Dilute Acid Hydrolysis of Cowpea Hulls: A Kinetic Study

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Abstract— In this study, dilute acid hydrolysis of cowpea hulls was carried out in two stages under the following conditions: pre-hydrolysis (4%v/v H_2SO_4, 121˙C, 30 minutes) and hydrolysis (at 10% and 15% v/v H_2SO_4,varied at different temperatures 150 ºC, 160 ºC, 170 ºC and 180 ºC for 2.5 hrs.). The substrate was characterized using both Fourier transform infrared spectroscopy and proximate analysis. The percentage lignocellulosic composition of the substrate was obtained for cellulose, hemicellulose and lignin as 34%, 14% and 4.7% respectively. Maximum glucose concentration of 8.09g was obtained using 10%v/v acid concentration at 170˙C after a reaction time of 90min. Saeman's model gave a good fit for the experimental data. Activation energy for glucose formation using 10%v/v and 15%v/v H_2SO_4 was obtained as 38.28KJ and 82.204KJ respectively. From the results obtained it can be concluded that cowpea hulls can be converted to a useful product.

Keywords— Acid hydrolysis; Agricultural waste; Cowpea Hulls; Glucose; Kinetic study

I. INTRODUCTION

As the world population continues to increase, the amount of waste generated also increases. According to United Nations [1], 140 billion metric tons of biomass is generated every year from agriculture and if not properly disposed or managed can cause environmental problems. One of these waste generated in Africa is cowpea hulls. Cowpea hull is the by-product generated from processing cowpea into flour and other cowpea food recipes such as moi-moi and akara, Cowpea is one of the most commonly consumed legumes in Nigeria. More than 5.4 million tons of dried cowpeas are produced worldwide, with Africa producing nearly 5.2 million: Nigeria, the largest producer and consumer, accounts for 61% of production in Africa and 58% worldwide [2]. With these large volume of cowpea produced in Nigeria, a larger amount of cowpea hulls are also generated. Cowpea hulls are classified as lignocellulosic biomass as it is a non-starchy biomass. These materials contain sugar polymerized in form of cellulose and hemicellulose, which can be liberated by hydrolysis and subsequently fermented to ethanol by microorganisms [3]. Hydrolysis can be achieved by using acids or enzymes. There are two types of acid hydrolysis: dilute and concentrated acid hydrolysis. Most times dilute acid hydrolysis is carried out as a pre-treatment method for enzyme hydrolysis. Dilute acid hydrolysis can also be carried out in two stages: The first stages under mild conditions while the second stage under severe conditions. In the first stage hemicellulose is converted to monomeric sugar leaving a residue mostly made up of cellulose and lignin. In the second stage the cellulose exposed is then converted to sugar also. According to [4], the two-stage dilute acid process is preferred to one-stage dilute acid hydrolysis because: a higher sugar yield is expected from separate hydrolysis of hemicellulose and cellulose, energy consumption is also minimized as liquid is removed after the first stage hydrolysis, less sugar degradation and formation of fewer fermentation-inhibiting components are formed.

The aim of this work is to convert cowpea hulls to a more useful product (glucose) via a two- stage dilute-acid hydrolysis process using sulphuric acid. This process route was chosen based on the advantages it offers over the single stage. The specific objectives of this work are: to characterize the substrate, to pre-hydrolyse the substrate using diluted Sulphuric acid (H2SO4), to determine the effect of process variables such as temperature, acid type and acid concentration on the second stage and evaluate the kinetics of the second stage of hydrolysis process for the substrate.

II. MATERIALS AND METHODS

A. Raw materials and characterization

The Cowpea hulls (CPH) was collected from local bean cake (akara) processors in Owerri Imo state, Nigeria. The substrate was sun dried for 3 days and afterwards grinded and sieved using a 1 mm mesh size sieve. The sievates were stored in a cool dry place for subsequent use. The lignin, ash and moisture composition of the substrate was quantified using standard analytical procedure for proximate analysis by [5] while the hemicellulose and cellulose composition were quantified using Crampton and Maryrand method [6].

