (v/v) of 0.25 to 0.75. Resulting from the fermentation processed can produce ethanol with a yield 30.5% and 36% of the weight of the bamboo. The result of the process of fermentation obtained bioethanol with low ethanol yield of 10-15%, which requires the flash distillation process to obtain yield bioethanol technical 90-95%.

1. Introduction

Biomass from plants has declared as an alternative raw material for gasoline fuel substitution in the form of bioethanol, bioethanol obtained from biomass and bioenergy crops has proclaimed as one of the feasible alternatives as gasoline fuel [1]. Sustainable bioethanol from rice straw [2]. Ethanol production from lignocellulose by hydrolysis process chemically or enzymatically, first conducted the process of pretreatment, the next process is the process of fermentation and distillation process. Pretreatment processes, the most important is to remove the lignin and pentosan, which can dissuade lignin and pentosan skarifikasi process. Various approaches have been performed earlier as pretreatment pretreatment in acids, bases, ammonia, sodium chlorite, and biological [3].

The research conducted to evaluate acid pretreatment from hydroxide paper waste as material for bioethanol production, optimized sulfuric acid hydrolysis, fermentation process of hydroxide acid of paper waste by using Pichia Stipites. The ethanol content obtained at 77.54%. By one more distillation process, the ethanol content received at the level of 95-96% [4]. Chemical pretreatment of lignocellulose biomass with Sulphur (H_2SO_4) and phosphorus (H_3PO_4) acids used since they are relatively cheap and efficient in hydrolyzing lignocellulose, though the letter gives a milder effect and is more benign to the environment. Hydrochloric (HCl) acid is more volatile and more natural to recover and attacks biomass better than H_2SO_4 [5]. Similarly, nitric acid (HNO_3) possesses good cellulose to sugar conversion rates [6]. However, both acids are expensive compared to Sulphur acid. Pretreatment of lignocellulose has received considerable research globally due to its affluence on the technical, economic and environmental sustainability of cellulose ethanol production. These paper reviews know, and emerging chemical pretreatment methods, the combination of chemical pretreatment with other ways to improve carbohydrate preservation reduce formation to degradation product, achieve high sugar yield at mild reaction conditions, reduce solvent loads and enzyme dose, reduce waste generation [7]. Technical and economic evaluation of bioethanol production from lignocellulose residues, a case of sugarcane and blue agave bagasse [8].

Initiatives of the future for lignin in biomass to bioethanol, pretreatment technologies to separate the main tree biopolymers (cellulose, hemicellulose, and lignin) [9]. Pretreatment for hydrogen and bioethanol production from olive oil waste products was ethanol yield 5.4 % treatment with 1.75 w/v Sulphur acid and heated it at 140 0C for 10 min, and was ethanol yield 5.0 % no pretreatment [10]. Pretreatment followed with simultaneous scarification and fermentation on bioconversion of microcrystalline cellulose for bioethanol production, the yield value of 67 % bioethanol bioconversion [11]. A sustainable feedstock bioethanol production, cellulose hydrolysis was microwave irradiation using hydrochloric acid as catalyst, fermentation with yeast (Saccharomyces cerevisiae), modest reaction conditions (2.38 M acid concentration), irradiation time 7 min, and yield of 0,67 g glucose / g cellulose [12]. Elements contained in the lignocellulose biomass of the plants are usually used lignocellulose biomass, a potential for bioethanol production globally. Agriculture (softwood), forestry (pretreatment method obtained ethanol content below 16%.

Pretreatment with dilute acid (sulfuric acid) eliminating the hemicellulose components and increase the sugar. Pretreatment method of sulfuric acid has long been recognized as an important step towards eliminating the hemicellulose fraction of lignocellulose substrates, and save the conversion of cellulosic biomass [20]. The research conducted by [21] about ethanol production from sago pith waste (SPW) using microwave hydrothermal hydrolysis catalyzed by carbon dioxide, resulted in higher energy saving compared to previous techniques in the absence of enzymes, acid or base catalyst. They obtained ethanol content below 15.6%. The production of bioethanol from lignocellulosic biomass through the different process steps, such as pretreatment, hydrolysis, fermentation, distillation and [26]. Ethanol production from lignocellulosic technologies largely determined by hydrolysis and pretreatment, whether chemical or biological [27].

Ethanol from the liquid waste of rise flour using fermentation by Saccharomyces, a maximum of 23.8% glucose and 40.5% ethanol yield, the developed technique for liquid waste of rise flour resulted in higher energy saving compared to the previous method in the absence of enzymes, acid or base catalyst [28]. hardwood), and industrial waste are a significant lignocellulose biomass for bioethanol production. The lignocellulose biomass is one of the potential main sources for economic bioethanol production globally. Agricultural, forestry (soft and hardwoods) and industrial wastes are the major lignocellulose biomasses [13]. The bioethanol production from lignocellulose biomass using process pretreatment, hydrolysis, fermentation, and recovery of ethanol, was obtained by ethanol under 16% v/v, with the distillation process will again be derived ethanol 95-96% v/v. The research conducted bioethanol production from lignocellulose biomass by using the pretreatment process, hydrolysis, fermentation, and ethanol recovery. Therefore, ethanol content obtained in the level below 16%, and by one more distillation process the ethanol content would receive at the level of 95-96% v/v [18].

The research conducted by [19] about bioethanol production from agricultural waste using PROFER Cellulosic or second generation (SG) bioethanol produced from lignocellulosic biomass (LB) in three main steps: pretreatment, hydrolysis, and fermentation. Pretreatment involves the use of physical processes, chemical methods, physic-chemical processes, biological methods, and several combinations thereof to fractionate the lignocellulose into its components. It results in the disruption of lignin seal to increase enzyme access to holo-cellulose [29, 30], reduction of cellulose crystallinity [31, 32], an increase in the surface area [33, 34] and porosity [35, 36] of pretreated substrates, resulting in increased hydrolysis rate. In hydrolysis, cellulose and hemicelluloses are broken down into monomeric sugars via the addition of acids or enzymes such as cellulose. Enzymatic hydrolysis offers advantages over acids such as low energy consumption due to the mild process requirement, high sugar yield, and no unwanted wastes. Enzymatic hydrolysis of cellulose affected by properties of the substrate such as porosity, cellulose fibre crystallinity, and degree of polymerization, as well as lignin and hemicellulose content [37, 38], optimum mixing [39], substrate and end-product concentration, enzyme activity, reaction conditions such as pH and temperature [40, 41].

From the previous research, it knows bioethanol from cellulose resulted in good bioethanol. The study was to search alternative material, review hydrolysis process, fermentation process to gain bioethanol product with a high level of ethanol. The originality of this research was the second generation that was bulrush, by using two methods (hydrolysis and fermentation) simultaneously, used two enzymes [Saccharomyces Cerevisiae (SC) and Zymomonas Mobilis (ZM)], and technical ethanol production with the level of 10-15% as the technical ethanol.

2. Experimental

The average concentration of cellulose was 48% in bamboo, and glucose was 5 % and impurities.

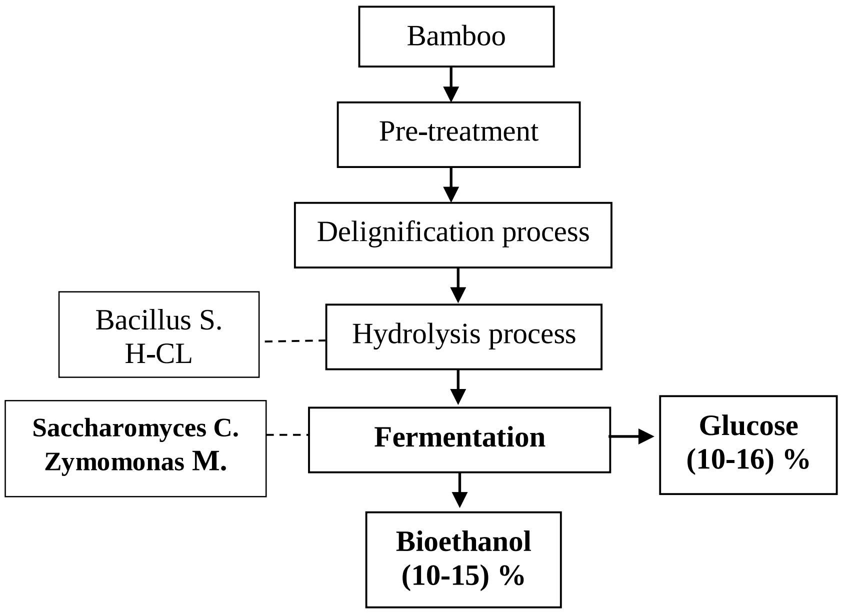

Figure 1. Glucose and bioethanol production flow used fermentation process

The pieces and refined fiber of bamboo with the approximate length of 5 cm and polished thread 200 mesh done to obtain the high level of glucose and cellulose during the hydrolyzed process by Bacillus and H-Cl. The quality bioethanol product determined by various influencing parameters such as the acidity (pH), the volume ratio of H-Cl to bamboo, the volume ratio of Bacillus Subtitles to the filtrate, the volume ratio of the enzyme (Saccharomyces C. and Zymomonas M.) to the filtrate, and fermentation time. Laboratory analysis did the quality analysis of raw materials and bioethanol product. The study conducted on the instrumentation and gravimetric analysis. Hydrolysis process in Figure 1 done in stable condition: temperature of 30 ^{0}C, water volume in 7 liters, and hydrolysis time in 1 hour with 200 rotations per minute (RPM). For the changing condition: bamboo weight of 50, 100, 150, 200, 250 (grams), the ratio of bacillus to filtrate volume 1:2; 5:4; 10:7 and H-Cl solution volume 10, 20, 30, 40, 50 (ml). The level of glucose in hydrolysis filtrate yield was analyzed before the fermentation process done previous research [42]. Fermentation process in Figure 1 done in stable condition: filtrate bamboo ratio of the varies Saccharomyces C. and Zymomonas M.: 5, 9, 13 (% v/v), fermentation time 4, 6, 8, 10, 12 days. Filtrate rate influences the residual glucose levels, obtained maximum residual glucose levels (1,3 - 3) %, and this is because in the tank hydrolysis reactor and the amount of filtrate starter Saccharomyces C. and Zymomonas M. in still little so that the fermentation process is not optimal. With the increasing amount of filtrate hydrolysis and starter Saccharomyces Cerevisiae then the smaller the residual glucose, because it fermented into ethanol.

