PRODUCTION, CHARACTERIZATION AND KINETIC MODELING OF BIOETHANOL PRODUCTION FROM CASSAVA TUBERS

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ABSTRACT

The rapid depletion of the world major energy source, the fossil fuel coupled with the numerous problems associated with its use has resulted in the search for renewable, sustainable and environmentally friendly fuel. Among the various options investigated, bio-ethanol ranks among the foremost alternatives to fossil fuel. This research work focused on production, characterization and kinetic modeling of bio-ethanol production from cassava tubers. Bio-ethanol was produced by enzyme hydrolysis of cassava pulp followed by simultaneous sacharification and fermentation of the glucose produced thereof using yeast. The fermentation process was optimized using one factor at a time method in other to determine the influence of fermentation parameters on the yield of bio-ethanol. The physiochemical properties of the bio-ethanol produced were determined based on American Standards for Testing and Materials (ASTM) method. The kinetics of bio-ethanol production was modeled based on Michaelis-Menten equation. The optimum values of fermentation parameters that gave the highest yield of bio-ethanol are, fermentation temperature 35^{O}C, fermentation time 6 days, yeast dosage 10g/L, glucose concentration 100g/L, pH 4.5 and agitation rate 160 rpm. The physiochemical properties of the bio-ethanol produce were determined as, kinematic viscosity 1.54cst at 40^{O}C, density 779 Kg/m^3, flash point 13.5^{O}C, refractive index 1.364, octane number 102, sulphur content 0.06%, water content 0.05 ppm boiling point 78^{O}C, lower heating value 20.8 KJ/dm^3, pH 6.6. The data of variation of bio-ethanol concentration with fermentation time was adequately modeled by Michalis-Menteen equation, hence the plot of $1/r_A$ versus $1/C_A$, gave a straight line graph of slope km/Vm = 0.795 and the intercept 1/Vm = 0.048. Insertion of the values into Michaelis-Menten equation gave the rate of fermentation reaction, r_A.

Keywords: Bio-ethanol, Fermentation, Hydrolysis, Kinetic modeling, Saccharification

1.0 Introduction

Various countries have made inroad into exploitation of energy from different sources including solar, wind, hydro, thermal, nuclear in addition to the worlds' main energy source, fossil that has not been rivaled in terms of quantity of reserves and suitability as fuel. However, the fast depletion of fossil fuel, coal, petroleum and natural gas and their attendant disadvantages, non-renewable, non-biodegradable, environmentally unfriendly, low lubricity, low octane number among others have made the search for alternative source of energy to fill the gap inevitable. As a result of the aforementioned problems associated with fossil fuel, bio-fuels namely bio-ethanol, biodiesel and biogas, have received much attention [1-3] and are envisaged to solve these problems. In contrast to fossil fuel, bio-ethanol is a renewable, biodegradable, environmentally friendly, of high octane number, high heating value, low emission of poisonous gases as a result of the high oxygen content that encourage complete combustion of the fuel [4, 5]. Bio-ethanol is a biochemical liquid derived from microorganism catalyzed fermentation of sugar in addition to distillation [6, 7]. However, bio-ethanol could be prepared chemically by the reaction of ethylene with steam [6]. It is a safer substitute for methyl tetra-butyl ester (MTBE}, a common additive used in gasoline for clean combustion.

Bio-ethanol is produced by fermentation of sugar catalyzed by microorganisms followed by distillation to obtain its pure and concentrated form [8, 6, 7, 9]. Apart from sugar, some other plant material consisting of carbohydrate like starch and lignocellulosic biomass could also be converted to bio-ethanol. Sugar, starch and lingocellulocic biomass are called first, second and third generation feed stocks respectively for production of bio-ethanol. The bio-ethanol produced from first, second and third generation feed stocks are known as first, second and third generation bio-ethanol respectively. While the first and second generation feed stocks are feared may compete with food sources, the lingocellulocic feed stock does not compete with food sources but rather its processing to bio-ethanol is more complicated as it involve four processing steps and therefore requiring more equipments and materials and higher cost. The four steps involved with conversion of lignocellulosic biomass to bio-ethanol are; pretreatment with acid, hydrolysis of the pretreated biomass, fermentation of the hydrolysate and distillation of the supernatant to concentrated bio-ethanol. Pretreatment is the most involving aspect of the processing as it entails breaking of its recalcitrant structure for easy access to the cellulose, hemicelluloses and lignin components by acid hydrolysis which converts the cellulose and hemicelluloses to fermentable sugar like glucose. Starch biomass is first hydrolyzed with acid or enzyme to obtain fermentable sugar. The fermentable sugar derived from lignocellulosics are then converted to bio-ethanol by fermentation with microorganisms like yeast. It is easier and more cost effective to use free sugar containing juice as feed stock for bio-ethanol production than starch or lignocellulosic biomass due to none requirement of costly steps such as hydrolysis or pretreatment followed by hydrolysis to get fermentable sugar [10-13]. Currently industrial bio-ethanol employ mainly two types of primary feed stocks, starch from cereal crops and juice or molasses from sugar crops [14, 15, 16]. About 60% of the global bio-ethanol is produced from sugar while the remaining 40% is produced from grains starch [17]. Bio-ethanol can also be produced from the free sugar containing juice of some crops containing sucrose or monosaccarides especially glucose, via fermentation with microorganism [18].

The feed stocks for bio-ethanol production are sugar, starch and lignocellulosic biomass. Sugar the feed stock for production of first generation bio-ethanol is principally sourced from energy crops like maize, sugar cane barley and wheat [19, 20]. Starch which is a cellulosic biomass used for preparation of second generation bio-ethanol is sourced from corn, wheat and cassava. The lignocelluloses materials including cassava peels, wheat straw, rice straw, corn straw, sugar cane bagasse are available in large quantity and are therefore suitable for large scale production of bio-ethanol [21, 22]. Similarly, starch biomass obtained as waste from starch processing factories, potato food industries, beverage and brewing industries are promising feed stock for large scale bio-ethanol production in tropical countries [23, 24].

