resource and is oxygenated thereby provides the potential to reduce emission in compression-ignition engines (Hansen *et al*, 2005).

In this era, bioethanol can be produced from staple food materials such as molasses from sugar cane, corn, and cassava tubers. Wastes from industrial agriculture are potentially used raw material for bioethanol production because of its availability is still abundant, the cost is relatively cheap and could reduce competition with staple food (Witantri *et al*, 2009). Lignocellulosic biomass is a cheap, renewable, abundantly available resource and its conversion to glucose and other fermentable sugars has been considered to be an attractive route to ethanol production (Cao *et al*, 1996; Cazetta *et al*, 2007).

Nigeria is the highest producer of cassava in the world, producing higher than Brazil, Thailand and Indonesia. Industrial and local processing of cassava to food and other product has led to useful products (Mohammed *et al*, 2013). In Nigeria, corn is processed to a variety of diets including pap which is a major diet for weaning, and the capacity for corn production in Nigeria is high (Orji *et al*, 2016). Corn cobs form 30% of maize agro wastes (Zakpaa *et al*, 2009). These wastes end up polluting the surface and underground water (Mohammed *et al*, 2013).

The present annual ethanol production rate of 134 million Liters in Nigeria is grossly inadequate and the industries producing bioethanol in the country inporttheir raw materials from Brazil in spite abundant of plant wastes that could serve as raw materials. Nigeria have to explore the abundant agricultural wastes to produce enough ethanol for consumption and exportation. This will serve as a source of employment and income to the citizenry in general. It will also curtail spending Nigeria's scarce resources in importation of ethanol (Mohammed *et al*, 2013). The aim

of this work is to utilize the reducing sugar from cassava fibre and corn cobs which are agricultural wastes for bioethanol production.

2. Materials and Methods

Sample collection

Cassava fibre was obtained from Nigeria Starch Mill, Uli town, while Corncob was obtained from Ihiala town, all in Ihiala LGA, Anambra State, Nigeria. These substrates were collected in sterile polythene bags and transported to Microbiology Laboratory, Chukwuemeka Odumegwu Ojukwu University, Uli.

Substrate preparation

The corncob and cassava fibre were cut into small pieces, then washed under running tap water to remove sand and other dirty particles. They were dried at 50 ^{o}C for 48h and ground to fine particles.

Proximate analysis of substrates

Proximate analysis of the substrates was determined according to AOAC (AOAC, 2003).

Percentage moisture content, fibre content, ash content, protein content and carbohydrate content of the substrates were analysed.

Sulphuric acid (H_2SO_4) Hydrolysis

One hundred and fifty (150) grams of cassava fibre and corncob, were hydrolyzed separately with sulphuric acid of various concentrations (0.1, 0.5 and 1.0 M) at temperature of 100^{o}C for

30min. The resulting hydrolysate was neutralized with 2M NaOH solution and then dried in an incubator at 50 ^{o}C for 48h. The dried particles were ground to fine particles.

Hydrochloric acid(HCL) Hydrolysis

One hundred and fifty (150) grams of cassava fibre and corncob, were hydrolyzed separately with hydrochloric acid of various concentrations (0.1, 0.5 and 1.0 M) at temperature of 100^{o}C for 30min. The resulting hydrolysate was neutralized with 2M NaOH solution and then dried in an incubator at 50 ^{o}C for 48h. The dried particles were ground to fine particles.

Effect of time on hydrolysis

The concentration of acid with the highest reducing sugar production was selected and used to separately hydrolyse the cassava fibre and corncob at temperature of 100^{o}C at various time interval of 30, 45, 60 and 75 min(Abidin *et al*., 2014)

Determination of Reducing Sugar

Reducing sugar was estimated using the method of Miller (1959). One millilitre of Dinitrosalicyclic acid (DNS) was added to 1 ml of the hydrolysate in test tubes and mixtures heated at a temperature of 100 ^{o}C for 10 min. The test tubes were cooled rapidly in tap water and volume adjusted to 12 ml with distilled water. A blank containing 1ml of distilled water and 1ml of DNS was prepared. The optical density (OD) was read against the blank in a spectrophotometer at 540 nm. The concentration of the reducing sugar was estimated from standard glucose curve.

