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BIOETHANOL PRODUCTION FROM POTATO PEEL USING DILUTE ACID HYDROLYSIS AND FERMENTATION PROCESS

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ABSTRACT

This research focused on the production of bioethanol from potato peels using dilute acid hydrolysis and fermentation process. The potato peels flour was subjected to pretreatment, hydrolysis, fermentation and distillation processes to produce bioethanol. The effect of hydrolysis and fermentation process variables on the yield of reducing sugar and bioethanol was studied to determine the optimal values that will give their maximum yields. The bioethanol produced was characterized using FTIR to determine the functional groups present in the distillate. The results obtained showed that maximum glucose yield of 44.16±0.89% was obtained at optimum parameter values of 2% acid concentration, 116 ^{o}C temperature and 25 minutes hydrolysis time; while the maximum bioethanol yield of 57.21±0.90% was obtained at optimum factor values of 6% yeast concentration, pH of 5.5 at 35 ^{o}C and 3-day fermentation time. The result of the FTIR analysis identified bioethanol as the main compound produced with stretching vibrations of OH at 3339.7 cm^{-1}, C-H at 2972.4 cm^{-1} and C-O at 1043.7 cm^{-1} stretch respectively. The results obtained showed that potato peel can be harnessed as potential and sustainable feedstock for bioethanol production in Nigeria.

Keywords: Bioethanol, fermentation, hydrolysis, potato peel, pretreatment.

INTRODUCTION

The quest for energy resources globally has increased due to massive population growth and industrialization. Presently, the world's major energy resources are from fossil fuels such as coal, petroleum and natural gas. These fossil fuels are produced from non-renewable sources that can be exhausted with time [1].

The combustion of these fossil fuels have negative impact on the environment among which is the emission of green house gasses known majorly to cause climate change. Other harmful substances that are released during fossil fuel combustion are sulphur oxides (SOx), nitrogen oxides (NOx) and methane [2]. Fossil fuels have a lot of disadvantages such as being non-biodegradable, non-renewable, environmentally unfriendly in nature and low octane number. This has led to the research into alternative and renewable sources of energy that can help combat these problems [3].

Bioethanol is a volatile, flammable, colourless biochemical liquid obtained by fermentation of sugar. It can also be chemically produced by reacting ethylene with steam [4, 5]. The equations are presented in equations 1 and 2.

$$C_6H_{12}O_6 \longrightarrow 2C_2H_5OH + 2CO_{2(g)} \quad \text{.....(1)}$$

$$C_2H_4(g) + H_2O_{(g)} \longrightarrow C_2H_5OH_{(g)} \quad \text{.....(2)}$$

Bioethanol is considered alternative to fossil fuel because of its high octane number, high heating value and high oxygen content [6]. It is also biodegradable, has lower CO_2 and dust emission because it is produced from renewable sources [5]. Bioethanol does not exhibit green house gas effect because it undergoes almost complete combustion when used in automobiles. This is due to its high oxygen content which results in low carbon monoxide emission when used as fuel additive to gasoline [3]. Other uses of bioethanol include as solvent in cosmetics, chemicals and preservatives.

In many European countries, the use of bioethanol serves as an alternative fuel to gasoline up to 15% [7]. The most common blend is 10% bioethanol and 90% petrol [8]. Vehicle engines require little or no modification to run on 10% bioethanol and 90% petrol and vehicle warranties are unaffected. When ethanol is blended with gasoline, the fuel mixture is oxygenated so that it burns more completely thereby reducing the emission of the unburned gases that causes environmental pollution [9].

Brazil and USA are the major producers of bioethanol. They have produced 16.12 billion liters of ethanol since 2005 [10]. These countries either use bioethanol fuel directly or as ethanol blended with fossil fuel. Bioethanol is yet to be commercialized especially in developing countries like Nigeria even though it has been extensively acknowledged to be a substitute to

petroleum product. This can be attributed to the production of bioethanol from edible feedstock such as starch and sugar crops (as maize, beet, sugarcane, corn and barley etc) which has led to their increase in price. This has created competition between food and fuel and the resultant increase in the price of bioethanol in the global market [11]. These problems can be resolved by the use of non-edible feedstock such as lignocellulose substrates like agricultural wastes, forest wastes and municipal wastes for bioethanol production [12].

