Optimization of bioethanol production from tapioca flour waste through the addition of a starter and fermentation duration

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Abstract. The production of tapioca flour often results in waste problems that have not been handled properly. Utilization of solid waste tapioca flour as an alternative energy for bioenergy is an energy innovation that is renewable and environmentally friendly. The purpose of this study was to determine the bioethanol content of tapioca flour waste based on variations in the addition of a starter and variations in fermentation time. The research method used was an experimental method with quantitative data collection. One of the industrial wastes is solid tapioca starch waste in the form of onggok and cassava peels used as the sample. The procedures of this research were: isolation of cassava peels and onggok, hydrolysis and fermentation. The study was conducted with 8 variations of treatment, namely a mixture of cassava skin-onggok plus a 24 hours starter, cassava skin plus a 24 hours starter, a mixture of cassava-onggok skin plus a starter, cassava skin plus starter, a mixture of cassava skin-onggok starter for 24 hours and Cassava skin D-1 starter 24 hours. Duration of fermentation treatments on the sample were 7 days, 14 days, 21 days, and 30 days. The results showed that the mixture of cassava peel and onggok added directly to the starter without waiting 24 hours was the most significant, the alcohol percentage is 55% and the glucose percentage is 17% with a 7 days of fermentation. It can be concluded that solid tapioca starch waste in the form of cassava peels and onggok had potential in bioethanol production with the addition of an optimal starter and not too long fermentation time.

1. Introduction
Bioethanol is a bioenergy that is renewable, low in pollution, and can be produced from materials that contain sugar and starch such as corn, potatoes, wheat, sugar cane, molasses and others. Bioenergy production from cultivated crops requires higher costs when compared to energy production from petroleum. Therefore we need an alternative source of cheap and abundant raw materials [1]. Cheap and abundant raw materials were wasted, one of which was found in the tapioca flour industry process [2]. Tapioca starch waste such as cassava peels and onggok (solid waste) can be used as an energy source in ethanol form [3]. The need for ethanol is increasing both as a solvent [4], disinfectant [5], raw material for chemical plants and as an alternative energy substitute for fuel [6]. Biofire gel made from bioethanol from tapioca starch waste can support zero waste. Biofire gel is safer than liquid fuel...
(not easily spilled and evaporated). The development of biofire gel made from waste is expected can increase the economic value of cassava peels and onggok.

This research had 4 main aspects, namely science and technology development, social, economic and institutional development. Aspects of science and technology development, this research had a specific objective to produce bioethanol that cheap, safe to produce and reduce waste from the tapioca flour industry to zero waste [7] which was divided into several stages, namely 1) isolation of cassava peels and onggok; 2) distillation of bioethanol from cassava peels and onggok using reflux column distillation; ethanol content test.

2. Method

At the research stage in 2020 that had been done was the process of ethanol production from tapioca flour waste directly into the application of liquid fuel products in gel form. This production stage started from (1) Preparation of raw materials, obtained from onggok and cassava peels which were cleaned and crushed to break down the flour composition; (2) Liquefaction and saccharification, at this stage the raw materials were converted into complex sugars using alpha amylase enzymes through a heating process at 90°C (hydrolysis). In this condition the enzymes will break down the flour structure chemically into dextrins. The liquefaction process was characterized by a parameter in which the slurry turns to be more liquid like soup. Then saccharification was carried out by cooling the slurry, adjusting the optimum pH of the enzyme, adding glucose amylase enzyme [20]. This process was completed by simple sugar produced level test; (3) Fermentation, flour had turned into simple sugars (glucose and partly fructose) with a sugar content of 5-12%. Next step, yeast was mixed in the liquid and left in a fermenter at a temperature of 27-32°C for 5-7 days; (4) Distillation, the distillation stage, was done by separating the alcohol from the fermented beer. Distillation using reflux column model and distillation technique. The result was ethanol content that can reach 90-95% through two stages of distillation.

3. Results and Discussion

Based on the research results, observational data had been obtained from the research stages, including the stages of raw material isolation, liquefaction and saccharification, fermentation, distillation and bioethanol gel manufacture. The results of tapioca flour waste isolation process to the fermentation process, the percentage of glucose was obtained based on 8 predetermined treatments. The 8 treatments are:

Treatment A: cassava skin and onggok without starter
Treatment B: onggok without starter
Treatment C: cassava skin and onggok plus starter before fermentation (D-1)
Treatment D: onggok plus starter before fermentation (D-1)
Treatment E: cassava peel and onggok added with a starter during the fermentation day D (D-Day)
Treatment F: onggok added with starter during fermentation on D-day
Treatment G: cassava skin and onggok plus starter after fermentation (D + 1)
Treatment H: onggok plus starter after fermentation (D + 1)

Based on the determination of the treatment, the results of glucose percentage before and after fermentation process were obtained (Figure 1).