2. Results and Discussion

Bamboo using as a study material derived from bamboo crops in the surrounding area. Assessment method is done, by doing a survey and laboratory analysis to obtain some data about the quality and quantity of the available bamboo. The expected result was data about the quality and bamboo quantity before processing to be ethanol. Ethanol forming elements were cellulose and glucose, the concentration of cellulose was 48.1 %, glucose was 4.8%, and impurities. If the entire cellulose hydrolyzed completely, it will be obtaining the glucose levels of 53%.

The results of fermentation process with ratio enzyme again filtrate cellulose as:

Table 1. Ethanol level and yield on fermentation process

Ratio Filtrate Sellulose (%v/v)	Fermentation time (day)	Glucose level Fermentation (%)		Ethanol Level Fermentation (%)	
		SC	ZM	SC	ZM
5	4	4.82	5.76	10.5	10.0
	6	5.30	5.23	11.0	12.3
	8	5.70	5.27	9.0	13.4
	10	5.78	5.54	12.5	10.3
	12	5.81	5.77	11.0	10.3
9	4	5.08	5.90	12.0	9.1
	6	5.68	5.76	14.5	13.2
	8	7.63	6.03	14.0	14.9
	10	7.78	6.78	15.0	13.5
	12	7.98	7.91	14.0	12.6
13	4	7.41	6.88	14.0	13.0
	6	8.35	7.95	14.5	13.7
	8	9.56	8.77	15.0	14.5
	10	9.87	9.05	14.5	12.3
	12	9.88	9.35	13.0	12.8

The pieces and refined fiber of bamboo with an approximate length of 5 cm and polished thread 200 mesh were done to obtain the high levels of glucose and cellulose before it hydrolyzed by Bacillus and H-Cl solution. Bamboo should be made in powder form so that cellulose hydrolyzed perfectly. However, that process took a higher cost. Besides, bulrush in the powder form could suffer the physical destruction, thus causing the damage of the glucose group. The drying process of bulrush was naturally done first in the room temperature. The drying process was done in an oven at 1000C for 3 hours. These done for cost savings. The drying process aimed to reduce the water content in ethanol. The water level that was permitted by Standart National Indonesia (SNI) was 1%. The decreasing of pH from pretreatment material was affected by the addition of H-Cl volume 7%v/v because the requiring pH for fermentation process was 4,5. Before doing the hydrolysis process, the pH of filtrate measured according to the terms of the fermentation process that is approximately 4.5. To obtain pH 4.5, the addition of Na-OH done if the pH of the filtrate was under 4.5 and the addition of citric acid if the filtrate pH was above 4.5. Filtrate rate influences the residual glucose levels, for a number of starter Saccharomyces C. 5, 9, and 13% v/v, obtained maximum residual glucose levels (1,3 - 3) %, this is because in the tank hydrolysis reactor and the amount of filtrate starter Saccharomyces C. still little, so that the fermentation process is not optimal. With the increasing amount of filtrate hydrolysis and starter Saccharomyces C. then the smaller the residual glucose, because it fermented into ethanol.

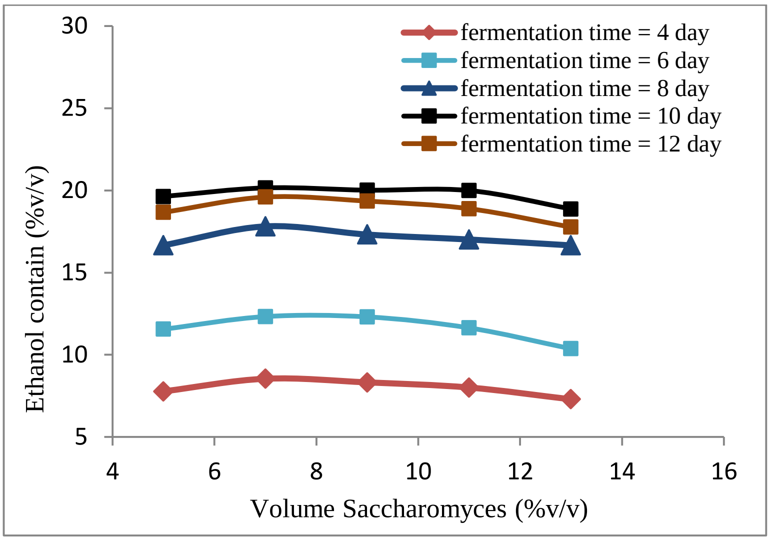

Figure 2. Effect Saccharomyces volume on the remaining ethanol contain

After analysis glucose levels in the rest of the fermentation process, with the addition of Saccharomyces C. 7 % of the volume of fluid (filtrate) showed small residual glucose levels compared to the addition of starter 5, 11 and 13 %. These are because the preliminary research has been conducted by following the Journal and the acquisition of 7% of the fluid volume. Filtrate rate influences the residual glucose levels, for a number of starter Saccharomyces C. 5, 9, and 13%, obtained maximum residual glucose levels (1.5-10) %, this was due to the amount in the tank reactor filtrate hydrolysis and starter Saccharomyces C. still little, so that the fermentation process is not optimal. With the increasing amount of filtrate hydrolysis and starter Saccharomyces C. then the smaller the residual glucose, because it fermented into ethanol.

5. Conclusion

Fermentation process from bamboo to bioethanol, glucose levels obtained in the fermentation process as (5-10) % for filtrate cellulose, levels of ethanol in the fermentation process equal 10 until 15 %. The Saccharomyces C. had higher glucose and bioethanol levels results of Zymomonas M., but durability Zymomonas M. stronger in a fermentation process.

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0,98 % :

Sulfuric acids (H_2SO_4) and phosphorus (H_3PO_4) acids are commonly used in the chemical pretreatment of lignocellulose biomass since they are relatively efficient in hydrolyzing lignocellulose. Meanwhile, hydrochloric acid (HCl) is more volatile and more natural to recover and attacks biomass better than H_2SO_4 [5]. Similarly, nitric acid (HNO_3) possesses good cellulose to sugar conversion rates [6].

Simple:

Sulfuric acids (H_2SO_4) and phosphorus (H_3PO_4) acids are **typically** used **withinside the** chemical pretreatment of lignocellulose biomass **given that they're** relatively **green** in hydrolyzing lignocellulose. Meanwhile, hydrochloric acid (HCl) is **extra unstable** and **extra herbal** to **get better** and **assaults** biomass **higher** than H_2SO_4 [5]. Similarly, nitric acid (HNO_3) possesses **right** cellulose to sugar conversion rates [6].

Advance:

Sulfuric acids (H_2SO_4) and phosphorus (H_3PO_4) acids are **normally** **utilized in** the chemical pretreatment of lignocellulose biomass since **they're** **comparatively** efficient in hydrolyzing lignocellulose. Meanwhile, **acid** (HCl) is more volatile and **additional** natural to recover and attacks biomass **higher** than H_2SO_4 [5]. Similarly, **aqua fortis** (HNO_3) possesses **smart** **polysaccharide** to sugar conversion rates [6].

Fluency:

Sulfuric acids (H2SO4) and phosphorus (H3PO4) are commonly used in the chemical pretreatment of **lignocellulosic** biomass **because they** are relatively efficient in hydrolyzing lignocellulose. **In the meantime,** hydrochloric **acid** (HCl) is more volatile and natural to recover and attack biomass **better** than H2SO4 [5]. **Similarly,** nitric acid (HNO3) **has** good **conversion rates from cellulose** to sugar [6].

Creative:

Sulfuric acid (H2SO4) and phosphorus (H3PO4) are commonly used in the chemical pretreatment of **lignocellulosic** biomass **because they** are relatively **effective** in hydrolyzing lignocellulose. **At the same time,** hydrochloric **acid** (HCl) is more volatile than H2SO4**, easier to recycle, and better to decompose biomass** [5]. **The conversion rate of nitric** acid (HNO3) **from** cellulose to sugar **is also very high** [6].

1,03% :

A sustainable feedstock bioethanol production, cellulose hydrolysis was microwave irradiation using hydrochloric acid as catalyst, fermentation with yeast (*Saccharomyces cerevisiae*), modest reaction conditions (2.38 M acid concentration), irradiation time 7 min, and yield of 0.67 g glucose/g cellulose [12]. Every plant containing biomass of lignocellulose has a potential for bioethanol production. Raw materials from agriculture (softwood) and forestry have the biomass potential to produce ethanol with a concentration of less than 16%.

Simple:

A sustainable feedstock bioethanol production, cellulose hydrolysis was microwave irradiation **the use of** hydrochloric acid as catalyst, fermentation with yeast (*Saccharomyces cerevisiae*), modest **response** conditions (2.38 M acid **awareness**), irradiation time 7 min, and yield of 0.**sixty seven** g glucose/g cellulose [12]. Every plant containing biomass of lignocellulose has a **capacity** for bioethanol production. Raw **substances** from agriculture (softwood) and forestry have the biomass **capacity** to produce ethanol with a **awareness** of **much less** than 16%.