The conversion of agricultural wastes to bioethanol involves pretreatment, hydrolysis, fermentation and distillation. The pretreatment step breaks the structure of the lignocellulose for easy access to cellulose, hemicellulose and lignin components. Hydrolysis step then converts the cellulose and hemicellulose to fermentable sugars like glucose while fermentation converts the hydrolyzed sugar to ethanol using microorganism like yeast [13,14]. In addition to the lignin, cellulose and hemicellulose, small amount of other materials such as ash, proteins and pectin can be found in these agricultural wastes in different degrees based on their sources [15]. Yeast strains such as *Sacchromyces cerevisiae*, and *Aspergillus niger* are generally used for the production of bioethanol and with the help of technology, bioethanol has become viable and feasible [16].

The agricultural industries generate a lot of solid wastes that includes peels from potato, banana, cassava, yam, sugarcane bagasse and orange as well as straws from cereals. Their disposal has become an environmental concern especially when they are not properly handled [17]. This has led to a new policy of waste to wealth which involves the complete utilization of raw waste materials so that little or no waste will be left that could pose pollution problem [18]. It is now realized that these wastes may be utilized as cheap raw materials for some industries or as cheap substitute for microbiological processes [19]. Studies have been conducted on the production of bioethanol from maize wastes and discarded Newspapers [20], sugar cane waste and maize waste [21], apple, Kiwi fruit, Peach wastes [22], mixture of cassava and potato peels [8] among others.

Potato peel is an underutilized waste with high carbohydrate content that requires simple pretreatment and hydrolysis processes [23,24]. Large amount of it is generated from food production which makes it suitable feedstock for ethanol production [25]. These wastes can be used as a carbon source for yeast during fermentation to produce bioethanol but they are mostly sold out cheaply to farmers to feed pigs while some are left to decay instead of being used to

produce useful products [9]. Enzymatic hydrolysis is mostly used but due to its high cost the research interest is focused on using dilute acid hydrolysis to determine its effect on the reducing sugar yield.

The main process variables which affect the hydrolysis and fermentation processes are concentration of acid, temperature, time, yeast concentration and pH. There is limited research on the optimization of these variables for maximum reducing sugar and bioethanol yield respectively. This study is aimed at producing bioethanol from potato peels using dilute acid hydrolysis and fermentation processes. Its use will contribute greatly in the conversion of the wastes into wealth thereby enhancing waste management, cost efficiency and environmental sanitation. [18].

MATERIALS AND METHODS

Sample Collection

Fresh potato peels were obtained after peeling potato tubers and were collected with plastic bags. The sample was transported to the Chemistry Laboratory, Akanu Ibiam Federal Polytechnic, Unwana, Afikpo, Nigeria, for further analysis.

Sample Preparation

The potato peels were cut into smaller sizes (between 3 to 4 cm) using a knife after it was rinsed with distilled water. The peels were dried in an oven to produce easily crushable materials for 72 hours at 60 ^{o}C and ground into fine powder using an electric grinding machine. The powdered sample was then sieved to a particle size of 1 mm, stored in sealed plastic containers and kept at room temperature until the next stage of analysis [18].

Sample Pretreatment

The sample was pretreated using steam in order to reduce the crystalline nature of the cellulose and promote the porosity of the material for easy hydrolysis. The steam pretreatment method was carried out according to the method described by Wondale [26]. Batch analysis using 50 g each of the powdered sample was carried out by dissolving the sample in distilled water at a ratio of 10:1 (v/w). The mixture was transferred into a 1000 ml conical flask, covered with aluminum foil and autoclaved. The autoclave's temperature was adjusted to a temperature of 121 ^{o}C and the pre treatment carried out for 15 minutes. The sample was allowed to cool after the pre

treatment and the soluble component separated from the non soluble component and stored for further analysis. The non-soluble component was used for acid hydrolysis.