Cassava (manihot esculentus is a tuberous root crop native to South America, and is cultivated in most parts of the world primarily for its starch as well as food source. Cassava can grow in marginal soils where other crops does not thrive [25]. Some researchers have shown that bio-ethanol production based on cassava is sustainable [26-28]. Therefore intensive cultivation of cassava coupled with biotechnological improvement of the species and in addition to harnessing the waste starch from the above mentioned industries will ensure sustainable commercial bio-ethanol production from cassava pulp and the peels as well as serving as food source for Nigeria that has much of the marginal soil lying fallow.

This research work is focused on production, characterization and kinetic modeling of bio-ethanol production from cassava pulp. Many researchers have reported on bio-ethanol production by fermentation of sugar using saccharomyces cerevisiae [29-34].

2.0 Materials And Methods

2.1 Materials

Cassava tubers, backers yeast (sacccharomyces cerevisiae), distillation unit, digital pH meter, magnetic hot plate, thermostatic water bath, glassware's, autoclave, viscometer, refractometer, flash point close cup tester.

2.2 Experimental Methods

2.2.1 Processing of the cassava tubers

The bulk of cassava tubers used in the research was peeled, washed and chopped into small pallets with knife. The pallets were sun-dried for three days and then dried in the oven at $60^{O}C$ for 72 hours to reduce the water content below 14%. The dried pulp was then size-reduced by grinding with a machine grinder and then sieved to obtain pulp flour of 150µm. The flour was bagged with polythene and kept for further experiments.

2.2.2 Production of bio-ethanol from cassava pulp flour

Production of bio-ethanol from cassava pulp flour which constitute principally of starch, a cellulose biomass involves hydrolysis, fermentation and distillation. Enzyme hydrolysis was used here to reduce the problems arising from acid hydrolysis. Again simultaneous saccharification and fermentation was used to avert high energy requirements of hydrolysis and saccharification coupled with enormous amount of water-based cooling system to bring down the temperature for fermentation 35-37].

2.2.3 Enzyme Hydrolysis of the cassava pulp flour

The pulverized cassava pulp flour was mixed with water in the ratio of 1:5 w/w. 30g of the pulp was mixed with 150ml of water in a 500ml round bottomed flask and gelatinized by heating on a water bath to a temperature of $80^{O}C$ until thick slurry was formed. The gelatinized pulp was liquefied by adding 1.3 ml α-amylase enzyme at $90^{O}C$ with continuous agitation of 160rpm for 40 minutes to form dextrin. The slurry was cooled to room temperature of $32^{O}C$ and then sterilized in an autoclave at $121^{O}C$ for 15 minutes.

2.2.4 Simultaneous Saccharification and fermentation of hydrolyzed pulp

The process of saccharification and fermentation were simultaneously carried out to avoid the problem associated with simultaneous liquefaction and saccharification with an enzyme. The sterilized and cooled hydrolysate was adjusted to a pH of 4.5 with citric acid. Then 0.35ml of glucoamylase enzyme solution and 10% mixture of backers yeast (saccharomyces ceravisiae] were inoculated onto the flask content. The flask and content was placed on a shaker in a water bath

2.2.5 Distillation

After fermentation, the residue was filtered and the filtrate containing bio-ethanol, water and other impurities like methanol was reflux-distilled at 80^{O}C. The bio-ethanol content at various intervals was analyzed with gas chromatogram (GC).

2.3 Optimization of fermentation process parameters

The cassava pulp fermentation process parameters, fermentation temperature, yeast concentration, glucose concentration, pH, fermentation time and agitation rate were optimized using classical method of optimization of studying the effect of one factor at a time while the other factors were kept constant. Here the temperature was varied from 15 to 45^{O}C, yeast concentration from 2 to 14g/L, glucose concentration from 15 to 135g/L, pH from 2.5 to 5.5, fermentation time from 1 to 7 days and agitation rate from 80 to 200rpm.

2.4 Modeling of kinetics of bio-ethanol production

The rate of the reaction A → product, is given by the expression,

$$r_A = kC^n_{A.} \qquad (1)$$

r_A is the rate of reaction of A, C_A is the concentration of the reactant A, k is the reaction rate constant and n is the order of reaction. The essence of modeling kinetics of reaction is to obtain the kinetic parameters r_A, k and n for designing the reaction experiment and the reactor in which the experiment is carried out.

3.0 Results and Discussion

3.1 Optimization of fermentation process parameters

The Influence of process parameters on the yield of bio-ethanol from fermentation of cassava pulp hydrolysate was studied in order to obtain the values of optimum parameters that gave the highest yield of bio-ethanol. The fermentation process parameters studied include, fermentation temperature, yeast concentration, glucose concentration, pH, fermentation time and agitation rate. The plots of the data of all the six parameters with ethanol concentration are given in figures 1 to 6. The data of all the plots are well fitted with quadratic model of coefficient of determination R^2 of approximately unity.

3.1.1 Effect of fermentation temperature on the yield of bio-ethanol from cassava pulp hydrolysate.

The influence of fermentation temperature on the yield of bio-ethanol from cassava pulp hydrolysate was studied by varying the fermentation temperature from 15 to 45^{O}C at a step increase of 5°C while keeping the other parameters , yeast concentration, glucose concentration, pH, fermentation time and agitation rate constant. The result of the experiment is as shown in Figure 1. From the Figure, it could be seen that the bio-ethanol yield increased with increase in fermentation temperature until it attained the maximum value of 17g/L at an optimum temperature of 35^{O}C. On exceeding the 35^{O}C optimum temperature, the bio-ethanol yield decreased with increase in temperature. This is because microorganisms like saccharomyces cerevisia are known to be stressed by high temperature which inactivate their ribosomes Again microbial activities and fermentation are regulated by different enzymes which are susceptible to deactivation by high temperature [38, 39].