Advance:
A **property** feedstock bioethanol production, **polyose** **reaction** was microwave irradiation **mistreatment** **acid** as catalyst, fermentation with yeast (*Saccharomyces cerevisiae*), modest reaction conditions (2.38 M acid concentration), irradiation time **seven** min, and yield of 0.67 g glucose/g cellulose [12]. Every plant containing biomass of lignocellulose has a potential for bioethanol production. Raw materials from agriculture (softwood) and **biology** have the biomass potential **to provide** ethanol with **a amount** of **below** 16%.

Fluency:
Sustainable production of bioethanol **as a raw material,** cellulose hydrolysis **consisted of microwave** irradiation **with** hydrochloric acid as **a catalyst,** fermentation with **yeast** (Saccharomyces cerevisiae), **moderate** reaction conditions (**concentration of 2.38** Macid), irradiation time **of 7** min and **a yield** of 0.67 g glucose **/ g Cellulose** [12]. **Plants** containing **lignocellulosic biomass have** potential for bioethanol production.**Agriculture** and forestry have the biomass potential to produce **ethanol** with a concentration of less than 16%.

Creative:
Sustainable production of bioethanol **as a raw material,** cellulose hydrolysis **includes the following steps: microwave** irradiation **with** hydrochloric acid as **a catalyst,** fermentation with **yeast** (Saccharomyces cerevisiae), **moderate** reaction conditions (**concentration of 2.38** Macid), irradiation time **of 7 minutes** and **production** of 0.67 g glucose/g **cellulose** [12]. **Plants** containing **lignocellulosic biomass can produce** bioethanol.**Agriculture** and forestry have biomass potential **for ethanol production with** a concentration of less than 16%.

0,72% :

The research conducted by [21] regarding ethanol production from sago pith waste using microwave hydrothermal hydrolysis catalyzed by carbon dioxide resulted in a higher energy saving compared to previous techniques in the absence of enzymes, acid, and base catalyst. They obtained ethanol content below 15.6%.

Simple:
The **studies performed via way of means of** [21] **concerning** ethanol **manufacturing** from sago pith waste **the usage of** microwave hydrothermal hydrolysis catalyzed **via way of means of** carbon dioxide resulted in a **better power** saving **in comparison** to **preceding strategies withinside the** absence of enzymes, acid, and base catalyst. They **acquired** ethanol **content material underneath** 15.6%.

Advance:
The research conducted by [21] **concerning** **plant product** production from **starch** pith waste using microwave hydrothermal **chemical reaction** catalyzed by **greenhouse emission** resulted in a higher energy saving compared to previous techniques **within the** absence of enzymes, acid, and base catalyst. They obtained **plant product** content below 15.6%.

Fluency:
Investigations by [21] **on the** production **of ethanol from** sago **marrow** waste **using carbon dioxide-catalyzed** hydrothermal **microwave hydrolysis** resulted in **greater** energy **savings** compared to **earlier** techniques **without enzymes,** acid and **basic** catalyst. **They received an ethanol** content below 15.6%.

Creative:
Compared with previous methods without enzymes, acid and base catalysts, the use of carbon dioxide-catalyzed hydrothermal microwave hydrolysis to **produce ethanol from sago core waste [21] saves more energy. Their** ethanol content **is less than 15.6%.**

0,69 % :
Waste from hardwood industries, agricultural, forestry (soft and hardwoods), and industrial wastes contain the major lignocellulose biomasses which have the potential to be main sources for economic bioethanol production [13]. The bioethanol production from lignocellulose biomass by using pretreatment, hydrolysis, fermentation, and recovery of ethanol resulted in ethanol less than 16% v/v. Meanwhile, by the distillation process, the obtained ethanol would be 95%–96% v/v. [18].

Simple:
Waste from hardwood industries, agricultural, forestry (**smooth** and hardwoods), and **commercial** wastes contain the **important** lignocellulose biomasses **that have** the **capability** to be **most important** sources for **monetary** bioethanol production [13]. The bioethanol production from lignocellulose biomass **with the aid of using the usage of** pretreatment, hydrolysis, fermentation, and **healing** of ethanol **ended in** ethanol **much less** than 16% v/v. Meanwhile, **with the aid of using** the distillation process, the **acquired** ethanol **could** be 95%–96% v/v. [18].

Advance:
Waste from hardwood industries, agricultural, **biology** (soft and hardwoods), and industrial wastes contain the major lignocellulose biomasses **that** have the potential to be main sources for economic bioethanol production [13]. The bioethanol production from lignocellulose biomass by **victimization** pretreatment, hydrolysis, fermentation, and recovery of **fermentation alcohol** resulted in ethanol **not up to** 16% v/v. Meanwhile, by the distillation process, the obtained **fermentation alcohol** would be 95%–96% v/v. [18].

Fluency:
Wastes from **the hardwood,** agricultural, forestry (soft and hardwood) and industrial **industries** contain **the main lignocellulosic** biomasses **that** have the potential to be **major** sources **for the economical production of bioethanol. [13] The production of bioethanol** from **lignocellulosic** biomass by pretreatment **and** hydrolysis **The** fermentation **and** recovery of ethanol resulted in ethanol **of less** than 16% v **/ v. Meanwhile, through** the **distillation** process, the **ethanol obtained** would be 95% **to 96%** v **/ v. [18].**

Creative:
Hardwood, agricultur**e and** forestry **(softwood** and hardwood), and industrial waste contain **important** lignocellulosic biomass **and may be the main source of economically produced bioethanol. [13] Bioethanol is produced from lignocellulosic biomass through** pretreatment, hydrolysis, fermentation **and separation. Ethanol results in an ethanol content of less** than 16% **(volume). In** the **distillation** process, the **ethanol obtained is** 95 **to 96% by volume. / Over. [18].**

3,4 %:

The research conducted by [19] regarding bioethanol production from agricultural waste using PROFER Cellulosic or second-generation bioethanol produced from LB involved three main steps, namely, pretreatment, hydrolysis, and fermentation. Pretreatment involves the use of physical processes, chemical process, physicochemical processes, biological process, and several combinations thereof to fractionate the lignocellulose into its components. It will lead to the disruption of lignin seal to increase enzyme access to cellulose [29, 30], reduction of cellulose crystallinity [31, 32], an increase in the surface area [33, 34] and porosity [35, 36] of pretreated substrates, resulting in increased hydrolysis rate. In hydrolysis, cellulose and hemicelluloses are broken down into monomeric sugars via the addition of acids or enzymes such as cellulose. Enzymatic hydrolysis offers advantages over acids, such as low energy consumption due to the mild process requirement, high sugar yield, and no unwanted wastes. Enzymatic hydrolysis of cellulose is affected by properties of the substrate, such as porosity, cellulose fiber crystallinity, and degree of polymerization, as well as lignin and hemicellulose content [37, 38], optimum mixing [39], substrate and end product concentration, enzyme activity, and reaction conditions such as pH and temperature [40, 41].

Simple:

The research **performed** by [19] **concerning** bioethanol **manufacturing** from agricultural waste using PROFER Cellulosic or second-**era** bioethanol **created from** LB **concerned** three **important** steps, namely, pretreatment, hydrolysis, and fermentation. Pretreatment **entails using bodily** processes, chemical **method**, physicochemical processes, **organic method**, and **numerous mixtures** thereof to fractionate the lignocellulose into its components. It will **cause** the disruption of lignin seal to **boom** enzyme **get entry to** to cellulose [29, 30], **discount** of cellulose crystallinity [31, 32], an **boom withinside the floor** area [33, 34] and porosity [35, 36] of pretreated substrates, **ensuing** in **extended** hydrolysis rate. In hydrolysis, cellulose and hemicelluloses are **damaged** down into monomeric sugars **thru** the addition of acids or enzymes **including** cellulose. Enzymatic hydrolysis **gives blessings** over acids, **including** low energy **intake because of** the **slight method** requirement, **excessive** sugar yield, and no **undesirable** wastes. Enzymatic hydrolysis of cellulose is **suffering from houses** of the substrate, **including** porosity, cellulose fiber crystallinity, and **diploma** of polymerization, **in addition to** lignin and hemicellulose content [37, 38], optimum mixing [39], substrate and **stop** product concentration, enzyme activity, and reaction **situations including** pH and temperature [40, 41].

Advance:

The **analysis** conducted by [19] **concerning** bioethanol production from agricultural waste **victimisation** PROFER **plastic** or second-generation bioethanol **made** from **avoirdupois unit concerned 3** main steps, namely, pretreatment, hydrolysis, and fermentation. Pretreatment involves **the utilization** of physical processes, chemical process, **chemical science** processes, biological process, **and several other mixtures thence** to fractionate the lignocellulose into its components. **it'll result in** the disruption of lignin seal **to extend protein** access to **polysaccharide** [29, 30], reduction of cellulose crystallinity [31, 32], **a rise within the expanse** [33, 34] and porosity [35, 36] of pretreated substrates, **leading to enlarged reaction** rate. In hydrolysis, **polysaccharide** and hemicelluloses are **softened** into monomeric sugars via the addition of acids or enzymes **corresponding to** cellulose. Enzymatic **reaction** offers **blessings** over acids, **corresponding to** low energy consumption **because of** the **gentle method** requirement, high sugar yield, and no unwanted wastes. **catalyst reaction** of **polysaccharide** is **littered with** properties of the substrate, **corresponding to** porosity, **polysaccharide** fiber crystallinity, and degree of polymerization, **further** as **polymer** and hemicellulose content [37, 38], optimum mixing [39], substrate and **ending** concentration, **protein** activity, and reaction conditions **corresponding to pH** and temperature [40, 41].