B. First stage: pre-hydrolysis

4% (v/v) sulphuric acid concentration was used for the hydrolysis process at 121°C for 30minutes using 20% substrate to acid solution ratio. Afterwards, the autoclaved substrate was placed inside an ice cold water to stop reaction. The solid residue obtained was oven dried at 60 °C for 2hours and was also analysed for its lignocellulosic fraction composition before been stored in a cool place for subsequent use. The filtrate obtained was analysed for its glucose concentration.

C. Second stage: hydrolysis

Solid residue obtained from the pre-hydrolysis process was dissolved in acid solution at the ratio of 1:10 respectively. The solution was stirred with magnetic stirrer at 150rpm and temperature at 140 °C. Samples were drawn at intervals of 30minutes for 2.5 hours. Each sample was placed in an ice cold water to quench the reaction process. The process was repeated for 150 °C, 160 °C, 170 °C and 180 °C using acid concentration of 10% (v/v) and 15% (v/v) sulphuric acid. The glucose composition of the hydrolysate were analysed using the DNS (3, 5- Di nitro salicylic acid) method with glucose as standard [7]. Absorbance was measured at 540nm with UV spectrophotometer (MODEL: Searchtech Spectrumlab 755s). Glucose concentration measured was reported in g/100 gram of dry weight of substrate.

D. Kinetic model

The experimental data obtained were fit into the Saeman model [8], this model was designed for the hydrolysis of cellulose from fir using sulphuric acid, and it assumes the reaction proceeds according to equation 1.

$$\text{Cellulose+ water} \xrightarrow{k_1} \text{glucose} \xrightarrow{k_2} \text{decomposition products} \quad (1)$$

Where k_1 is the rate of conversion of cellulose to glucose and k_2 is the rate of decomposition of glucose. Both have units of the reciprocal of time (min^{-1}). The reaction of cellulose and water is a pseudo first order reaction with water in excess and irreversible. Solving Equation 1 gives Equation 2.

$$G = \left(\frac{k_1 C_0}{k_2 - k_1} \right) \quad \left(e^{-k_1 t} - e^{-k_2 t} \right) \quad (2)$$

It is assumed that all the cellulose hydrolyses to glucose; therefore the initial cellulose concentration, C_O (in gL^{-1}) is equal to the potential concentration of glucose Gn_0

obtainable from the cellulose. Therefore Equation 2 becomes:

$$G = \left(\frac{k_1 Gn_0}{k_2 - k_1} \right) \quad \left(e^{-k_1 t} - e^{-k_2 t} \right) \quad (3)$$

Gn_0 can be determine analytically as described by [9] equation 4

$$Gn_0 = \frac{FZ\rho}{WSR} \quad (4)$$

Where F is stoichiometric factor due to hydration of molecule during the hydrolysis, ρ is the density of hydrolysis, z is composition of the raw material for the polysaccharides and WSR is the water to solid ratio. The temperature dependence of reaction rates was described using Arrhenius equation, Equation 5.

$$k_i = k_{i0} e^{\frac{-E_a}{RT}} \quad (5)$$

Where,

ki = Kinetic coefficient (i = 1 or 2) (min-1)
$ki0$ = Pre-exponential factor (i = 1 or 2) (min-1)
Ea = Activation Energy (kJ mol^{-}1)
R = Gas Constant, 8.314 (kJ mol^{-}1 K^{-}1)
T = Temperature (K)
Linearizing Equation 5 gives Equation 6.

$$Ink_i = \frac{-E_a}{R} \frac{1}{T} + Ink_{i0} \quad (6)$$

Activation Energy was obtained by plotting Ink versus $1/T$

III. RESULT AND DISCUSSION

A. Characterisation

The following results were obtained from the proximate analysis of the cowpea hull: cellulose 34%, 14% hemicellulose, 38% lignin, 4.70 % ash. The high cellulose and hemicellulose content makes this waste a good substrate for hydrolysis. The compositional analysis of bean hulls by [10], gave 78% hollocellulose (cellulose and hemicellulose) 51% α-cellulose, Pentosane 26%, lignin 10%, ash 3.36%. The values for Lignocellulosic fractions of cowpea seed hulls were also reported by [11] to be 28% hemicellulose, 40% cellulose and acid detergent lignin 13%.