Alcoholic fermentation

Each of the hydrolysed corncob and cassava fibre (150g) was added into each of cotton plugged vessel (4L size) containing 1 litre of sterile distilled water and pH was adjusted to 5.0. The fermentation medium was sterilized and thereafter one percent (v/v) of a 24h seed inoculum of

Baker's yeast was used to inoculate it. The fermentation was carried out at 27^0C for 168 h. At interval of 24 h, samples were taken from the medium and used for determination of ethanol, reducing sugar concentration and pH.

3. Results

Proximate Analysis

Figure 1 shows the results of the proximate analysis of cassava fibre and corncob. The cassava fibre had 17.8% moisture, 1.3% ash, 4.1% fibre, 5.3% crude protein, 68.4% carbohydrate and 2.6% lipid, while corncob had 15.7% moisture, 4.0% ash, 25.7% fibre, 5.9% crude protein, 45.8% carbohydrate and 3.3% lipid. Corncob gave the highest values in all except for moisture and carbohydrate content.

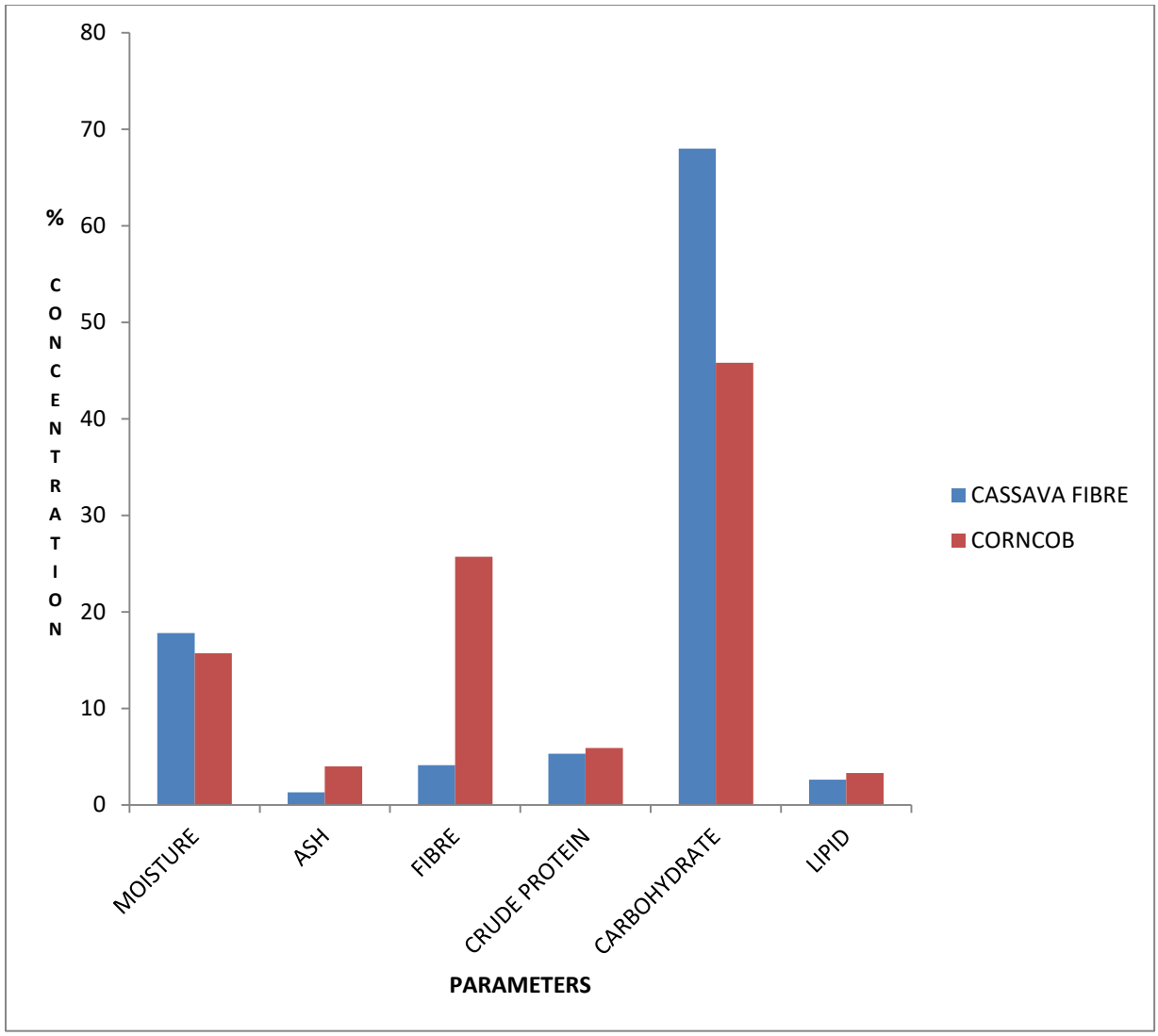

Figure 1: Proximate analysis of cassava fibres and corn cobs

Figure 2 shows the result of the effect of different concentrations of sulphuric acid on hydrolysis of cassava fibre and corncob for reducing sugar production. Sulphuric acid (0.1M) gave the highest reducing sugar concentration of 5.08 (mg/ml) and 5.10 (mg/ml) for cassava fibre and corncob respectively. Higher concentrations