3.1.2 Effect of yeast dosage (concentration) on the yield of bio-ethanol from cassava pulp hydrolysate

Yeast concentration was varied from 2 to 14g/L, at a step increase of 2g/L in order to determine its influence on the bio-ethanol yield while keeping the other parameters constant. The results obtained are shown in Figure 2. From the Figure it is clear that the bio-ethanol yield increased with increase in yeast concentration till it reached a maximum value of 17g/L at an optimum yeast concentration of 10g/L. On exceeding 10g/L optimum yeast concentration, the bio-ethanol yield remained constant with increase in yeast dosage. After exceeding the optimum yeast concentration, the inoculums size no longer had significant effect on bio-ethanol yield but rather affected the rate of sugar consumption. Above the optimum yeast concentration, the cells grow rapidly resulting in rapid consumption of the glucose to produce bio-ethanol at a reduced fermentation time.

3.3.3 Effect of glucose concentration on the yield of bio-ethanol from cassava pulp hydrolysate

The influence of glucose concentration on bio-ethanol yield was studied by varying the glucose concentration from 15 to 135g/L with step increase of 20g/L while keeping the other parameters constant. The results obtained for the experiment are shown in Figure 3. From the Figure it could be seen that bio-ethanol concentration increased with increase in glucose dosage till it attained the highest value of 17g/L at the optimum glucose concentration of 100g/L when the bio-ethanol yield started decreasing with increase in glucose concentration. Decrease in bio-ethanol production after exceeding the optimum glucose concentration stem from the fact that such high concentration of glucose overwhelmed the microorganism intake capacity, resulting in the reduction of the activity of the microorganism and hence the bio-ethanol production.

3.1.4 Effect of pH on the yield of bio-ethanol from cassava pulp hydrolysate

In order to determine the influence of pH on the bio-ethanol yield from cassava pulp hydrolysate, the pH was varied from 2.50 to 5.50 at a step increase of 0.5 while keeping the other parameters constant. The results obtained are shown in Figure 4. From the Figure it could be observed that bio-ethanol yield increased with increase in pH till it attained highest value of 17g/L at an optimum pH of 4.5 when it started to decrease with increase in pH value. Microorganisms have a certain range of pH at which its activity is optimal. It has been reported that pH of the broth is a key factor to be considered in bio-ethanol production as it influence the microorganism directly as well as its cellular processes. The optimum pH for bio-ethanol production using saccharomyces cerevisiae has been reported to be 4.0-5.0. On exceeding the pH of 4.50 saccharomyces cerevisiae is strained, denatured and the catalytic activities reduced, resulting to reduction of the bio-ethanol produced.

3.1.5 Effect of fermentation time on bio-ethanol yield from cassava pulp hydrolysate

The fermentation time was varied from 1 to 7 days with step increase of 1 day while keeping the other parameters constant in order to determine its influence on bio-ethanol yield. The results obtained are as shown in Figure 5. From the Figure it could be seen that the bio-ethanol yield increased with increase in fermentation time till it attained highest value of 17g/L at an optimum time of 6 days. On exceeding the optimum fermentation time of 6 days, the bio-ethanol yield started decreasing with increase in fermentation time. At the initial time of the first day, the bio-ethanol produced was low as the yeast was at the lag phase. As the time progresses, the microorganism enters its exponential period of growth when the bio-ethanol production increase exponentially as well. From the Figure, the optimal period for bio-ethanol production is 6days. On exceeding the optimum fermentation period of 6 days, the bio-ethanol produced started declining with increase in time as the death rate of the microorganism become greater than the birth rate.

3.1.6 Effect of agitation rate on bio-ethanol yield from cassava peel hydrolysate

The agitation rate was varied from 80 to 200rpm at a step increase of 20rpm while the other parameters were kept constant in order to study the effect of agitation rate on bio-ethanol yield. The result obtained is as shown in Figure 6. From the Figure it could be seen that bio-ethanol production increased with increase of agitation rate till it reached maximum concentration of 17g/L at an optimal agitation rate of 160 rpm. On exceeding the optimum agitation rate of 160 rpm, the bio-ethanol produced started decreasing. Moderate agitation rate is required for efficient fermentation of glucose to produce ethanol using saccharomyces cerevisiae. Agitation enables permeability of nutrients into the cells and removal of bio-ethanol from the interior of the cells to the fermentation broth. It also increases the level of sugar consumption by the yeast and reduced the inhibitory effect of bio-ethanol on the activities of the cells. Excess agitation is however counterproductive for bio-ethanol production as it lowers the metabolic activities of the cells resulting in lower bio-ethanol yield.