B. Pre-hydrolysis

Results obtained analysing the solid residue form pre-hydrolysis shows lignocellulosic fraction composition of cellulose: 39.95%, hemicellulose: 2.08%, lignin: 46.34%, ash: 1.50% and moisture content of 10.13%. The first stage hydrolysis caused 85% reduction in the initial hemicellulose composition of the cowpea hulls. This shows that hemicellulose is easily hydrolysed and can be reduced significantly under this reaction condition. This makes the cellulose more exposed to the actions of acid in the second stage.

C. Effect of process parameters on hydrolysis of cowpea hulls

1) Effect of temperature and time on glucose yield

The glucose yield produced from acid hydrolysis of CPH at different temperatures using 10% v/v sulphuric acid can be seen in Fig. 3 and 4. At 140^0C to 170^0C, the peak glucose yield occurred at 90 minutes reaction time. At 140^0C, there was a steady rise in glucose concentration until a peak of 7.090g of glucose was obtained. But due to glucose decomposition 10% of the maximum glucose formed was lost. 7.437g of maximum glucose was obtained at 150^0C; this also experienced a slight loss of 9% due glucose degradation. At 160^0C, 7.664g of glucose was obtained at a reaction of 90mins. 12% was lost due decomposition as time increases. At 170^0C, a steady rise was observed until a maximum point of 8.090g of glucose was obtained which afterwards experienced a decline in concentration resulting to 25% of the maximum glucose lost due to decomposition of glucose. At 180^0C however, the peak (6.925g) occurred at 60 minutes. This yield which was lower than the peak obtained at other temperatures also experienced loss (43%) as time increases. This is attributable to the decomposition reaction which was severe at this temperature.

Fig. 5 and 6 shows the glucose yield produced from acid hydrolysis of CPH at different temperatures using 15% v/v sulphuric acid. For temperatures of 140^0C to 160^0C, the peak values of the glucose yields were all obtained at 90 minutes reaction time. At 170^0C and 180^0C, the peak values occurred at 60 minutes and 30 minutes respectively. At 140^0C a peak value of 2.87 g of glucose was obtained at 90 minutes reaction time with 74% of this left by the end of the reaction. At 150^0C a peak value of 4.87 g of glucose was obtained with 75% of it left by the end of the reaction. At 160^0C a peak value of 4.68 g of glucose was obtained with 84 % of this left by the end of the reaction. At 170^0C a peak value of 5.00 g of glucose was obtained with 62 % of this left by the end of the reaction. Also the glucose generation at 180^0C had the highest reaction rate and reached its peak value of 5.47 g of glucose/100 g of dry CPH after 30 minutes of reaction. The reaction also decomposed rapidly with only 5.6 % of this glucose yield remaining by the end of the reaction. The decreases in glucose concentration observed at the different temperatures are due to the decomposition of the glucose to degradation products which increases as time increases.

A similar trend of rise and fall in glucose yield was also observed by [12] in their kinetic study on acid hydrolysis *Cenchrus ciliaris*. Reference [13] obtained optimum condition for wheat bran pre-treatment to be: acid concentration of 1.75% (w/v), at 190 °C for 10 min. Another study obtained 73.4 g l^{-1} glucose-rich solution in the second-stage of hydrolysis by using 40% acid concentration at 80 °C [14]. Reference [15] also obtained a high glucose yield at 180°C after 5mins reaction time. These result generally agrees with the fact that high sugar yield will be obtained at high acid concentration, low temperature and reduced reaction time or low acid concentration, high temperature and reduced reaction time.
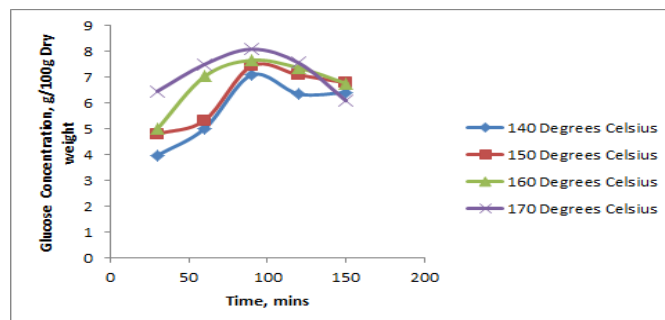