of the acid (0.5 and 1.0 M) produced lower concentration of sugar. There was no significant difference ($P \geq 0.05$) in reducing sugar concentration and sulphuric acid hydrolysis of corncob and cassava fibre.

Fig 3 shows the result of effect of different concentrations of hydrochloric acid on hydrolysis of cassava fibre and corncob for reducing sugar production. Hydrochloric acid (0.1M) gave the highest reducing sugar concentration of 2.06 (mg/ml) for cassava fibre, while 1.0 M of hydrochloric acid gave the highest reducing sugar concentration of 2.89 (mg/ml) for corncob. Higher concentrations of acid (0.5 and 1.0 M) produced lower concentration of sugar in the case of cassava fibre, while lower acid concentrations (0.1 and 0.5 M) produced lower sugar yield for corncob. There was no significant difference ($P \geq 0.05$) in reducing sugar concentration and hydrochloric acid hydrolysis of corncob and cassava fibre.

Fig 4 shows the result of the effect of time on hydrolysis of cassava fibre and corncob with 0.1M H_2SO_4. The highest reducing sugar concentrations of 5.32 and 5.51 (mg/ml) were obtained from cassava fibre and corncob wastes at 75min. The lower time intervals (30, 45 and 60 min) produced reduced amount of reducing sugar. There was no significant difference ($P \geq 0.05$) in reducing sugar concentration and time of hydrolysis of cassava fibre and corncob.