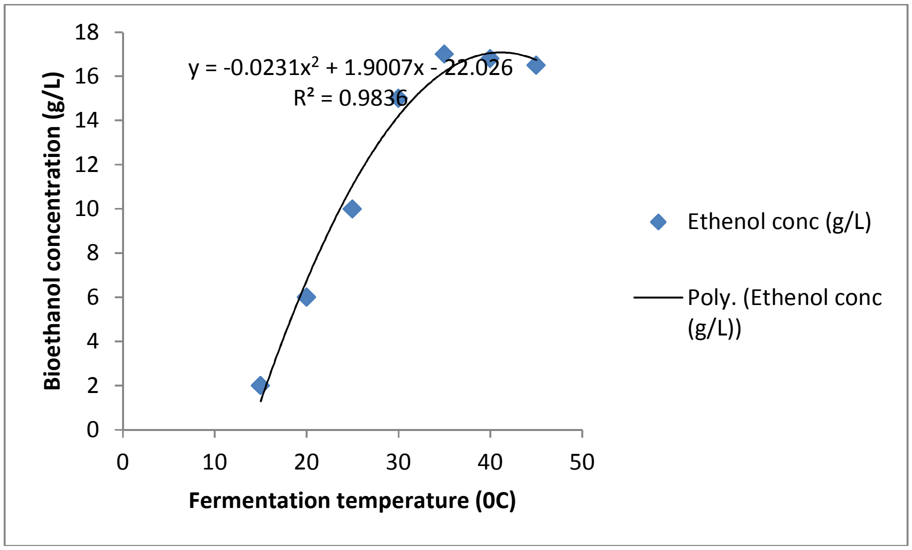

Figure 1: Effect of fermentation temperature on the yield of bio-ethanol from

cassava pulp hydrolysate

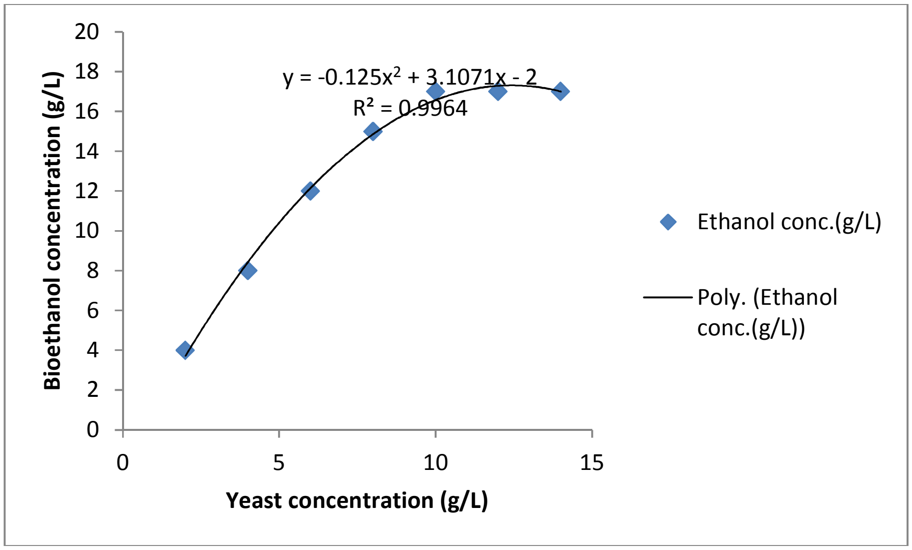

Figure 2: Effect of yeast dosage on the yield of bio-ethanol from cassava pulp hydrolysate

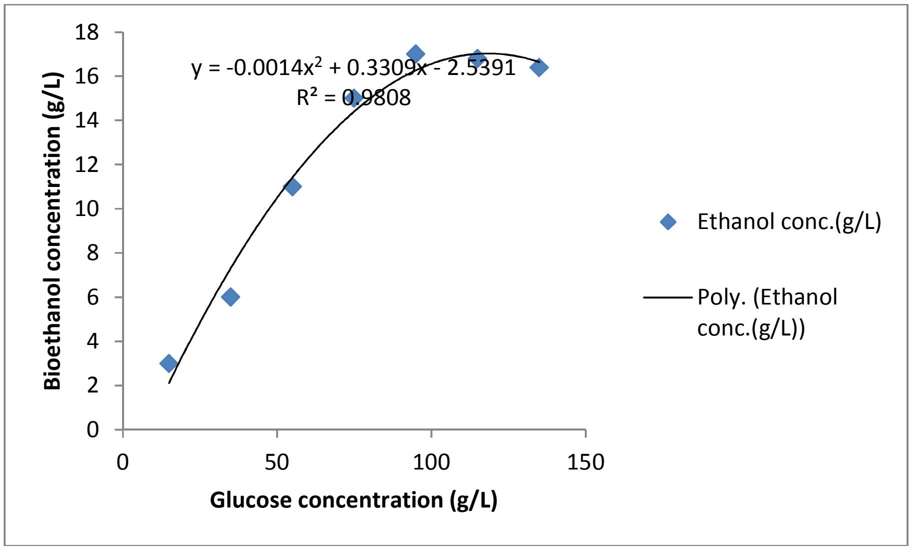

Figure 3: Effect of glucose concentration on the yield of bio-ethanol from cassava pulp hydrolysate