Hydrolysis

Dilute acid hydrolysis using H_2S0_4 was carried out on the non soluble fraction of the sample to break down the cellulose and hemicellulose polymers into fermentable sugars that will be used to produce bioethanol [27]. The pretreated sample was used for the hydrolysis at different factor conditions of Acid concentration (1.0-3.0%), Temperature (100 – 132 ^oC) and Time (10 – 30 minutes) to determine the optimal factor conditions that will give maximum yield of glucose. After hydrolysis, in order to eliminate the non fermentable lignin, the solid particles were separated from the sugar rich liquid by filtration and added to the previously filtered solution from pretreatment process. The liquid portion was boiled for sometime so as to obtain the original sugar concentration and the solution stored at room temperature for further analysis.

Fermentation Process

Fermentation was carried out under anaerobic condition according to the method described by Wondale [26]. The fermentation media was first prepared before the fermentation process to enable a conducive environment for yeast growth and to supply the required amount of nutrients [28]. The fermentation media was prepared using dextrose sugar (10 g), urea (1.0 g), yeast extract (0.2 g), $MgSO_4$. $7H_2O$ (1.0 g) and distilled water to make up 100 ml. The sample was then mixed with the fermentation media in the ratio of 1:10 and added into the reactor using separating funnel. The conical flask was properly covered to maintain anaerobic condition with an outlet provided for the release of CO_2 as it turns lime water milky. Triplicate fermentation broths of the same composition were prepared and incubated under the same condition. The fermentation process was carried out initially at varying yeast concentration of 2 – 10%, temperature of 30 ^oC for 72 hours at pH 5.5. The pH of the fermentation was adjusted using pH meter to accommodate yeast growth by adding the required amount of 4 M NaOH and 2.5 M HCl [29]. Otherwise, the yeast will die in hyper acidic or basic state [18]. Subsequent fermentation process was carried out at various conditions of pH (4.0 – 6.0), temperature (20 - 40 ^oC), time (1 – 5 days) to determine the optimum factor conditions that will give maximum yield of bioethanol. After fermentation, the sample were taken out and distilled.

Filtration and Distillation of Bioethanol Produced

The sample was filtered using muslin cloth to separate the solid substrate from the liquid. The resulting mixture of bioethanol, water and other impurities were then transferred into the distillation flask and placed on a heating mantle fixed to a distillation column enclosed in a tap running water in order to obtain pure bioethanol. The distillate was collected at 78.5 ^{o}C (boiling point of ethanol) for 3 hours with another flask fixed at the other end of the column [18]. Distillation is the method used to separate two liquids based on their difference in boiling points.

Quantitative Estimation of Bioethanol Produced

The amount of bioethanol produced was estimated quantitatively using specific gravity method as described by Aleme *et al*. [30]. A 25 ml pycnometer (specific gravity bottle) was cleaned and dried first and weighed and the weight noted as Wo at 20 ^{o}C. The bottle was filled with bioethanol and reweighed at 20 ^{o}C to give W_1. The bioethanol sample was substituted with water after washing and drying the bottle and weighed at 20 ^{o}C to give W_2. Using these observations, specific gravity was calculated and the percentage of bioethanol in the distillate was estimated from the relationship between the specific gravity and the proportion of the ethanol in alcohol solution using AOAC table. The specific gravity of the sample was obtained using the following equation:

$$\text{Specific gravity} = \frac{W_1 - W_0}{W_2 - W_0}$$

Where W_0 = Weight (g) of the empty bottle

W_1 = Weight (g) of bottle + Sample (Bioethanol)

W_2 = Weight (g) of bottle + water

Optimization of Hydrolysis Process

The effect of the variables on the yield of glucose produced from potato peels was studied to determine the optimal factor conditions that gave the maximum yield. The hydrolysis process parameters studied include acid concentration, temperature and time. The results are presented as mean ± standard deviation at n = 3 for $P < 0.05$. The acid concentration was varied from 1.0 to 3.0% at a step increase of 0.5% while the other parameters were kept constant in order to determine the optimal acid concentration that will give the maximum glucose yield.