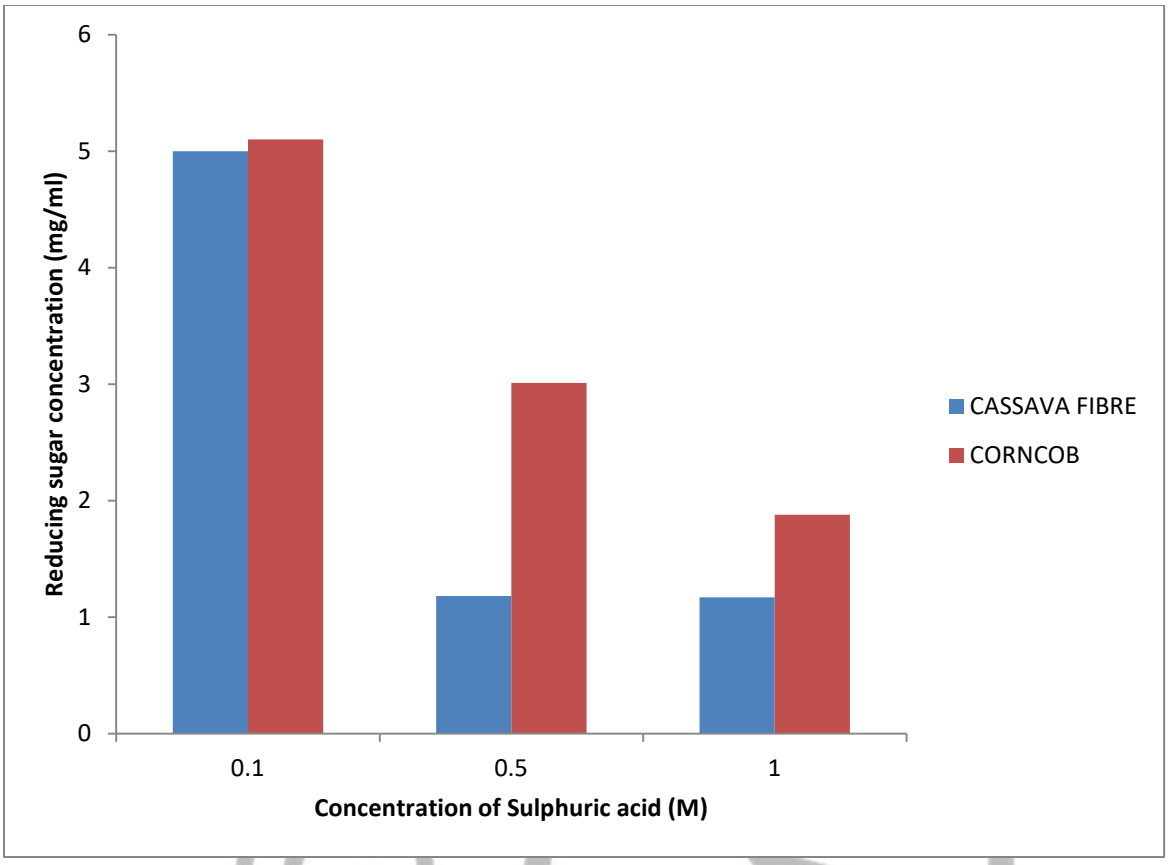

Figure 2: Effect of different concentrations of sulphuric acid on hydrolysis of cassava fibre and corncob wastes