Fluency:
The **investigations carried out by** [19] **on the production of bioethanol** from agricultural **residues** using **PROFER** Cellulosic or second **generation** bioethanol from LB **comprised** three **main steps:** pretreatment, hydrolysis and fermentation. **Pretreatment involves** the use of physical processes, chemical processes, **physico-chemical processes, biological processes,** and **various** combinations thereof to fractionate lignocellulose into its **constituents. It** will **break down** the lignin seal **in order to improve the enzyme's** access to cellulose [29, 30], reduction of**Cellulose** crystallinity [31, 32], an increase in the surface [33, 34] and **the porosity** [35, 36] of **the pretreated** substrates, **which leads to** increased hydrolysis**. During** hydrolysis, cellulose and hemicelluloses are broken down **into intomonomic** sugars **by adding** acids or enzymes such as cellulose. **Enzymatic** hydrolysis offers advantages over acids, such as low energy **consumption** due to the requirement **of the gentle process,** sugar **with high yield** and **undesired** waste. **Enzymatic The hydrolysis** of cellulose is **influenced** by **substrate properties** such as porosity, **crystallinity of the cellulose** fiber and **the degree** of **cellulosePolymerization** as well as lignin and hemicellulose content [37, 38], optim**al mixing** [39], **concentration of substrate** and end product, **enzymatic** activity and reaction **conditions** such as pH **value and** temperature [40, 41]**.**

Creative:
Research on the production of bioethanol from agricultural **residues** using **PROFER cellulose** or **LB second-generation** bioethanol **[19] includes** three **main steps:** pretreatment, hydrolysis and fermentation. **Pretreatment involves** the use of physical processes, chemical **processes,** physicochemical **processes,** biological **processes,** and **various** combinations thereof to **separate** lignocellulose into its components. **This will** lead to the **destruction of lignin fillers, thereby improving the absorption** of cellulose **by enzymes [29,** 30], **thereby reducing the content** of **lignin.The** crystallinity **of cellulose [31,** 32], **the** increase in surface area [33, 34] and **the pretreatment of porosity** [35, 36] **increase the hydrolyzed** substrate. **In the hydrolysis process,** cellulose and hemicellulose are broken down into **monomeric** sugars **by adding** acids or enzymes **(such** as **cellulose). Enzymatic** hydrolysis **has** advantages over acid, such as mild process **requirements, low energy consumption, high** sugar yield and **poor** waste. **The hydrolysis** of cellulose **depends on the characteristics** of **the matrix,** such as porosity, **crystallinity and grade of cellulose** fibers**The polymerization and content of** lignin and hemicellulose [37, 38], optim**al mixing** [39], substrate and **final** product concentration, enzyme activity and reaction **conditions,** such as pH and temperature [40, 41]**.**

0,65 % :

Fig. 10. Graph of alcohol content optimization results

This work was successful in obtaining bioethanol from the waste of wheat flour. The liquid waste of wheat flour was hydrolyzed by using H_2SO_4, producing 11% glucose in a form of the filtrate. This filtrate was fermented with Turbo yeast in various concentrations and fermentation times, producing optimum bioethanol of 37%. This work presents the optimum condition to obtain a high ethanol concentration (37% (v/v)), which was 10% Turbo yeast with fermentation time of 5 days. Modeling with RSM using Minitab 17 application shows the optimum results acquired were 11.66 (% v/v) Turbo yeast, and fermentation time of 5 days resulted in a bioethanol content of 37.21% (v/v).

Simple:

Advance:

Fluency:

Creative:

Simple:

Advance:

Fluency:

Creative:

Simple:

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Fluency:

Creative:

Process Fermentation of Filtrate Bamboo with Saccharomyces Cerevisiae and Zymomonas Mobilis

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Process Fermentation of Filtrate Bamboo with Saccharomyces Cerevisiae and Zymomonas Mobilis

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Abstract. Fermentation is the process of the formation of ethanol from glucose by using enzymes. Bamboo is one of the materials containing glucose is high enough, that is previously done hydrolysis in advance. Bamboo used when the hydrolysis process of bamboo that does not include lignin and the pentose done process of pretreatment and not lignification. The purpose of this research is to produce ethanol as a raw material substitution of bioethanol, knowing pentose and dirt left in the bamboo. Therefore, need to be studied in the future, with the best process, that we used biological processes that can optimize the production of ethanol. The use of the enzyme (Saccharomyces Cerevisiae and Zymomonas Mobilis) is also significant because of the optimum enzyme conditions. Temperature, pH, and the yeast with optimal conditions when it can raise the level of his work. The fermentation process at temperature 25 C and 45 C, the filtrate is 500 ml solution of bamboo and the stirring speed of 200 rpm. The variable composed enzyme with a ratio (v/v) of 0.25 to 0.75. Resulting from the fermentation processed can produce ethanol with a yield 30.5% and 36% of the weight of the bamboo. The result of the process of fermentation obtained bioethanol with low ethanol yield of 10-15%, which requires the flash distillation process to obtain yield bioethanol technical 90-95%.

1. Introduction

Biomass from plants has declared as an alternative raw material for gasoline fuel substitution in the form of bioethanol, bioethanol obtained from biomass and bioenergy crops has proclaimed as one of the feasible alternatives as gasoline fuel [1]. Sustainable bioethanol from rice straw [2]. The technology for lignocellulose ethanol production relies mainly on pre-treatment, chemical or enzymatic hydrolysis, fermentation and product separation or distillation. An appropriate pretreatment strategy is essential for the efficient enzyme hydrolysis of lignocellulose biomass as lignin hinders the scarification process. Various pretreatment approaches have exploited in the past such as acid or alkali pretreatment, hydrogen peroxide pretreatment, steam explosion, liquid hot water, ammonia fiber expansion pretreatment, sodium chlorite pretreatment, and biological pretreatment [3].

The research conducted to evaluate acid pretreatment from hydroxide paper waste as material for bioethanol production, optimized sulfuric acid hydrolysis, fermentation process of hydroxide acid of paper waste by using Pichia Stipites. The ethanol content obtained at 77.54%. By one more distillation process, the ethanol content received at the level of 95-96% [4]. Chemical pretreatment of lignocellulose biomass with Sulphur (H_2SO_4) and phosphorus (H_3PO_4) acids used since they are relatively cheap and efficient in hydrolyzing lignocellulose, though the letter gives a milder effect and is more benign to the environment. Hydrochloric (HCl) acid is more volatile and more natural to recover and attacks biomass better than H_2SO_4 [5]. Similarly, nitric acid (HNO_3) possesses good cellulose to sugar conversion rates [6]. However, both acids are expensive compared to Sulphur acid. Pretreatment of lignocellulose has received considerable research globally due to its affluence on the technical, economic and environmental sustainability of cellulose ethanol production. These paper reviews know, and emerging chemical pretreatment methods, the combination of chemical pretreatment with other ways to improve carbohydrate preservation reduce formation to degradation product, achieve high sugar yield at mild reaction conditions, reduce solvent loads and enzyme dose, reduce waste generation [7]. Technical and economic evaluation of bioethanol production from lignocellulose residues, a case of sugarcane and blue agave bagasse [8].

Initiatives of the future for lignin in biomass to bioethanol, pretreatment technologies to separate the main tree biopolymers (cellulose, hemicellulose, and lignin) [9]. Pretreatment for hydrogen and bioethanol production from olive oil waste products was ethanol yield 5.4 % treatment with 1.75 w/v Sulphur acid and heated it at 140 0C for 10 min, and was ethanol yield 5.0 % no pretreatment [10]. Pretreatment followed with simultaneous scarification and fermentation on bioconversion of microcrystalline cellulose for bioethanol production, the yield value of 67 % bioethanol bioconversion [11]. A sustainable feedstock bioethanol production, cellulose hydrolysis was microwave irradiation using hydrochloric acid as catalyst, fermentation with yeast (Saccharomyces cerevisiae), modest reaction conditions (2.38 M acid concentration), irradiation time 7 min, and yield of 0,67 g glucose / g cellulose [12]. Elements contained in the lignocellulose biomass of the plants are usually used lignocellulose biomass, a potential for bioethanol production globally. Agriculture (softwood), forestry (pretreatment method obtained ethanol content below 16%.

The purpose of dilute acid pretreatment is the removal of hemicelluloses and the recovery of the sugar component. Among all pretreatment methods, the acid pretreatment methods of biomass with dilute sulfuric acid has long recognized as a critical step of removing the hemicellulose fraction from the lignocellulose substrate to economize the biological conversion of cellulosic biomass to ethanol [20]. The research conducted by [21] about ethanol production from sago pith waste (SPW) using microwave hydrothermal hydrolysis catalyzed by carbon dioxide, resulted in higher energy saving compared to previous techniques in the absence of enzymes, acid or base catalyst. They obtained ethanol content below 15.6%. Bioethanol production from lignocellulosic biomass involves different step such as pretreatment, hydrolysis, fermentation and ethanol recovery [26]. The technology for lignocellulosic ethanol production relies mainly on pre-treatment, chemical or enzymatic hydrolysis, fermentation and product separation or distillation. An appropriate pretreatment strategy is essential for the efficient enzyme hydrolysis of lignocellulosic biomass as lignin hinders the saccharification process. Various pre-treatment approaches exploited in the past such as acid or alkali pretreatment, hydrogen peroxide pretreatment, steam explosion, liquid hot water, ammonia fiber expansion pretreatment [27].