Fig. 1: Effect of temperature and time on glucose yield from CPH hydrolysed at 140^0C - 170^0C with 10% v/v sulphuric acid
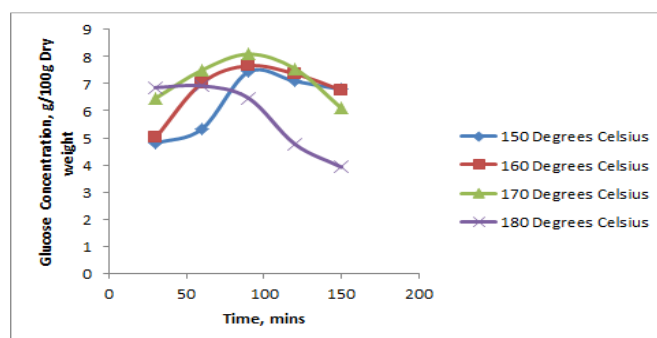

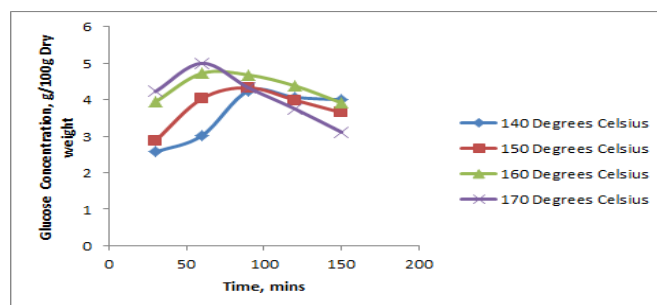Fig. 2: Effect of temperature and time on glucose yield from CPH hydrolysed at 150^0C - 180^0C with 10% v/v sulphuric acid

Fig. 3: Effect of temperature and time on glucose yield from CPH hydrolysed at 140^0C - 170^0C with 15% v/v sulphuric acid.
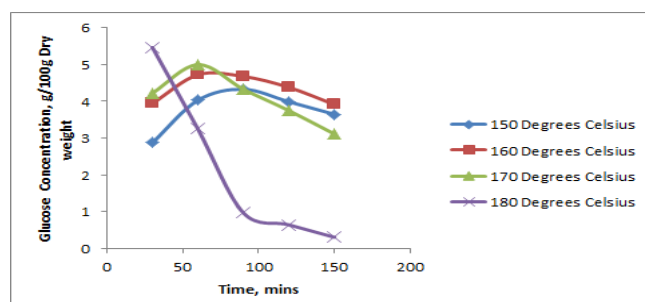

Fig. 4: Effect of temperature and time on glucose yield from CPH hydrolysed at 150^0C - 180^0C with 15% v/v sulphuric acid.