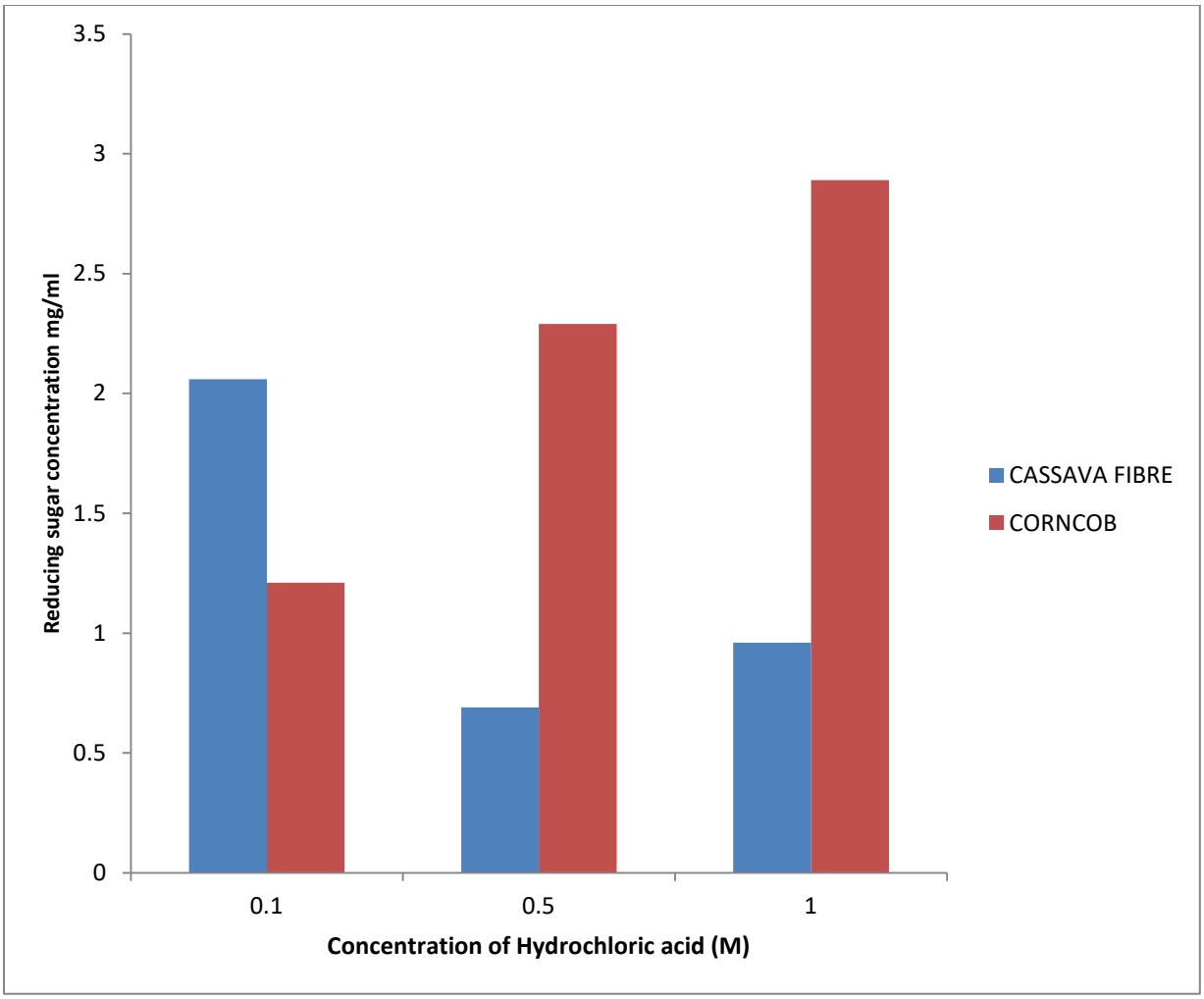

Figure 3: Effect of different concentrations of hydrochloric acid on hydrolysis of cassava fibres and corncob wastes