Bioethanol production from the liquid waste of rise flour using fermentation by Saccharomyces, a maximum of 23.8% glucose and 40.5% ethanol yield, the developed technique for liquid waste of rise flour resulted in higher energy saving compared to the previous method in the absence of enzymes, acid or base catalyst [28]. hardwood), and industrial waste are a significant lignocellulose biomass for bioethanol production. The lignocellulose biomass is one of the potential main sources for economic bioethanol production globally. Agricultural, forestry (soft and hardwoods) and industrial wastes are the major lignocellulose biomasses [13]. The bioethanol production from lignocellulose biomass using process pretreatment, hydrolysis, fermentation, and recovery of ethanol, was obtained by ethanol under 16% v/v, with the distillation process will again be derived ethanol 95-96% v/v. The research conducted bioethanol production from lignocellulose biomass by using the pretreatment process, hydrolysis, fermentation, and ethanol recovery. Therefore, ethanol content obtained in the level below 16%, and by one more distillation process the ethanol content would receive at the level of 95-96% v/v [18].

The research conducted by [19] about bioethanol production from agricultural waste using PROFER Cellulosic or second generation (SG) bioethanol produced from lignocellulosic biomass (LB) in three main steps: pretreatment, hydrolysis, and fermentation. Pretreatment involves the use of physical processes, chemical methods, physic-chemical processes, biological methods, and several combinations thereof to fractionate the lignocellulose into its components. It results in the disruption of lignin seal to increase enzyme access to holo-cellulose [29, 30], reduction of cellulose crystallinity [31, 32], an increase in the surface area [33, 34] and porosity [35, 36] of pretreated substrates, resulting in increased hydrolysis rate. In hydrolysis, cellulose and hemicelluloses are broken down into monomeric sugars via the addition of acids or enzymes such as cellulose. Enzymatic hydrolysis offers advantages over acids such as low energy consumption due to the mild process requirement, high sugar yield, and no unwanted wastes. Enzymatic hydrolysis of cellulose affected by properties of the substrate such as porosity, cellulose fibre crystallinity, and degree of polymerization, as well as lignin and hemicellulose content [37, 38], optimum mixing [39], substrate and end-product concentration, enzyme activity, reaction conditions such as pH and temperature [40, 41].

From the previous research, it knows bioethanol from cellulose resulted in good bioethanol. The study was to search alternative material, review hydrolysis process, fermentation process to gain bioethanol product with a high level of ethanol. The originality of this research was the second generation that was bulrush, by using two methods (hydrolysis and fermentation) simultaneously, used two enzymes [Saccharomyces Cerevisiae (SC) and Zymomonas Mobilis (ZM)], and technical ethanol production with the level of 10-15% as the technical ethanol.

2. Experimental

From the result of laboratory analysis, it known ethanol forming elements were cellulose, glucose, and impurities. The average concentration of cellulose was 48% in bamboo, and glucose was 5 % and impurities.

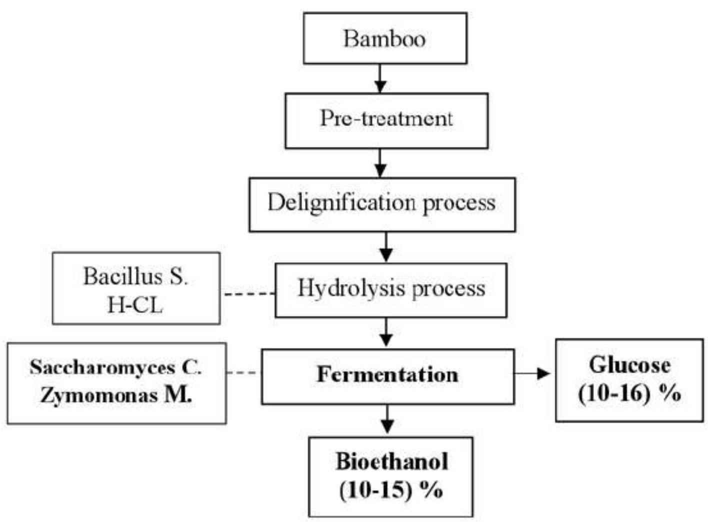

Figure 1. Glucose and bioethanol production flow used fermentation process

The pieces and refined fiber of bamboo with the approximate length of 5 cm and polished thread 200 mesh done to obtain the high level of glucose and cellulose during the hydrolyzed process by Bacillus and H-Cl. The quality bioethanol product determined by various influencing parameters such as the acidity (pH), the volume ratio of H-Cl to bamboo, the volume ratio of Bacillus Subtitles to the filtrate, the volume ratio of the enzyme (Saccharomyces C. and Zymomonas M.) to the filtrate, and fermentation time. Laboratory analysis did the quality analysis of raw materials and bioethanol product. The study conducted on the instrumentation and gravimetric analysis by using Gas Chromatography

(GC) and Spectrophotometer, which analyzed items were the concentration of bamboo, glucose, ethanol, H-Cl, and impurities.

Hydrolysis process in Figure 1 done in stable condition: temperature of 30 C, water volume in 7 liters, and hydrolysis time in 1 hour with 200 rotations per minute (RPM). For the changing condition: bamboo weight of 50, 100, 150, 200, 250 (grams), the ratio of bacillus to filtrate volume 1:2; 5:4; 10:7 and H-Cl solution volume 10, 20, 30, 40, 50 (ml). The level of glucose in hydrolysis filtrate yield was analyzed before the fermentation process done previous research [42]. Fermentation process in Figure 1 done in stable condition: filtrate bamboo ratio of the varies Saccharomyces C. and Zymomonas M.: 5, 9, 13 (% v/v), fermentation time 4, 6, 8, 10, 12 days. Filtrate rate influences the residual glucose levels, obtained maximum residual glucose levels (1,3 - 3) %, and this is because in the tank hydrolysis reactor and the amount of filtrate starter Saccharomyces C. and Zymomonas M. in still little so that the fermentation process is not optimal. With the increasing amount of filtrate hydrolysis and starter Saccharomyces Cerevisiae then the smaller the residual glucose, because it fermented into ethanol.

2. Results and Discussion

Bamboo using as a study material derived from bamboo crops in the surrounding area. Assessment method is done, by doing a survey and laboratory analysis to obtain some data about the quality and quantity of the available bamboo. The expected result was data about the quality and bamboo quantity before processing to be ethanol. Based on the results of laboratory analysis, it is known, that ethanol forming elements were cellulose and glucose. The average concentration of cellulose was 48.1 %, glucose was 4.8%, and impurities. If the entire cellulose hydrolyzed completely, it will be obtaining the glucose levels of 53%.

The results of fermentation process with ratio enzyme again filtrate cellulose as:

Table 1. Ethanol level and yield on fermentation process

Ratio Filtrate Sellulose (%v/v)	Fermentation time (day)	Glucose level Fermentation (%)		Ethanol Level Fermentation (%)	
		SC	ZM	SC	ZM
5	4	4.82	5.76	10.5	10.0
	6	5.30	5.23	11.0	12.3
	8	5.70	5.27	9.0	13.4
	10	5.78	5.54	12.5	10.3
	12	5.81	5.77	11.0	10.3
9	4	5.08	5.90	12.0	9.1
	6	5.68	5.76	14.5	13.2
	8	7.63	6.03	14.0	14.9
	10	7.78	6.78	15.0	13.5
	12	7.98	7.91	14.0	12.6
13	4	7.41	6.88	14.0	13.0
	6	8.35	7.95	14.5	13.7
	8	9.56	8.77	15.0	14.5
	10	9.87	9.05	14.5	12.3
	12	9.88	9.35	13.0	12.8

The pieces and refined fiber of bamboo with an approximate length of 5 cm and polished thread 200 mesh were done to obtain the high levels of glucose and cellulose before it hydrolyzed by Bacillus and H-Cl solution. Bamboo should be made in powder form so that cellulose hydrolyzed perfectly. However, that process took a higher cost. Besides, bulrush in the powder form could suffer the physical destruction, thus causing the damage of the glucose group. The drying process of bulrush was naturally done first in the room temperature. The drying process was done in an oven at 1000C for 3 hours. These done for cost savings. The drying process aimed to reduce the water content in ethanol. The water level that was permitted by Standart National Indonesia (SNI) was 1%. The decreasing of pH from pretreatment material was affected by the addition of H-Cl volume 7%v/v because the requiring pH for fermentation process was 4,5. Before doing the hydrolysis process, the pH of filtrate measured according to the terms of the fermentation process that is approximately 4.5. To obtain pH 4.5, the addition of Na-OH done if the pH of the filtrate was under 4.5 and the addition of citric acid if the filtrate pH was above 4.5. Filtrate rate influences the residual glucose levels, for a number of starter Saccharomyces C. 5, 9, and 13% v/v, obtained maximum residual glucose levels (1,3 - 3) %, this is because in the tank hydrolysis reactor and the amount of filtrate starter Saccharomyces C. still little, so that the fermentation process is not optimal. With the increasing amount of filtrate hydrolysis and starter Saccharomyces C. then the smaller the residual glucose, because it fermented into ethanol.