The effect of hydrolysis temperature on the yield of glucose from potato peels was determined by varying the temperature from 100 – 132 ^{o}C at step increment of 8 ^{o}C while keeping other parameters constant. The hydrolysis time was varied from 10 to 30 minutes with step increase of 5 minutes and other parameter kept constant in order to determine its effect on glucose yield.

Optimization of Fermentation Process

The effect of fermentation process variable on the yield of bioethanol production from potato peel hydrolysate was investigated in order to determine the optimum factor conditions that gave the maximum bioethanol yield. The fermentation process variables studied were: yeast concentration, pH, temperature and time. The results are presented as mean ± standard deviation at n = 3 for $P < 0.05$. The concentration of yeast was varied from 2 to 10% at a step increment of 2% in order to determine its effect on the bioethanol yield while keeping the other parameters constant. To determine the effect of pH on the yield of bioethanol the pH was varied from 4.0 to 6.0 at a step increment of 0.5 while keeping other parameters constant. The effect of fermentation temperature on the yield of bioethanol was studied by varying the temperature from 20 – 40 ^{o}C at a step increment of 5 ^{o}C while keeping other parameter constant. The fermentation time was varied from 1 to 5 days with step increase of 1 day while keeping the other parameters constant.

RESULTS AND DISCUSSION

Optimization of Acid Concentration

Table 1 shows the results of the glucose yield at different acid concentrations. From the results, increase in acid concentration increased the glucose yield till it reached the maximum value of 35.68±0.87% at optimum acid concentration of 2% when it started decreasing with further increase in the concentration of acid. The decreased glucose yield with high acid concentration results from the fact that low acid concentration is conducive for glucose production during hydrolysis. However, the use of high acid concentration for hydrolysis results to browning and charring of the hydrolysates as a result of the degradation of monomeric sugars (xylose, glucose) to fufural and 5 HMF which leads to decrease in glucose yield [3, 29, 31].

Optimization of Temperature

The result of the effect of temperature on reducing sugar yield is presented in Table 2 and it was observed that glucose yield increased with increase in hydrolysis temperature till it attained a maximum value of 41.04±1.00% at optimum hydrolysis temperature of 116 ^{o}C when it started decreasing with further increase in temperature. The results indicated that extreme temperature has unfavorable effect on the conversion of sugar from the substrates due to the degradation of the simple sugar which result in the formation of fufural and 5 HMF that are toxic for *Saccharomyces cerevisiae* in fermentation [31].

Optimization of Hydrolysis Time

The result of the effect of time on the reducing sugar yield presented in Table 3 shows that increase in hydrolysis time increased the yield of glucose till it reached the maximum value of 44.16 ± 0.89% at optimum time of 25 minutes. On exceeding the optimum time, the glucose yield reduced with further increase in time. The decrease in glucose yield on exceeding the optimum time is attributed to the decomposition of glucose to degradation product (Fufural and 5 HMF). Therefore maximum yield of glucose was obtained at optimum 2% acid concentration, 116 ^{o}C temperature and 25 minutes hydrolysis time.