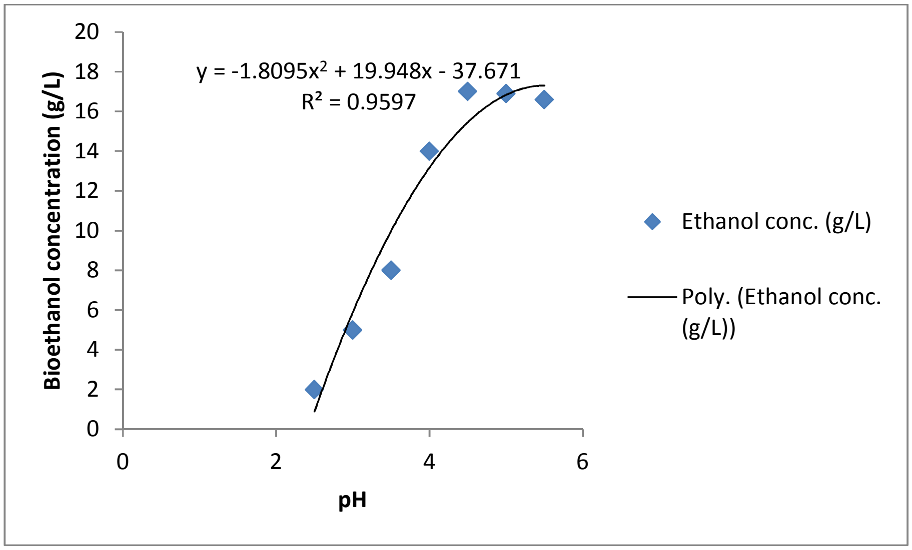

Figure 4: Effect of pH on the yield of bio-ethanol from cassava pulp hydrolysate

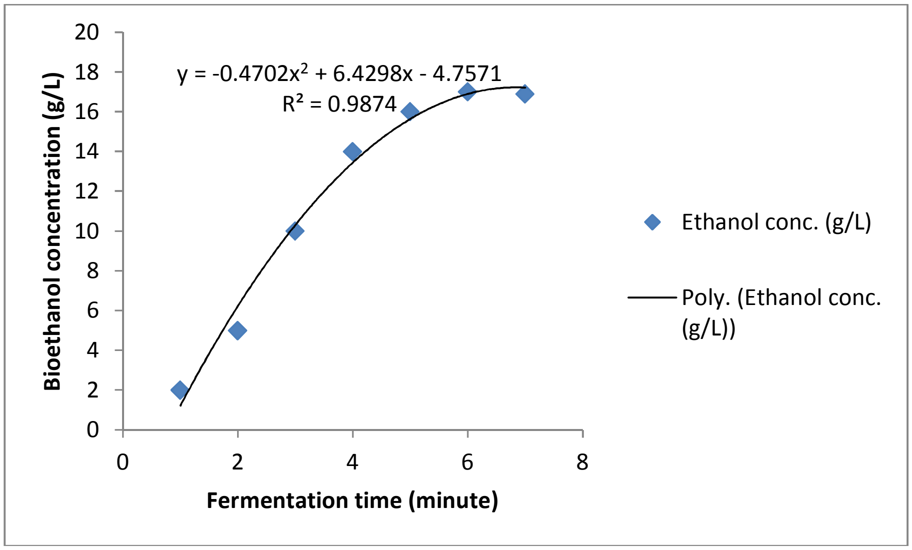

Figure 5: Effect of fermentation time on the yield of bio-ethanol from cassava pulp hydrolysate

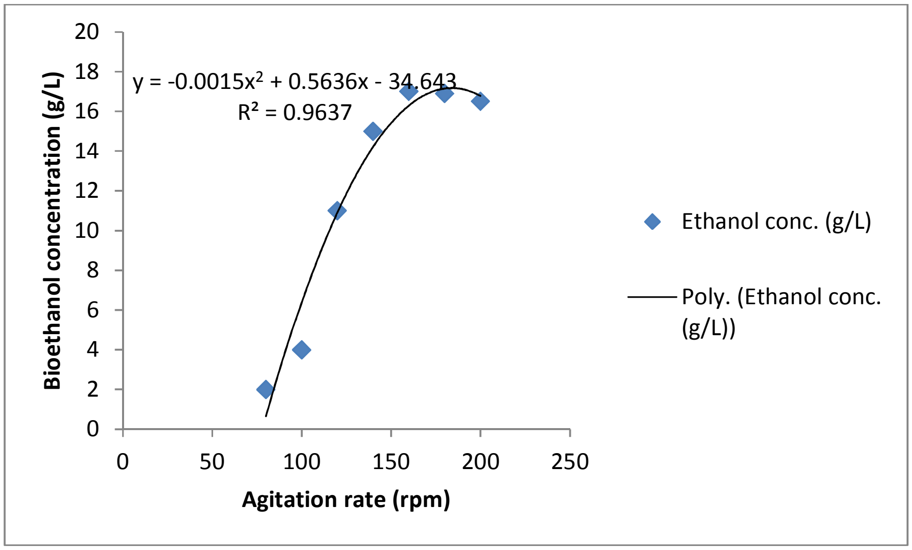

Figure 6: Effect of agitation rate on the yield of bio-ethanol from cassava pulp hydrolysate

3.2 Characteristics of produced cassava pulp bio-ethanol

The physiochemical properties of the produced bio-ethanol as compared to those of gasoline are presented in Table 1. Density the ratio of mass to volume of the fuel produced is 779kg/dm3., which is within the ASTM standards. The bio-ethanol density is greater than that of gasoline, therefore more volume of gasoline should give an equivalent mass of bio-ethanol, in addition of gasoline possessing better ignition quality.

Viscosity is another very important characteristic of fuel. The bio-fuel should neither be too viscous nor too thin. The kinematic viscosity of the bio-ethanol produced was evaluated as 1.54 cst at 40^{O}C and is therefore within the ASTM limit. High kinematic viscosity of bio-fuels result in poor atomization and incomplete combustion which give rise to cocking of injector tips and hence engine power loss. This conforms to the findings of Tat and Van Gerpen [41]. On the other hand very low viscosity fuel produces very subtle spray which cannot properly get into the combustion cylinder, thus forming a fuel rich zone that give rise to sooth formation [42-44]. The viscosity value obtained for the bio-ethanol conformed with the literature value of Muhaj and Sutjahio [45]. The viscosity of bio-ethanol is higher than that of gasoline. This is indicative of the fact that bio-ethanol is prone to less atomization efficiency, incomplete combustion and hence higher engine power loss than gasoline.

Flash point measures the degree of flammability of the fuel. This was evaluated as 13.5^{o}C for the bio-ethanol, and is therefore within the ASTM limit for bio-ethanol. The flash point of bio-ethanol is higher than that of gasoline and therefore exhibit less flammability than gasoline. The flash point of bio-ethanol 13^{O}C is higher than that of gasoline and therefore less dangerous to handle and store. The octane number of fuel expresses the degree of knocking of such fuel in internal combustion engine. Fuels of high octane number do not knock in the engine. The degree of knocking in the engine is a function of the ratio of branched chain molecules like 2,2,4-trimethylpentane to the straight chain molecules like n-pentane contained in the fuel.