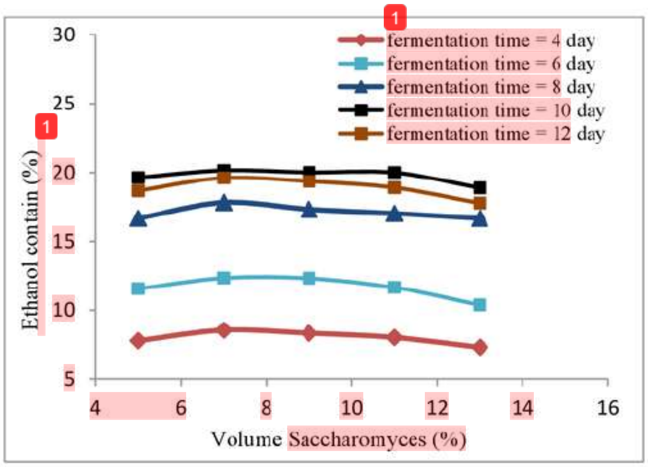

Figure 2. Effect Saccharomyces volume on the remaining ethanol contain

After analysis glucose levels in the rest of the fermentation process, with the addition of Saccharomyces C. 7 % of the volume of fluid (filtrate) showed small residual glucose levels compared to the addition of starter 5, 11 and 13 %. These are because the preliminary research has been conducted by following the Journal and the acquisition of 7% of the fluid volume. Filtrate rate influences the residual glucose levels, for a number of starter Saccharomyces C. 5, 9, and 13%, obtained maximum residual glucose levels (1.5-10) %, this was due to the amount in the tank reactor filtrate hydrolysis and starter Saccharomyces C. still little, so that the fermentation process is not optimal. With the increasing amount of filtrate hydrolysis and starter Saccharomyces C. then the smaller the residual glucose, because it fermented into ethanol.

5. Conclusion

Fermentation process from raw materials (bamboo) to produce bioethanol, glucose levels obtained in the fermentation process as (5-10) % for filtrate cellulose, levels of ethanol in the fermentation process equal 10 to 15 %. The Saccharomyces C. had higher glucose and bioethanol levels results of Zymomonas M., but durability Zymomonas M. stronger in a fermentation process.

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15	C-11	Response Surface Analysis on the Microwave Integrated-Rumen Based Extraction of Natural Vanillin from Cured Vanilla Pods	Vita Paramita, Mohamad Endy Yulianto, Altaera Yuha Syahputri

16	C-12	The Effect of Ionic Strength on Protonation Constant of Monoethanolamine in Water	S Ma'mun, A Chafidz, E Indrayanto and P K Setiawan
17	C-13	Browning Prevention of Chips from Freshly Harvested Porang (Amorphophallus oncophyllus) Tubers through Immersion in Ascorbic Acid Solutions at Various Times	A C Kumoro, M Amyranti, D S Retnowati and R Ratnawati
18	C-14	Composite Of Ramie, Cotton, And Rayon Double Ply Combination For Bulletproof Vests Body Armor	Kanthi Setyani and Nita Aryanti
19	C-15	Flux Decline and Blocking Mechanism in Ultrafiltration of Glycerine Rich Solution	Nita Aryanti, Awali Sir Kautsar harivram, Dyah Hesti Wardhani
20	C-17	Production Of Natural Colorant Powder From Clitoria Ternatea L. Using Tray Dryer Which Is Dehumidified By Zeolite	Mohamad Djaeni, Friska Mauludifia, Shanintya Dhivya Astrinia, Kania Adelia Meiranti
21	C-18	Effect of Onion (Allium Cepa L.) Drying Using Hot Air Dehumidified by Activated Carbon, Silica gel and Zeolite	Mohamad Djaeni, Arninda Mahar Perdanianti
22	C-19	Kinetics of Colour Degradation during Application of Dried Colorant from Roselle Extract with Foaming Agent	Mohamad Djaeni and Febiani Dwi Utari
23	C-20	Effect Of Sago Starch Concentrations, Stirring Speeds, And Lemon Grass Oil Concentration For Edible Film Production Using Solvent Casting Method	Herry Santosa, M. Djaeni, Ratnawati, Nur Rokhati, Asri Pertiwi Setiatun, Afriyanti
24	C-21	Optimization Processing Of Batik Waste Using Used Iron Metal As Anodes With Electroadsorbsi Methods (With The Multi Varian Regression Model)	Suhartana, Purwanto, Adi Darmawan
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26	C-24	Development of Cellulose Acetate – PVP blend membrane and UV irradiation treatment to increase membrane selectivity for clove oil purification	Tutuk Djoko Kusworo, Anggun Anaulia Siahaan, Goldi Kharisma Iskandar, Dani Puji Utomo
27	D-01	The Effect of Saccharomyces cerevisiae Concentrations on Second Generation Bioethanol Production from Oil Palm Frond	Meliana Dewi, A Ahmad, S R Muria
28	D-02	Production Of Second Generation Bioethanol From Palm Fruit Fiber Biomass Using Saccharomyces cerevisiae	Masroah Tuljannah,A Ahmad, S R Muria,
29	D-03	Stability of Mixture Honey, Black Seed Oil and Olive Oil With Tween 80 as Emulsifier	M Sahlan, A Ferdianti, A B Wicaksono, H Hermasnyah, and A Wijanarko
30	D-04	Fructose Syrup Production from Tapioca Solid Waste (Onggok) By Using Enzymatic Hydrolysis In Various pH And Isomerization Process	Ayu Ratna Permanasari, Saripudin,Maulani, L Ramdhayani
31	D-05	Process Fermentation of Filtrate Bamboo with Saccharomyces Cerevisiae and Zymomonas Mobilis	N K Sari, D Ernawati
32	D-06	Effectiveness Of Eugenol As An Antibacterial Toward Staphylococcus Epidermidis	Rudi Firyanto, Ery Fatarina P., Nur Azizah

33	D-07	Production of bioethanol from suweg (Amorphophallus campanulatus B) by hydrolysis at low temperature and fermentation using Saccharomyces cerevisiae	Hargono, Bakti Jos, Andri Cahyo Kumoro, Kristinah Haryani
34	D-08	Effect of KOH as Deacetylation Agent on Physicochemical Properties of Glucomannan	Dyah H Wardhani, H Cahyono, N Aryanti and DR Pangestuti
35	D-09	The Quality and physical properties of dried noodle fortified by phycocyanin	Hadiyanto
36	E-01	Adsorption of Cobalt-60 (II) on Silica Xerogel from Rice Husk	Noor Anis Kundari, M. G Permadi, K. Megasari, G Nurliati
37	E-02	Betaine-based deep eutectic solvents with diol, acid and amine hydrogen bond donors for carbon dioxide absorption	Kamarza Mulia,Evan Libriandy, Elsa Anisa Krisanti and Nasruddin
38	E-04	Recycling and processing of solid waste into products in the cosmetic packaging industry	P. Purwanto and A.D. Permana-Citra
39	E-06	Banana peals as adsorbent	Priyono Kusumo, Rudi Firyanto, Rizqi Dwi Jayanti
40	E-07	Reduction Of Cod Content In Domestic Waste Water Using Combination Activated Sludge Method - Active Carbon Continuously	Mukhtar G, Retno I, Fajar N, Satria A
41	E-08	Production of Fish Feed from Soy Residue and Shrimp Waste using Cassava Starch as Binding Agent	Zulhaq Dahri Siqhny, Siswo Sumardiono
42	F-02	Condensate water as a compressor discharge cooler to generate subcooling on the residential air conditioning using R32 as refrigeran	A S Margana, S Hidayat, Kasni Sumeru
43	F-03	Kinetics and Equilibrium Studies of Electro Adsorption of Remazol Red on Modified Stainless Steel Electrode	Ristiyanti Riska, Purwanto
44	F-04	Ultrasonic Pretreatment On Biogas Production From Wood-Dust Mahogany (Swietenia Mahagoni) With Solid-State Anaerobic Digestion Method: Effect Of Time And Temperature Pretreatment	Hanif Ardhiansyah, Budiyono and Siswo Sumardiono
45	F-08	Simplex Lattice Design Method for the Optimization of Non-Edible Oils Mixture Composition as Feedstock for Biodiesel Synthesis Using Reactive Distillation	Ratna D. Kusumaningtyas, Bayu Triwibowo, Reshita A. Ramadhani, Dody H.S. Riyadi, I Istadi
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48	F-11	Drying rate and efficiency energy analysis of paddy drying using dehumidification with zeolite	M Djaeni, F Irfandy and F D Utari
49	G-02	Production Technology and Ultilization of Nano Cellulose	Henny Herawati
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51	G-04	Morphological and Optical Properties of Poly (lactic acid) Bionanocomposite Film Reinforced with Oil Palm Empty Fruit Bunch Nanocrystalline Cellulose	E Indarti, Marwan and W D. Wan Rosli
52	G-05	Nano-ZnO impregnated-cellulose acetate hybrid membrane for increasing eugenol content in clove oil	Tutuk Djoko Kusworo, Anggun Anaulia Siahaan, Goldi Kharisma Iskandar, Dani Puji Utomo
53	G-07	Effect of Combination High Alumunium Doping in Zinc Oxide and Ozonation to Degrade dyes of Rhodamine B and direct blue 71	Heri Susanto
54	H-01	Isolation of Lignin from Rice Husk at Low Temperature	Anwar Ma'ruf,Bambang Pramudono,Nita Aryanti, Aulia Usriyati
55	H-02	Gas permeation properties and preparation of carbon membrane by PECVD method using indene as precursor	Myat Kyaw, Nathaniel Dugos, Shinsuki Mori , Susan Roces ,Arnel Beltran, Shunsuke Suzuki
56	I-01	Study of Plasma Electrolysis Method on Starch-Based Hybrid Latex Synthesis"	Nelson Saksono, Andream Bais Jr, Edward Gustaf,Jhangir Desfrandanta,Mochamad Kholid
57	I-02	Effect of iron and manganese concentration on the sulfate-reducing process in acid mine drainage	Nurandani hardyanti,Sudarno Utomo, , Katrin Serafina, Angelica Oktaviana
58	I-04	Effect of pH on Depolimerization of k-Caragenan by Ultrasound, Ozon and Their Combination	Aji Prasetyaningrum, Bakti Jos, Yudhy Dharmawan, Ratnawati Ratnawati, S. Riyandita, and R. Scesario
59	I-05	Effect of Temperature and Reaction Time on the Swelling Power and Solubility of Gadung (Dioscorea hispida Dennst) Tuber Starch during Heat Moisture Treatment Process	A C Kumoro, D S Retnowati, R Ratnawati and M Widiyanti
60	L-02	Scale-up simulation and economic evaluation of encapsulated eugenol with casein micelle using spray drying method	Aryo Bayu Wicaksono, H Hermasnyah, A Wijanarko, and M Sahlan
61	L-03	Artificial Rice Production Using Granulation, Hot Extrution, and Cold Extrution Methods : An Overview	Heny Kusumayanti, Siswo Sumardiono, and Bakti Jos
62	L-04	The Modification of Sago Starch by Combination of Lactic Acid Hydrolysis and H2O2 Oxidation Methods to Increase Baking Expansion	Siswo Sumardiono, Assalaam U. Abdurahman, Arland, Bakti Jos and Isti Pudjihastuti
63	L-05	The effect of temperature and composition of composite materials flour (Cassava, Dioscorea Esculenta, Corn, and Canavalia Ensiformis) on Analog Rice Production by Using Hot Extrusion Method	Siswo Sumardiono, Arsy Novitasari, Fiky Z.Awaliyah, Herry Santosa, and Isti Pudjihastuti
64	L-06	Central Composite Design for Optimation of Starch-Based Bioplastic with Bamboo Microfibrillated Cellulose as Reinforcement Assisted by Potassium Chloride	Silviana Silviana, Puji Rahayu