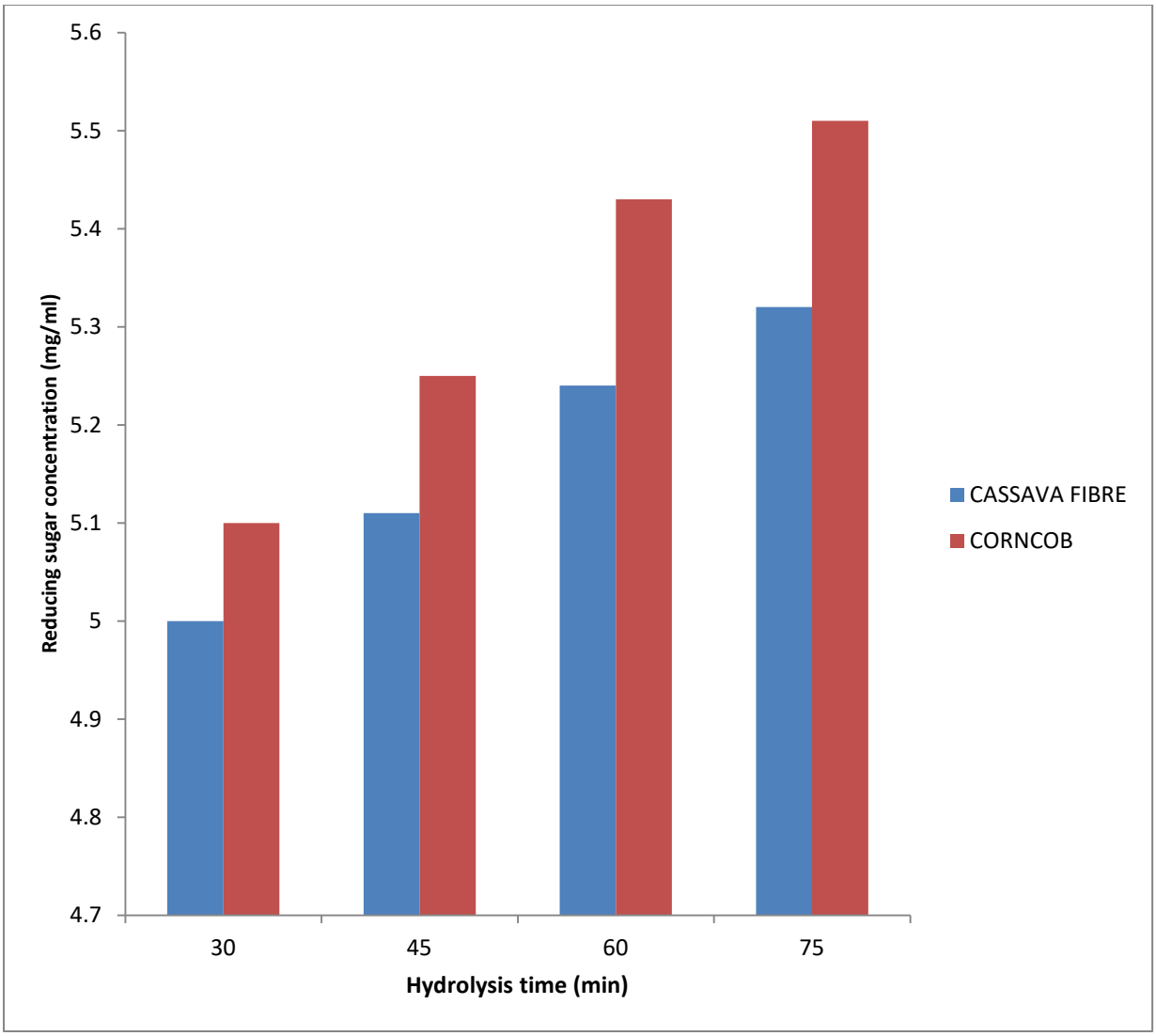

Figure 4: Effect of time on hydrolysis of cassava fibres and corncob wastes with 0.1M H_2SO_4

Figure 5 shows the effect of fermentation time on bioethanol production from corncob by Baker's yeast. Maximum ethanol yield 7.2 (%v/v) was recorded at 72 h and the yield reduced progressively with increase in fermentation duration. It was noticed that both the pH and reducing sugar , decreased progressively till the end of the fermentation.

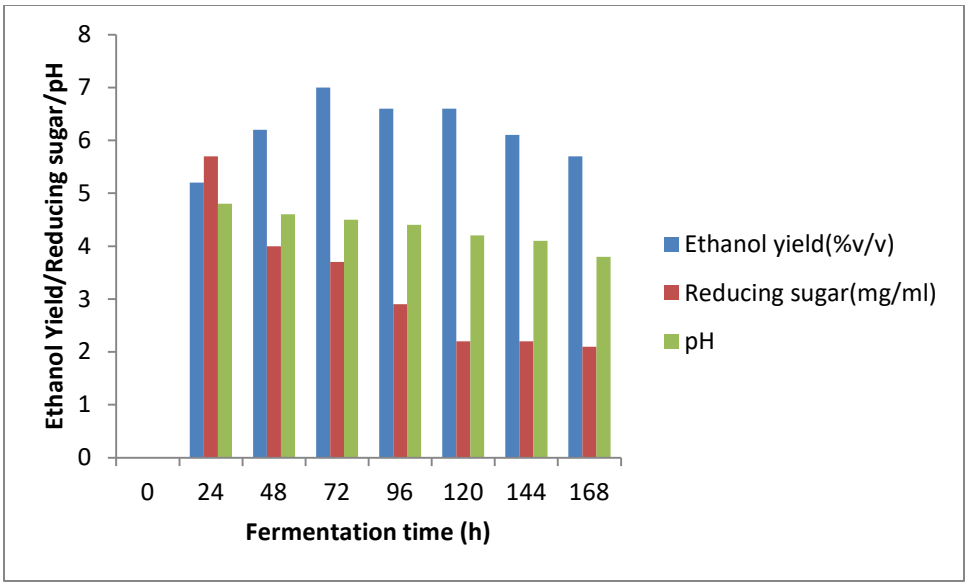

Figure 5: Effect of Fermentation time on Bioethanol production from corncob by Baker's yeast.

Figure 6 shows the effect of fermentation time course on bioethanol production from cassava fibre by Baker's yeasts. Maximum ethanol yield 6.4 (%v/v) was recorded at 120 h and the yield reduced progressively with increase in fermentation duration. It was noticed that both the pH and reducing sugar, decreased progressively till the end of the fermentation.