Table 1: Glucose Yield from Hydrolysis of Potato Peels at Different Concentration of Acid

Acid Concentration (%)	Glucose yield (%)
1.0	28.06± 0.96
1.5	31.86 ±0.77
2.0	35.68 ± 0.87
2.5	34.40 ± 0.75
3.0	33.48 ± 0.85

Table 2: Glucose Yield from Hydrolysis of Potato Peels at Different Temperature

Temperature (°C)	Glucose Yield %
100	30.62 ±0.75
108	38.07± 0.98
116	41.04± 1.00
124	40.33 ±0.78
132	37.35 ±0.79

Table 3: Glucose yield from hydrolysis of Potato peels at Different Time

Time (Minutes)	Glucose yield (%)
10	28.06± 0.97
15	34.38± 0.76
20	40.33 ±0.77
25	44.16± 0.89
30	32.48 0.86

Optimization of Fermentation Yeast

The result of the effect of yeast concentration on the bioethanol yield presented in Table 4 reveals that the yield of bioethanol is increased with increase in yeast concentration till it reached a maximum value of 33.08 ± 1.06% at an optimum yeast concentration of 6%. On exceeding 6% optimum concentration of yeast, the bioethanol yield decreased. This may be due to the fact that above the optimum yeast concentration, the cells grow rapidly resulting in rapid consumption of the glucose to produce bioethanol at a reduced fermentation time [3].

Optimization of pH

The result for the effect of pH on the bioethanol yield is presented in Table 5 which revealed that bioethanol yield increased with increase in pH till it attained the highest value of 37.45 ± .0.91% at an optimum pH of 5.5 when it started to decrease as the pH increased further. The optimum pH for bioethanol production using *saccharomyces cerevisiae* has been reported to be 4.0 to 5.5. On exceeding pH of 5.5, the yeast may be denatured which will lead to the reduction of the catalytic activities and subsequent decrease in the bioethanol production [3].

Optimization of Fermentation Temperature

The result of the effect of temperature on the bioethanol yield shown in Table 6 indicated that bioethanol yield increased with increase in fermentation temperature until it attained the maximum value of 47.28 ±1.05% at optimum temperature of 35 ^{o}C. On exceeding the optimum temperature, the bioethanol yield decreased. One of the characteristics of microorganism is that temperature has a significant effect on them over narrow range [3]. According to Egbosiuba *et al* [11], fermentation process above 45 ^{o}C could lead to the death yeast of cells which enhances reduction in their activity thereby decreasing the yield of bioethanol produced while lower temperature slows them down. The results is similar to that obtained by Duhan *et al* [32] who studied the effect of temperature on bioethanol yield and obtained maximum yield at 35 ^{o}C.

Optimization of Fermentation Time

The result obtained is presented in Table 7 which revealed that bioethanol yield increased with increase in fermentation time until it attained the highest value of 57.21 ± 0.90% at an optimum time of 3 days. On exceeding the optimum fermentation time, the bioethanol yield started decreasing. The optimum time of 3 days is similar to the result obtained by Phisalaphong *et al* [33].

Result of FTIR Characterization of the potato bioethanol

FTIR analysis was used to identify the functional groups present in the bioethanol sample. The FTIR spectra (Figure 1) obtained in this study showed the presence of broad absorption peak between 3300-3500 cm^{-1} which indicates the presence of OH stretching in alcohol [34]. The peaks at 2972.4 and 1043.7 cm^{-1} are attributed to C-H and C-O stretching in ethanol [35]. This further confirms that bioethanol could be produced from potato peels.

Table 4: Bioethanol Yield Potato Peel Hydrolysate at Different Concentration of Yeast

Yeast extract %	Bioethanol yield %
2	30.38 ±0.98
4	31.23 ±0.99
6	33.08 ±1.06
8	33.51 ±0.74
10	31.35 ±0.75

Table 5: Bioethanol Yield from Potato Peel Hydrolysate at Different pH

pH	Bioethanol yield
4.0	33.20±0.96
4.5	35.22 ±0.92
5.0	36.47 ±0.76
5.5	37.45 ±0.91
6.0	32.76 ±0.80

Table 6: Bioethanol Yield from Potato Peel Hydrolysate at Different Temperatures

Temperature (^oC)	Bioethanol yield %
20	41.71 ± 0.74
25	43.22 ± 1.06
30	46.22 ± 0.99
35	47.28 ± 1.05
40	46.07 ± 0.97