Table 1 : Physiochemical properties of produced cassava peel bio-ethanol

Properties	Produced Bio-ethanol	ASTM Bio-ethanol Standards	ASTM Gasoline Standards	Test Method
Kinematic viscosity @ 40^{0}C (cst)	1.54	1.525	1.00	D445
Density (Kg/m^{3})	779	750-850	690-790	D1293
Flash point (^{0}C)	13.5	10-15	-43	D93
Refractive index	1.362	1.360-1.364	1.350-1.355	
Octane number	102	95-110	88-100	
Sulphur content (%)	0.06	0.05		D3452

Water content (ppm)	0.05	0.05	0.05	D203
Boiling point (^{0}C)	78.6	78.5	27-225	
Lower heating value (KJ/dm^3)	20.4	22	30.33	D240
pH	6.7	6.5-9.0		D684
Acidity (As acetic acid)	0.006	0.007max.		D1613
Solubility in water	Soluble	Soluble	Insoluble	

A fuel rated 88% octane number shows that it contained 88% of the branched molecules and 12% of the straight chain molecules. The octane number of the bio-ethanol produce is 102, which is within the ASTM standard limit, indicative of its smooth burning in the engine cylinder. The octane number of gasoline is less than that of bio-ethanol and therefore is prone to more knocking in internal combustion engine. The water content of the produced bio-ethanol was evaluated as 0.05 which is within the standard limit of ASTM. High water content in bio-ethanol predisposes the oxidation of the fuel as well as being the cause of corrosion in storage tank when stored for a long time. The infinitesimally small water content of bio-ethanol which is equivalent to that of the gasoline is within the ASTM standards. The water content of the bio- ethanol therefore has little or no effect on the degradation of the fuel or on corrosion in tanks during storage. The boiling point of the bio-ethanol was evaluated as 78^{O}C which corresponds with the ASTM standards. Refractive index indicates the state of purity of a substance as any substance has more or less a specific refractive index. The refractive index of the bio-ethanol produce is 1.362 and is within the ASTM standard limit.

3.3 Modeling of kinetics of bio-ethanol production

The rate of the reaction A → product, according to [43, 44] is given by the expression,

$$r_A = kC^n_A. \qquad (1)$$

where r_A is the rate of consumption or production of A. r_A is negative if A is reactant or positive if A is product. A is the reacting component, C_A is the concentration of A (gL^{-1}), k is the reaction rate constant ($gL^{-1}d^{-1}$) and n is the order of reaction, d^- is day. The essence of modeling kinetics of reaction is to obtain the kinetics parameters r_A, k and n for designing the reaction experiment and the reactor in which the experiment is carried out. The product ethanol is denoted by A and its concentration by C_A. The data of ethanol concentration with fermentation time is given in figure 5. The differential of bio-ethanol concentration C_A with respect to fermentation time t, dC_A/dt or r_A at designated C_A's were evaluated based on the graphical differential method enunciated by [45] . The rate of reaction r_A is then given by

$$r_A = dC_A/dt = kC^n_A \qquad (2)$$

Michaelis-Menten equation (3) was adopted for modeling of the bio-ethanol production.

$$V = Vm \frac{S}{Km + S} \qquad (3)$$

Where V is the velocity of the reaction, Vm is the maximum attainable velocity of the reaction, Km is Michaeli-menten constant and S is the concentration of the reactant. By denoting Vm with r_{Am} and with the notations in equation (2), equation (3) could be written as equation (4).

$$r_A = r_{Am} \frac{C_A}{km + C_A} \qquad (4)$$

The reciprocals of equations (3) and (4) respectively are equations (5) and (6),

$$\frac{1}{V} = \frac{km\ 1}{Vm\ S} + \frac{1}{Vm} \qquad (5)$$

$$\frac{1}{r_A} = \frac{km\ 1}{r_{Am}\ C_A} + \frac{1}{r_{Am}} \qquad (6)$$

The plot of $\frac{1}{r_A}$ versus $\frac{1}{C_A}$ of equation (6) known as line weaver Burk reciprocal plot is given in figure 7. This is a straight line graph of slope $\frac{Km}{r_{Am}}$ and slope $\frac{1}{r_{Am}}$ a condition for Michaelis-Menteen equation to have satisfactorily fitted the bio-ethanol concentration versus fermentation time data.

The graphical method of differenciation by [43] was employed to evaluate the differential of bio-ethanol concentration with fermentation time, $\frac{dC_A}{dt}$ (or r_A) at specified concentration C_A.

Here r_A, r_{Am} and C_A of equation (6) are the same as V, Vm and S of equation (5) respectively.
The plot of $\frac{1}{r_A}$ versus $\frac{1}{C_A}$ of equation (6) gave a straight line graph with coefficient of determination R^2=0.983 indicative of the adequacy of straight line fit of the data. The straight line has the intercept of 0.048 and slope 0.795. Inserting the values into the expressions above give r_{Am} = Vm = 20.83g/Ld, km = 16.56L/gd. Therefore the rate of fermentation r_A is give by equation (7).