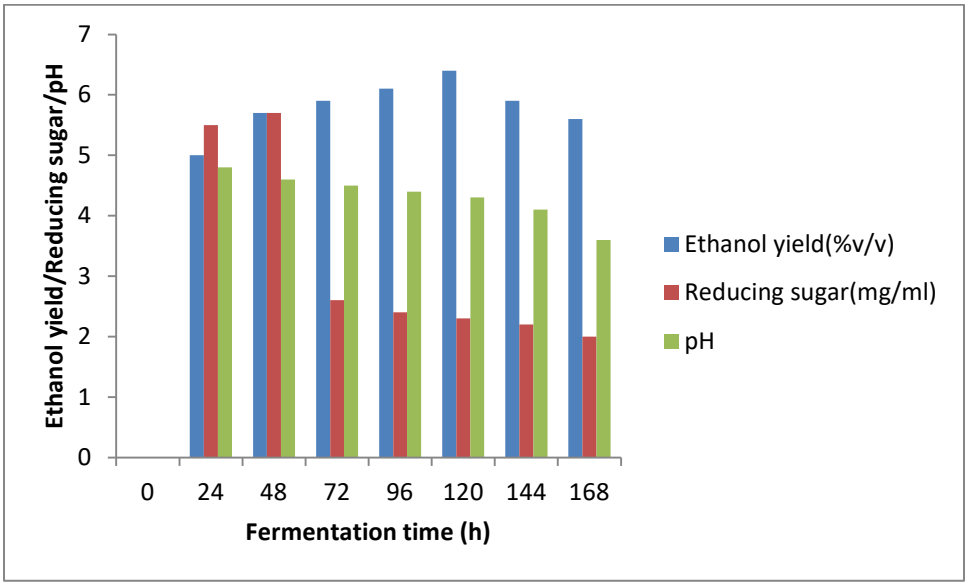

Figure 6: Effect of Fermentation time on Bioethanol production from cassava fibre by Baker's yeast.

4. Discussion

In the study the proximate analysis values for corncob were different from that reported by Ibeto *et al*. (2014). According to Ibeto *et al*. (2014) crude fibre was 22%, moisture content 60.31%, Ash 6.4%, fat 1.5%, protein 9.09% and carbohydrate 1.64%. Also, the proximate values for cassava fibre in the study differed from that reported for cassava peel and cassava pulp by Archibong *et al* (2016). Archibong *et al* (2016), had reported that protein was 4.9%, fibre 16.16%, Ash 5.9%, fat 1.3% for cassava peels, while for cassava pulp protein was 2.3%, fibre 3.4%, ash 2.5%and fat 1.4%. He opined that the variability in the values shown above could be attributed to differences in substrate, climatology, soil fertility, age of harvest and methods of processing.

The results obtained for reducing sugar production, revealed that 0.1 M concentration of H_2SO_4 stimulated maximum reducing sugar production from corncobs. The observation was contrary to the report of Akpan *et al*. (2005) who reported that 4.5M H_2SO_4 stimulated optimum yield of reducing sugar production from corncob. Also, Abidin *et al* (2014) reported increased reducing sugar production at 0.5 M of H_2SO_4. There was a drop in glucose concentration when hydrolyzed at a higher concentration of acid, glucose can be converted to levulinic and formic acid (Akpan *et al*., 2005) which leads to decreases in glucose yield.

It was found during the study that optimum ethanol production from hydrolyzed cassava fibre by Baker's yeast was recorded at 72 h. This was in contrast with the report of Akponah and Akpomie (2012). They observed enhanced ethanol yield from cassava effluent after 24 h. Again, it was also observed in this study that reducing sugar concentration decreased progressively with increase in fermentation time, which was similar to the findings of Akponah and Akpomie

(2012) and Akponah (2011). The progressive decrease in reducing sugar concentration could be due to the efficiency of the strains in utilization of reducing sugars. Reduction in reducing sugar concentration beyond the time for maximum ethanol yield in both substrates could be attributed to increased production of aldehydes, phenols, lactic acid and an increased sensitivity of strains to ethanol tolerance (Shyam *et al*., 2011).

5. Conclusion

Reducing sugar could be produced from cassava fibre and corncob by hydrolysis with sulphuric and hydrochloric acid. This has been revealed in the above study where various yields of reducing sugars were recorded, when the substrates were hydrolysed with sulphuric acid and hydrochloric acid respectively. The study also showed that the hydrolysed substrates(cassava fibre and corncob) can be utilized by baker's yeast for bioethanol production. The process for bioethanol production if well developed could lead to the commercial production of the product locally.

6. References

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