Table 7: Bioethanol Yield from Potato Peel Hydrolysate at Different Fermentation Time

Time (days)	Bioethanol Yield
1	49.47±0.77
2	47.52±1.24

3	57.21±0.90
4	56.24±0.95
5	47.30±0.85

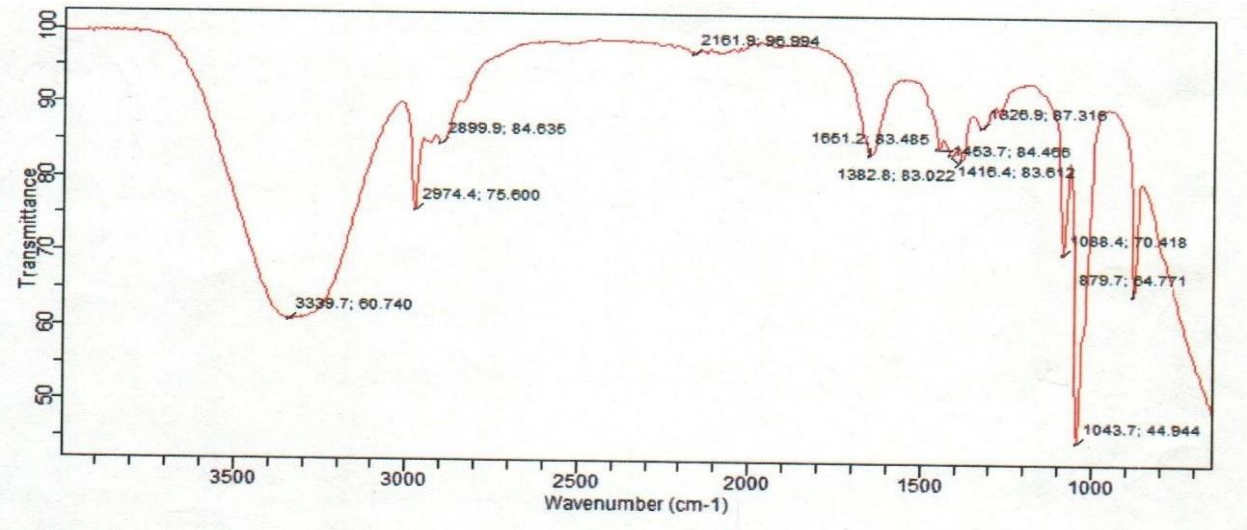

Figure 1: FTIR Spectrum for Potato Peels Bioethanol

CONCLUSION

This study focused on the pretreatment and hydrolysis of potato peels to produce glucose and its subsequent fermentation to bioethanol. The results for hydrolysis and fermentation process indicated that acid concentration, temperature, and time of hydrolysis affect the optimum yield of glucose significantly as the yield of 44.16± 0.89% was obtained at optimum factor condition of 2.0% acid concentration temperature of 116 ^{o}C an time of 25 minutes while the maximum bioethanol yield of 57.12±0.90% was obtained at optimal factor condition of 6% yeast concentration, pH of 5.5 at 35 ^{o}C and 3-day fermentation time. Also the FTIR analysis of the distillate produced showed the presence of OH group which confirms that bioethanol was produced from potato peels. This demonstrates that potato peels have high amount of glucose which accounted for 50% of the bioethanol produced. Hence potato peel is a renewable source and abundant agricultural waste that can serve as rich source of sugar which can be converted into bioethanol through hydrolysis and fermentation process.

RECOMMENDATION

Potato peels are rich in carbohydrate content and are in abundance in Nigeria which makes bioethanol production from them to have tremendous potential in terms of meeting the energy needs and promoting environmental benefits. It is recommended that the use of these wastes as alternative sources of producing bioethanol should be encouraged to help alleviate the problem of waste disposal and environmental pollution thereby boosting the economy of Nigeria.

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