$$r_A = \frac{20.83CA}{16.56 + C_A} \frac{g}{Ld} \qquad (7)$$

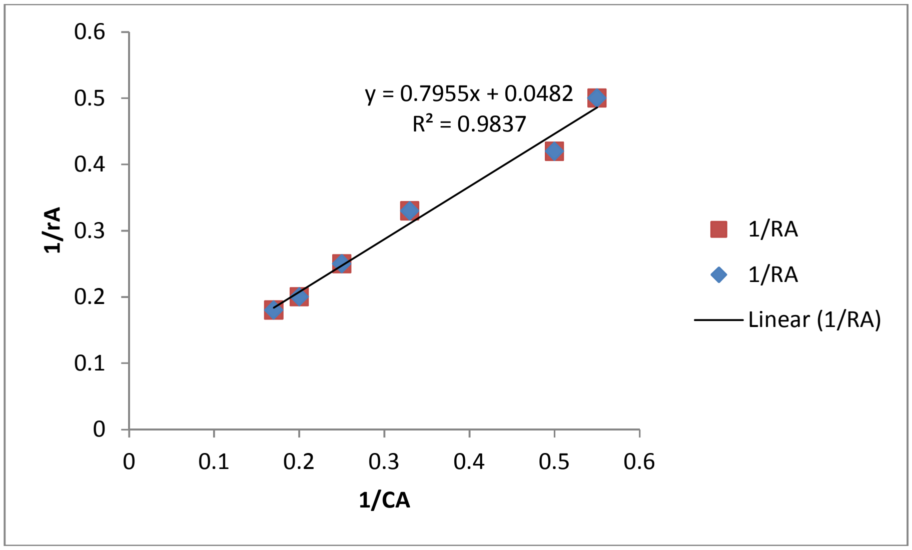

Figure 7: Plot of $1/r_A$ versus $1/C_A$ for bio-ethanol production by fermentation

CONCLUSION

Production of bio-ethanol from cassava pulp hydrolysate yielded bio-ethanol of physiological properties within the ASTM standards. The use of enzyme hydrolysis with simultaneous saccharification and fermentation method for bio-ethanol production reduced the problem associated with acid hydrolysis and saccharification of high energy and cooling water requirement The process parameters, fermentation temperature, fermentation time, yeast concentration, glucose concentration, pH and agitation rate have varying significant impact on the yield of bio-ethanol.

REFERENCES

1 Ahorsu, R., Medima, F., & Clement M.(2018). Significance and challenges of biomass as a suitable feedstock for bioenergy and biochemical production: A review, Energies 11(3366),2018, 1-19.
2 Abdeshahian, P., Dashti, M.G., Kalil, M.S., Yusoff. W.M.W. (2010). Production of biofuel using biomass as a sustainable biological resourse. Biotechnology. 9, 274-282.
3 Vimond, R., Medina, F. & Constant, M. (2013).Valorization of agricultural solid residue and residues from biofuel production chains by thermochemical conversion. A review citing Brazil as a case study. Brazilian Journal of Chemical Engineering Vol. 30, 197-229.
4 Tan, J.H., Sigaud, J., Claros Baptista, A. & Ozza, T. (2015). Alcohol fuel type for mobility PMS project-energy transition, IFP School publication Paris.
5 Busic, A., Mardetko, N., Kundas, S., Morzak, G., Belskaya, H., Santek, M.I.,Komes, D., Novak, S., and Santek, B. (2018). Bioethanol production from renewable raw materials and its separation and purification: A review, Food Technology and Biotechnology, 56(3),

298-311.
6 Anuj, K.C., Ravinder, R., Lakashmi, V. R.& Ravindra, P. (2007). Economic and environmental impact of bio-ethanol production Technology. Biotechnology and Molecular Biology Review2(1); 14-32.
7 Amutha, R., and Gunasekaran, P. (2001). Production of ethanol from liquefied cassava starch using co-immobilized cells of zymomonas mobilis and saccharomyces diastaticus. Journal of Bioscience Bioengineering. 92(6), 560-564.
8 Adelekan, B.A., (2010), Investigation of ethanol productivity of cassava crop as a sustainable source of biofuel in tropical countries. African Journal of Biotechnology, 9(15) pp5643-5650.
9 Ledward, D.A., Taylor, A.J., & Lawrence, R.A. (2003). Upgrading waste for food and feeds, 3rd edn. Butterworth,U.S.A. 321.
10 Rolz, C., & Cab,S.,rera Ethanol from sugar cane flask experiment using the EX-FERM Technique, Applied and environmental microbiology 40(3) pp466-471, 1980.
11 Genshi, S., Khan, A.F., Suresh, M., Senthil, (1995). Character association for alcohol yield in sweet Sorghum. Madras Agricultural journal 82(5), pp361-363.
12 Bryan, W.L., (1890). Solid state fermentation of sugars in sweet sorghum, Enzyme and microbiology, 12(6), pp437-442.
13 Nikolov, T., Bakalova, N., Petrova, S., Spasov, S., Kalev, D. (2000). An effective method of bioconversion of delignified waste cellulose fibers from the paper industry with a cellulose complex, Resource Technology 71(1), pp 1-.
14 Balat, M., Balat, H. (2009). Recent trends in global production and utilization of bio-ethanol fuel, Applied Energy 86(11), pp 2273-2282.
15 Wikie, A.C., Richard, K.J., & Owens J.M. (2000). Spillage characterization and anaerobic treatment of ethanol spillage from conventional and cellulosic feed stock, Biomass and Bioenergy 19(2)pp 63-102.
16 Mojovic, I., Nicolic, S., Rakin, M., Vukasinovic, M (2006).rom corn meal Production of bioethanol from corn meal hydrolysate, Food, 8(12), pp 1750-1755.
17 Ensinas, A.V., Modesto, M., Nebra, S.A., Serra, I. (2009). Reduction of irreversibility generation in sugar and ethanol production from sugar cane, Energy 34(5), pp 680-688.
18 Salassi, M.E. (2007). The economic feasibility of ethanol production from sugar crops, Lauisiana Agricultural magazine. Winter issue.
19 Naik, S.N., Goud, V.V., Rout, P.K., Dalai, A.K. (2010). Production of first and second generation biofuels: A comprehensive review. Renewable and Sustainable Enegy Reviews. 14(2), 578-597.
20 Ekert, C.T., Trigo, E.P. Albreecht, L.P. Albrecht, A.J.P., Christ, D., Santos, W.G., Berkembrock, E., Egewart, V.A. (2018). Maze ethanol production in Brazil: Characteristics and perspectives, Renewable and Sustainable Energy Review, 82, 3907-3912.
21 Sarkar, O.J., Hhosh, S.K., Baenerjee, S., Aikat K. (2012). Bioethanol production from agricultural waste : An overview, Renew. Energy, 37(1), pp 19-27.
22 Cardona, C.A., Quintero, J.A. & Paz, I.C. (2010). Production of ethanol from sugarcane bagasse: Status and prospective, Bioresource Technology, 101(13), pp4754-4766.
23 Kosugi, A., Kondo, A., Ueda, M., Murata, Y., Vaithanomstat, P., Thanapase, W. (2009). Production of ethanol from cassava pulp via fermentation with a surface engineered yeast strain displaying glucoamylase, Renew Energy, 34, 1354-1358.
24 Lingang, S., Phang, L.Y., Wasoh, M.H., Abd-Aziz, S. (2012). Sago pitch residue as an alternative cheap substrate for fermentable sugar production, Applied Biotechnology, 167(1) pp122-131.
25 Srioth, K., Piyachomkwan, K., Wanlapatit, S., Nivitchanyong, S. (2010). The promise of a technology revolution in cassava bioethanol: From Thai practice to the world practice. Fuel, doi: 10.1016/j.fuel.2009.17.008
26 Leng, R., Wang, C., Zheng, C. Dial, D., Pu, G., (2008). Life cycle inventory and energy analysis of cassava-based fuel ethanol in China. J. Cleaner Prod 16:374-384.
27 Yu, S., & Tao, J. (2009). Energyefficiency assessment by the cycle simulation of cassava based fuel ethanol for automotive use in Chinese Guangxi context. Energy, 34:22-31.
28 Zhou, Z., Jang, H., & Quin, L. (2007). Life Cycle Sustainability assessment of fuels.