65	L-07	A Comparative Study on FeCrAl Alloy and NiCrSi Alloy Materials as Heating Element	Veinard Vingtsabta, Abdul Syakur
66	L-08	Application of an Integrated Cooking Pan in Sambal Production	Dyah Hesti Wardhani, Nita Aryanti, Luqman Buchori, Heri Cahyono
67	L-09	Synthesis of Cassava Bagasse Starch-Based Biocomposite Reinforced Woven Bamboo Fibre with Lime Juice as Crosslinker and Epoxidized Waste Cooking Oil (EWCO) as Bioplasticizer	Silviana Silviana, M Chilmi
68	L-10	Enhanced conductivity of supercapacitor based PAni-GO-Cellulose-Lanthanum using modification of Al current collector surface and gamma irradiation	D Swantomo, C A L Wijaya, and Sigit

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Process Fermentation of Filtrate Bamboo with Saccharomyces Cerevisiae and Zymomonas Mobilis

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Abstract. Fermentation is the process of the formation of ethanol from glucose by using enzymes. Bamboo is one of the materials containing glucose is high enough, that is previously done hydrolysis in advance. Bamboo used when the hydrolysis process of bamboo that does not include lignin and the pentose done process of pretreatment and not lignification. The purpose of this research is to produce ethanol as a raw material substitution of bioethanol, knowing pentose and dirt left in the bamboo. Therefore, need to be studied in the future, with the best process, that we used biological processes that can optimize the production of ethanol. The use of the enzyme (Saccharomyces Cerevisiae and Zymomonas Mobilis) is also significant because of the optimum enzyme conditions. Temperature, pH, and the yeast with optimal conditions when it can raise the level of his work. The fermentation process at temperature 25 C and 45 C, the filtrate is 500 ml solution of bamboo and the stirring speed of 200 rpm. The variable composed enzyme with a ratio (v/v) of 0.25 to 0.75. Resulting from the fermentation processed can produce ethanol with a yield 30.5% and 36% of the weight of the bamboo. The result of the process of fermentation obtained bioethanol with low ethanol yield of 10-15%, which requires the flash distillation process to obtain yield bioethanol technical 90-95%.

1. Introduction

Biomass from plants has declared as an alternative raw material for gasoline fuel substitution in the form of bioethanol, bioethanol obtained from biomass and bioenergy crops has proclaimed as one of the feasible alternatives as gasoline fuel [1]. Sustainable bioethanol from rice straw [2]. Ethanol production from lignocellulose by hydrolysis process chemically or enzymatically, first conducted the process of pretreatment, the next process is the process of fermentation and distillation process. Pretreatment processes, the most important is to remove the lignin and pentosan, which can dissuade lignin and pentosan skarifikasi process. Various approaches have been performed earlier as pretreatment pretreatment in acids, bases, ammonia, sodium chlorite, and biological [3].

The research conducted to evaluate acid pretreatment from hydroxide paper waste as material for bioethanol production, optimized sulfuric acid hydrolysis, fermentation process of hydroxide acid of paper waste by using Pichia Stipites. The ethanol content obtained at 77.54%. By one more distillation process, the ethanol content received at the level of 95-96% [4]. Chemical pretreatment of lignocellulose biomass with Sulphur (H_2SO_4) and phosphorus (H_3PO_4) acids used since they are relatively cheap and efficient in hydrolyzing lignocellulose, though the letter gives a milder effect and is more benign to the environment. Hydrochloric (HCl) acid is more volatile and more natural to recover and attacks biomass better than H_2SO_4 [5]. Similarly, nitric acid (HNO_3) possesses good cellulose to sugar conversion rates [6]. However, both acids are expensive compared to Sulphur acid. Pretreatment of lignocellulose has received considerable research globally due to its affluence on the technical, economic and environmental sustainability of cellulose ethanol production. These paper reviews know, and emerging chemical pretreatment methods, the combination of chemical pretreatment with other ways to improve carbohydrate preservation reduce formation to degradation product, achieve high sugar yield at mild reaction conditions, reduce solvent loads and enzyme dose, reduce waste generation [7]. Technical and economic evaluation of bioethanol production from lignocellulose residues, a case of sugarcane and blue agave bagasse [8].

Initiatives of the future for lignin in biomass to bioethanol, pretreatment technologies to separate the main tree biopolymers (cellulose, hemicellulose, and lignin) [9]. Pretreatment for hydrogen and bioethanol production from olive oil waste products was ethanol yield 5.4 % treatment with 1.75 w/v Sulphur acid and heated it at 140 0C for 10 min, and was ethanol yield 5.0 % no pretreatment [10]. Pretreatment followed with simultaneous scarification and fermentation on bioconversion of microcrystalline cellulose for bioethanol production, the yield value of 67 % bioethanol bioconversion [11]. A sustainable feedstock bioethanol production, cellulose hydrolysis was microwave irradiation using hydrochloric acid as catalyst, fermentation with yeast (Saccharomyces cerevisiae), modest reaction conditions (2.38 M acid concentration), irradiation time 7 min, and yield of 0,67 g glucose / g cellulose [12]. Elements contained in the lignocellulose biomass of the plants are usually used lignocellulose biomass, a potential for bioethanol production globally. Agriculture (softwood), forestry (pretreatment method obtained ethanol content below 16%.

Pretreatment with dilute acid (sulfuric acid) eliminating the hemicellulose components and increase the sugar. Pretreatment method of sulfuric acid has long been recognized as an important step towards eliminating the hemicellulose fraction of lignocellulose substrates, and save the conversion of cellulosic biomass [20]. The research conducted by [21] about ethanol production from sago pith waste (SPW) using microwave hydrothermal hydrolysis catalyzed by carbon dioxide, resulted in higher energy saving compared to previous techniques in the absence of enzymes, acid or base catalyst. They obtained ethanol content below 15.6%. The production of bioethanol from lignocellulosic biomass through the different process steps, such as pretreatment, hydrolysis, fermentation, distillation and [26]. Ethanol production from lignocellulosic technologies largely determined by hydrolysis and pretreatment, whether chemical or biological [27].

Ethanol from the liquid waste of rise flour using fermentation by Saccharomyces, a maximum of 23.8% glucose and 40.5% ethanol yield, the developed technique for liquid waste of rise flour resulted in higher energy saving compared to the previous method in the absence of enzymes, acid or base catalyst [28]. hardwood), and industrial waste are a significant lignocellulose biomass for bioethanol production. The lignocellulose biomass is one of the potential main sources for economic bioethanol production globally. Agricultural, forestry (soft and hardwoods) and industrial wastes are the major lignocellulose biomasses [13]. The bioethanol production from lignocellulose biomass using process pretreatment, hydrolysis, fermentation, and recovery of ethanol, was obtained by ethanol under 16% v/v, with the distillation process will again be derived ethanol 95-96% v/v. The research conducted bioethanol production from lignocellulose biomass by using the pretreatment process, hydrolysis, fermentation, and ethanol recovery. Therefore, ethanol content obtained in the level below 16%, and by one more distillation process the ethanol content would receive at the level of 95-96% v/v [18].

The research conducted by [19] about bioethanol production from agricultural waste using PROFER Cellulosic or second generation (SG) bioethanol produced from lignocellulosic biomass (LB) in three main steps: pretreatment, hydrolysis, and fermentation. Pretreatment involves the use of physical processes, chemical methods, physic-chemical processes, biological methods, and several combinations thereof to fractionate the lignocellulose into its components. It results in the disruption of lignin seal to increase enzyme access to holo-cellulose [29, 30], reduction of cellulose crystallinity [31, 32], an increase in the surface area [33, 34] and porosity [35, 36] of pretreated substrates, resulting in increased hydrolysis rate. In hydrolysis, cellulose and hemicelluloses are broken down into monomeric sugars via the addition of acids or enzymes such as cellulose. Enzymatic hydrolysis offers advantages over acids such as low energy consumption due to the mild process requirement, high sugar yield, and no unwanted wastes. Enzymatic hydrolysis of cellulose affected by properties of the substrate such as porosity, cellulose fibre crystallinity, and degree of polymerization, as well as lignin and hemicellulose content [37, 38], optimum mixing [39], substrate and end-product concentration, enzyme activity, reaction conditions such as pH and temperature [40, 41].