3.1. Process of pre-post fermentation treatment

The effects of fermentation on various predetermined treatments, are shown in Figure 1.
Figure 1 shows that the effect of fermentation on the sample had a significant result on the glucose percentage. Observations were made until the 7th day of fermentation. The average glucose levels tend to decrease with the length of fermentation time. Almost all treatments showed a significant decrease in fermentation time. This is in line with previous research which states that glucose levels will decrease with increasing fermentation time in pineapple skin samples [8]. This situation occurs because glucose has been converted into bioethanol. The length of hydrolysis time resulted in a decrease in the amount of glucose formed, the optimum time to hydrolyze starch to sugar is around 2 hours [9]. These changes occur due to the activity of *Saccharomyces cerevisiae* from the inversion enzyme [11,22]. The activity of microorganisms in the manufacture of bioethanol were very helpful in the catalysis process [12]. Only treatment E and F experienced an increase in glucose levels, although only slightly increased. Glucose levels will affect the levels of bioethanol produced. There were supported by previous research which states that A linear relationship was found between the total sugar concentration of the pulp which can be used to estimate the potential ethanol yield of the raw matter by the same field analytical test used in the sugarcane industry [13].

3.2. Bioethanol levels in various treatments

Fermentation in bioethanol manufacture is closely related to bioethanol level in a sample. The study results showed significant results in fermentation time in various treatments. This effect, can be seen in Table 1.

The observation results of the 8 treatments showed that on the 7th day of fermentation produced the optimal bioethanol content. After passing the 7th day of fermentation, the bioethanol content was decreases. This is in accordance with previous research, that bioethanol levels after 7 days decreased due to loss of protease, cell lysis, and enzyme degradation [10]. When fermentation started, the alcohol content was 0% in all treatments with various glucose levels according to Figure 1. The E and F treatments showed the optimal bioethanol content compared to other treatments. The effect of saccharification and proper fermentation with the help of *Saccharomyces cerevisiae* would help produce high ethanol levels [16-17,23].

The effect of starter on the bioethanol formation on various fermentation time was very significant. The starter will help *Saccharomyces cerevisiae* in converting glucose into bioethanol. The addition of a suitable and appropriate starter volume will greatly affect the process of bioethanol formation in each treatment [9],[21]. The starter in the study had a composition of 10 grams of sample, the 100 ml of water and add 8 ml of starter was added to the sample. Therefore, the addition of the starter used was 0.08%. This was also in line with previous research that the maximum addition of starter volume
during the fermentation process was 5% of the fermentation volume [9]. Therefore, all treatment added with starter all increased the optimal bioethanol content compared to treatment without starter (Treatment A and B). In addition, the bioethanol content from cassava peels and onggok yields was higher than the bioethanol that only from onggok samples. So the tapioca flour waste, had potential to produce high bioethanol. There were supported states that cassava waste can be converted into sugar to produce ethanol [14-15].

Table 1. Percentage of alcohol content in various treatments during fermentation

| No. | Treatment | Days to-- | 1  | 3  | 5  | 7  | 9  | 11 | 14 |
|-----|-----------|-----------|----|----|----|----|----|----|----|
| 1   | A         |           | 0  | 15 | 17 | 22 | 22 | 12 | 8  |
| 2   | B         |           | 0  | 15 | 18 | 22 | 22 | 12 | 8  |
| 3   | C         |           | 0  | 26 | 32 | 38 | 38 | 22 | 20 |
| 4   | D         |           | 0  | 24 | 30 | 35 | 35 | 23 | 15 |
| 5   | E         |           | 0  | 30 | 40 | 55 | 55 | 30 | 25 |
| 6   | F         |           | 0  | 25 | 33 | 45 | 45 | 28 | 22 |
| 7   | G         |           | 0  | 27 | 35 | 41 | 41 | 28 | 23 |
| 8   | H         |           | 0  | 25 | 32 | 35 | 35 | 24 | 15 |

Furthermore, the relationship between bioethanol levels and glucose levels in each treatment can be seen as follows.

Figure 2. Percentage of bioethanol and glucose levels in various treatments

In the fermentation process, alcohol content and glucose levels will differ significantly. During fermentation, *Saccharomyces cerevisiae* will use its glucose as a source of nutrition to produce bioethanol [18-19]. *Saccharomyces cerevisiae* was used respectively for fermentation [24-25]. As a result, the glucose levels will decrease with the length of fermentation time, and the bioethanol levels will be higher. These results can be seen in Figures 1 and 2 showing that all treatments experienced a decrease in glucose levels and an increased in bioethanol content.

4. Conclusion
The glucose levels percentage after fermentation treatments tends to increase slightly than before fermentation. The low glucose levels percentage was inversely related to the high alcohol content.
percentage. The optimum fermentation time to obtain optimal ethanol content was on the 7th day. From various treatment variations, it was found that 92% ethanol content in the treatment of cassava and onggok peels with the addition of a starter on the first day of sample fermentation.

Treatment without using a starter obtained the lowest ethanol content. The starter helps microorganism activity in fermentation process to produce optimal ethanol levels.

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