Fuel. 88:256-263.

29 Baki, A.S., Bande, Y.M. and Bello, A. (2020). Coparatve Studies on bioethanol Production fromCassava peels using saccharomyces cerevisiae and Zymomonas Mobilis. Open Access Journalof Biomedical Engineering and Biosciences, 3(5), DOI:10.32474/OAJBEB.2020.03.000174 .

30 Obianwa, C., Edak, E.A. Igile, G. (2016). Bioethanol Production from cassava peel using different microbial inoculants. African Journal of Biotechnology, 15(10), 1608-1612.

31 Aditomere, K.O. (2015). Production of Bioethanol As An Alternative Source Of Fuel Using Cassava And Yam Peels As raw aterials. International Journal of Innovative Scientific and Engineering Techologies Research 3(2):28-44.

32 Oyeleke, S.B., Dauda, B.E.N., Oyewole, O.A., Okoliegbe, L.N. & Ojebode, T. (2012) Production of Bioethanol From Cassava and Sweet Potato peels. Advances on environmental Biology, 6(1): 241-245.

33 Daniel, C.S.R., Evariste, B.G.K. (2017). Kinetics of Bioethanol Production from Waste Sorghu Leaves Using Saccharomyces cerevisiae BY4743. Fermentation, 3, 19, doi.10.3390/fermentation3020019

34 Sanette, M. and Tando, Y.Q. (2012). Cassava as feedstock for ethanol production in South Africa. African Journal of Biotechnology, 12(310, pp4975-4983.

35 Cardona, C.A. & Sanchez, O.J. (2007). Fuel ethanol production process design trends and integration opportunities. Bioresource Technology, 98(12), pp 2415-2457

36 Kroumov, A.D., Modenes, A.N. & Tait, D.M.C. (2006) . Development of new unsaturated model for simultaneous saccharification and fermentation of starch to ethanol by recombinant strain. Journal of Biochemical Engineering, 28: 243-255.

37 Nicholas, S.B., Hyde, M. & Henry, W. (2002). Fruit pomace as fuel. Transaction of American Society of Agricultural Engineers, 121, 11-14.

38 McMikeen, E.A., Olley, J., Ratkowsky, D.A., Ross, T. (2002). Predictive microbiology: towards the interface and beyond, Intrernational Journal of Food Microbiology, 73(2-3), pp395-407.

39 Phisalaphung, M., Srirattana, N. & Tanthapanichakoon, W. (2006). Mathematical modeling to investigate temperature effect on kinetic parameters of ethanol fermentation, Biochemical Engineering Journal , 28(1), 36-43.

30 Tat, M.E. 7 Van Gerpen, J.H. (1999). The kinematic viscosity of Biodiesel and its blends with diesel Fuel. JAOCS. 76, 1511-1513.

41 Endah, M.M.P., Rachimoellah, M., Nidya, S. & Ferdy, P. (2012). Biodiesel production from kapok seed oil (*Ceibai pentandra*) through the transesterification process by using Cao as catalyst, *International Research Journal,* 12(2),3-7.

42 Umeuzuegbu, J.C. (2020). Synthesis and Optimization of Biodiesel from Castor Seed Oil. World Journal of Innovative Research, 8(5), 47-60.

43 Ezekwe, C.C. & Ajiwe, V. (2014). The variations of physiochemical properties of biodiesel blends with the blend ratios. *International Journal of Science Innovations and Discoveries*, 4, 11-14.

44 Muhaj, S. & Sutjahjo, D.H. (2018). The Characteristics of bioethanol made of vegetable raw materials, IOP Conference Series: Material Science and Engineering 296, 012019 doi: 10.1088/1757-899X/296/1/012019.

45 Fogler, H.S. (2004). Elements of chemicalreaction engineering. Third edition .Prentice Hall of India Private Limited, New Delhi, pp 223-281.

46 Octave, L. Chemical reaction engineering.(2004). Third edition. John Wiley & Sons, New York, pp1-77.