2) Effect of acid concentration on Glucose yield from CPH

Fig. 7 to 11 shows the effects of sulphuric acid concentration on glucose yield for cowpea hulls hydrolysed at 140^0C to 180^0C. For all the temperatures, the hydrolysis with 15% sulphuric acid gave lower glucose yields than with 10% sulphuric acid. This implies that the 15% sulphuric acid was harsh on the substrate leading to fast degradation of sugar produced to unwanted products.
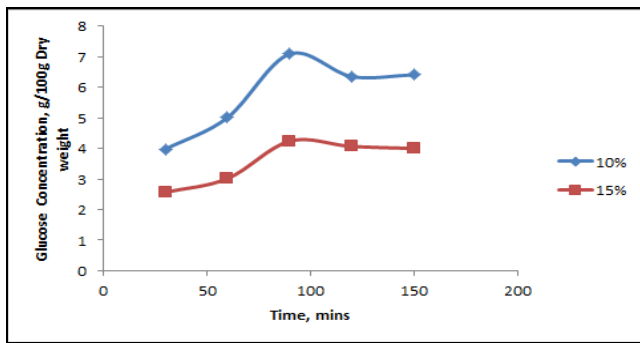
Fig.5: Effect of acid concentration on glucose yield from CPH hydrolysed at 140^0C with sulphuric acid.
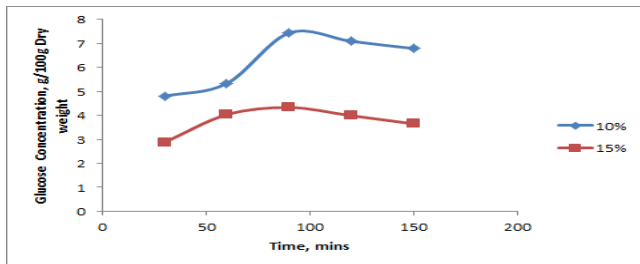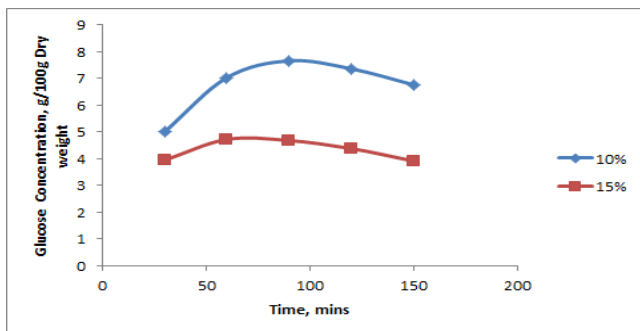

Fig. 6: Effect of acid concentration on glucose yield from CPH hydrolysed at 150^0C with sulphuric acid.

Fig. 7: Effect of acid concentration on glucose yield from CPH hydrolysed at 160^0C with sulphuric acid.
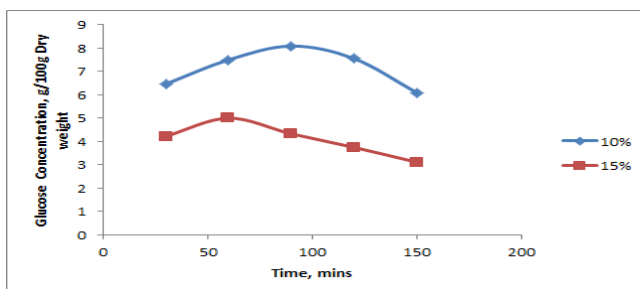

Fig. 8: Effect of acid concentration on glucose yield from CPH hydrolysed at 170^0C with sulphuric acid.
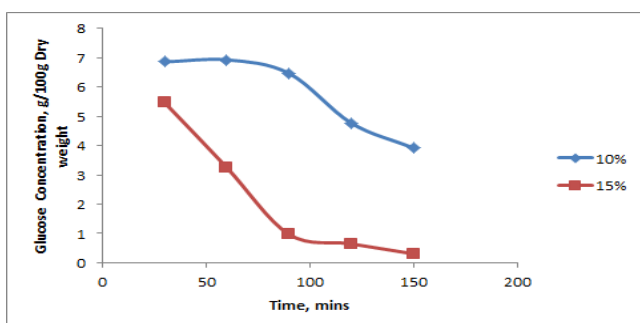

Fig. 9: Effect of acid concentration on glucose yield from CPH hydrolysed at 180^0C with sulphuric acid.

D. Kinetic study

Equation (4) gives the potential glucose concentration, Where F is stoichiometric factor due to hydration of molecule during the hydrolysis and is given by 180/162 for hexoses such as glucose [9], ρ is the density of hydrolysate (10 g/l), z is composition of the raw material for the polysaccharides (52.51g/100g of raw material dry basis) and WSR is the water to solid ratio used (10.636g/g). From equation (5), the potential composition of glucose in the liquors was obtained as 40.56g glucose/l.