From the previous research, it knows bioethanol from cellulose resulted in good bioethanol. The study was to search alternative material, review hydrolysis process, fermentation process to gain bioethanol product with a high level of ethanol. The originality of this research was the second generation that was bulrush, by using two methods (hydrolysis and fermentation) simultaneously, used two enzymes [Saccharomyces Cerevisiae (SC) and Zymomonas Mobilis (ZM)], and technical ethanol production with the level of 10-15% as the technical ethanol.

2. Experimental

The average concentration of cellulose was 48% in bamboo, and glucose was 5 % and impurities.

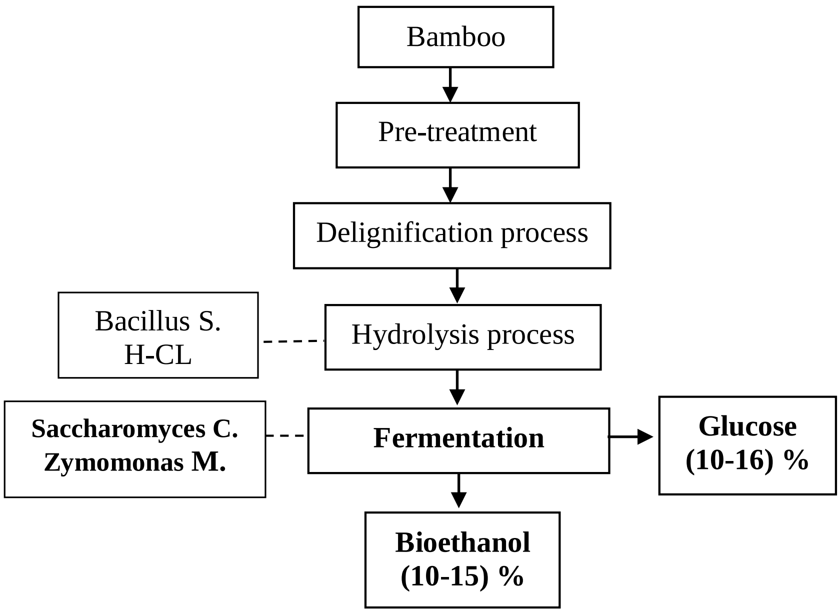

Figure 1. Glucose and bioethanol production flow used fermentation process

The pieces and refined fiber of bamboo with the approximate length of 5 cm and polished thread 200 mesh done to obtain the high level of glucose and cellulose during the hydrolyzed process by Bacillus and H-Cl. The quality bioethanol product determined by various influencing parameters such as the acidity (pH), the volume ratio of H-Cl to bamboo, the volume ratio of Bacillus Subtitles to the filtrate, the volume ratio of the enzyme (Saccharomyces C. and Zymomonas M.) to the filtrate, and fermentation time. Laboratory analysis did the quality analysis of raw materials and bioethanol product. The study conducted on the instrumentation and gravimetric analysis. Hydrolysis process in Figure 1 done in stable condition: temperature of 30 ^{0}C, water volume in 7 liters, and hydrolysis time in 1 hour with 200 rotations per minute (RPM). For the changing condition: bamboo weight of 50, 100, 150, 200, 250 (grams), the ratio of bacillus to filtrate volume 1:2; 5:4; 10:7 and H-Cl solution volume 10, 20, 30, 40, 50 (ml). The level of glucose in hydrolysis filtrate yield was analyzed before the fermentation process done previous research [42]. Fermentation process in Figure 1 done in stable condition: filtrate bamboo ratio of the varies Saccharomyces C. and Zymomonas M.: 5, 9, 13 (% v/v), fermentation time 4, 6, 8, 10, 12 days. Filtrate rate influences the residual glucose levels, obtained maximum residual glucose

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levels (1,3 - 3) %, and this is because in the tank hydrolysis reactor and the amount of filtrate starter Saccharomyces C. and Zymomonas M. in still little so that the fermentation process is not optimal. With the increasing amount of filtrate hydrolysis and starter Saccharomyces Cerevisiae then the smaller the residual glucose, because it fermented into ethanol.

3. Results and Discussion

Bamboo using as a study material derived from bamboo crops in the surrounding area. Assessment method is done, by doing a survey and laboratory analysis to obtain some data about the quality and quantity of the available bamboo. The expected result was data about the quality and bamboo quantity before processing to be ethanol. Ethanol forming elements were cellulose and glucose, the concentration of cellulose was 48.1 %, glucose was 4.8%, and impurities. If the entire cellulose hydrolyzed completely, it will be obtaining the glucose levels of 53%.
The results of fermentation process with ratio enzyme again filtrate cellulose as:

Table 1. Ethanol level and yield on fermentation process

Ratio Filtrate Sellulose (%v/v)	Fermentation time (day)	Glucose level Fermentation (%)		Ethanol Level Fermentation (%)	
		SC	ZM	SC	ZM
5	4	4.82	5.76	10.5	10.0
	6	5.30	5.23	11.0	12.3
	8	5.70	5.27	9.0	13.4
	10	5.78	5.54	12.5	10.3
	12	5.81	5.77	11.0	10.3
9	4	5.08	5.90	12.0	9.1
	6	5.68	5.76	14.5	13.2
	8	7.63	6.03	14.0	14.9
	10	7.78	6.78	15.0	13.5
	12	7.98	7.91	14.0	12.6
13	4	7.41	6.88	14.0	13.0
	6	8.35	7.95	14.5	13.7
	8	9.56	8.77	15.0	14.5
	10	9.87	9.05	14.5	12.3
	12	9.88	9.35	13.0	12.8

The pieces and refined fiber of bamboo with an approximate length of 5 cm and polished thread 200 mesh were done to obtain the high levels of glucose and cellulose before it hydrolyzed by Bacillus and H-Cl solution. Bamboo should be made in powder form so that cellulose hydrolyzed perfectly. However, that process took a higher cost. Besides, bulrush in the powder form could suffer the physical destruction, thus causing the damage of the glucose group. The drying process of bulrush was naturally done first in the room temperature. The drying process was done in an oven at 1000C for 3 hours. These done for cost savings. The drying process aimed to reduce the water content in ethanol. The water level that was permitted by Standart National Indonesia (SNI) was 1%. The decreasing of pH from pretreatment material was affected by the addition of H-Cl volume 7%v/v because the requiring pH for fermentation process was 4,5. Before doing the hydrolysis process, the pH of filtrate measured according to the terms of the fermentation process that is approximately 4.5. To obtain pH 4.5, the addition of Na-OH done if the pH of the filtrate was under 4.5 and the addition of citric acid if the filtrate pH was above

4.5. Filtrate rate influences the residual glucose levels, for a number of starter Saccharomyces C. 5, 9, and 13% v/v, obtained maximum residual glucose levels (1,3 - 3) %, this is because in the tank hydrolysis reactor and the amount of filtrate starter Saccharomyces C. still little, so that the fermentation process is not optimal. With the increasing amount of filtrate hydrolysis and starter Saccharomyces C. then the smaller the residual glucose, because it fermented into ethanol.

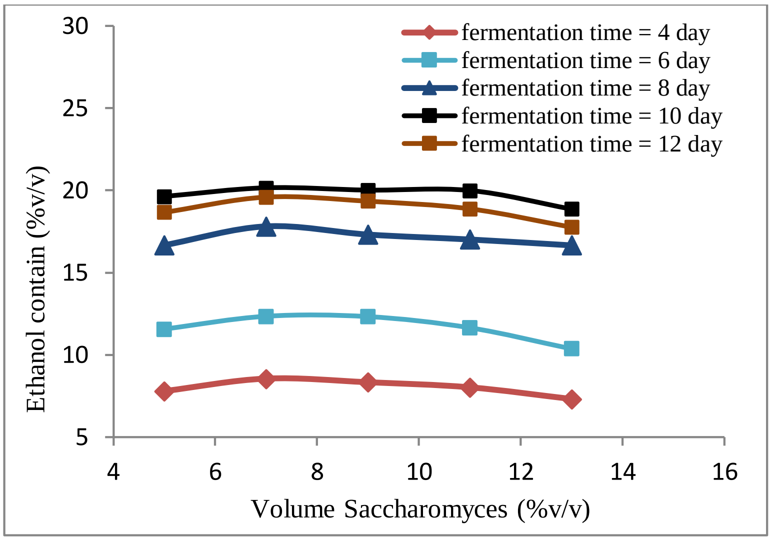

Figure 2. Effect Saccharomyces volume on the remaining ethanol contain

After analysis glucose levels in the rest of the fermentation process, with the addition of Saccharomyces C. 7 % of the volume of fluid (filtrate) showed small residual glucose levels compared to the addition of starter 5, 11 and 13 %. These are because the preliminary research has been conducted by following the Journal and the acquisition of 7% of the fluid volume. Filtrate rate influences the residual glucose levels, for a number of starter Saccharomyces C. 5, 9, and 13%, obtained maximum residual glucose levels (1.5-10) %, this was due to the amount in the tank reactor filtrate hydrolysis and starter Saccharomyces C. still little, so that the fermentation process is not optimal. With the increasing amount of filtrate hydrolysis and starter Saccharomyces C. then the smaller the residual glucose, because it fermented into ethanol.

4. Conclusion

Fermentation process from bamboo to bioethanol, glucose levels obtained in the fermentation process as (5-10) % for filtrate cellulose, levels of ethanol in the fermentation process equal 10 until 15 %. The Saccharomyces C. had higher glucose and bioethanol levels results of Zymomonas M., but durability Zymomonas M. stronger in a fermentation process.

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