The R-square values obtained from fitting the experimental data into the Saeman model are shown in Tables 1 and 2. These values are high which indicates that the experimental data can be described by Saeman model. It was observed from Table 1, that the values of K_2 are greater than the values obtained of K_1 and both increased with increase in temperatures. This shows that the rate of glucose degradation was more than the rate of glucose formation. Similar trend was also obtained for 15% acid concentration. Arrhenius plot for glucose formation from Cowpea hulls hydrolysed at 140^0C - 180^0C with 10% v/v and 15% Sulphuric acid (see Fig. 12 and 13 respectively). The R^2 values obtained shows a good agreement between the natural log of K and inverse of temperature. The activation energy obtained for 10% acid concentration is 38.28 KJ for K_1 and 28.497 KJ for K_2. A similar trend was also observed with 15% acid concentration which gave activation energy 82.204 KJ for K_1 and 60.499 KJ for K_2. Reference [12], obtained activation energy of 24KJ/mol for glucose formation in a acid hydrolysis of *Cenchrus ciliaris* particles. Reference [16], obtained the activation energy value of 29.96KJ for glucose production form cassava root sieviate using 10% acid concentration.

TABLE I
KINETIC PARAMETERS OF SAEMAN MODEL FOR 10% V/V H_2SO_4 HYDROLYSIS OF CPH

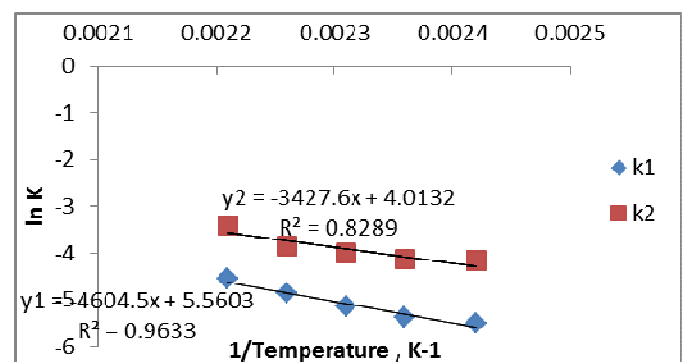| Temperature (°C)	K_1	K_2	R^2
140	0.004096	0.01562	0.8520
150	0.004646	0.01592	0.7458
160	0.005827	0.01809	0.9990
170	0.007692	0.02154	0.8125
180	0.010630	0.03314	0.9905

Fig. 10: Arrhenius plot for glucose formation from CPH hydrolysed at 140^0C - 180^0C with 10% v/v H_2SO_4

454

TABLE II
KINETIC PARAMETERS OF SAEMAN MODEL FOR 15% V/V H_2SO_4
HYDROLYSIS OF CPH

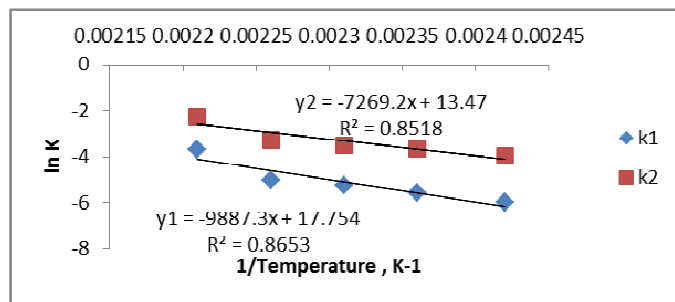| Temperature ($^{\bullet}$C)	K_1	K_2	R^2
140	0.002513	0.01849	0.8715
150	0.003759	0.02626	0.9712
160	0.005170	0.02975	0.9274
170	0.006676	0.03836	0.9854
180	0.025120	0.10260	0.9841

Fig. 11: Arrhenius plot for glucose formation from CPH hydrolysed at 140^0C - 180^0C with 15% v/v H_2SO_4

IV. CONCLUSIONS

Dilute acid hydrolysis of cowpea hull with sulphuric acid is a process that can be harnessed in the production of glucose and its allied products. The composition and abundance of this substrate makes it a useful waste for biotechnological processes. The hydrolysate obtained can be used for fermentation because of it high glucose composition.

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