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COMPARATIVE ANALYSIS OF THE NUTRITIONAL COMPOSITION OF SUGAR PRODUCED FROM LIGNOCELLULOSIC SAWDUST AND COMMERCIAL GRANULATED SAMPLE FOR INDUSTRIALIZATION

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ABSTRACT

Sugar is the generic name for sweet-tasting, soluble carbohydrates, many of which are used in foods, found in the tissues of most plants. Scientifically, it is loosely refers to a number of carbohydrates, such as monosaccharides, disaccharides, or oligosaccharides, whose biopolymers are common in nature, through photosynthesis, this precipitate sourcing for sugar from lignocellulosic biomass like sawdust. *Lignocellulosic biomass which is the plant dry matter is the most abundantly available raw material on the Earth for the production of biofuel, concerns around greenhouse gas emissions necessitates the development of sustainable processes for the production of chemicals, foods materials, and fuels from alternative renewable sources. The steps involved in the production of sugar from lignocellulosic sawdust were in two stages; acid hydrolysis of blended sawdust using HCl been tested for higher percentage yield of sugar, and anaerobic fermentation of hydrolyzed sawdust which was then crystalized.* Proximate analysis was conducted on the sugar produced from lignocellulosic sawdust to ascertain its nutritional composition using FAO (2003) method, while the mineral content was determined using Instrumental Techniques. The overall nutritional potential was observed to be more beneficial than commercialized sugar except for the lower concentration of micro-element in the lignocellulosic sugar. There was higher moisture content in the prepared sugar which makes it susceptible to structural damage if granulized and it had lower carbohydrate content compare to the commercial sugar. It could be inferred from this work that, the sugar from ligocellulosic biomass was richer in macro nutrient and micro element than commercial granulated sample. *Therefore, Lignocellulosic sawdust is a good source of raw material for sugar production, which invariably may serve as a good source of food supplements*.

Key words: Anaerobic, Biomass, Biopolymers, Crystallization, Fermentation, Hydrolysis, Lignocellulosic, saccharides.

1.0 INTRODUCTION

Sugar is the generic name for sweet-tasting, soluble carbohydrates, many of which are used in foods. Simple sugars, also called monosaccharides, include glucose, fructose, and galactose. Compound sugars, also called disaccharides or double sugars, are molecules made of two bonded monosaccharides; Sucrose (glucose + fructose), lactose (glucose + galactose), and maltose (two molecules of glucose). White sugar is a refined form of sucrose. In the body, compound sugars are hydrolyzed into simple sugars. Longer chains of monosaccharides (>2) are not regarded as sugars, and are called oligosaccharides or polysaccharides. Starch is a glucose polymer found in plants, and is the most abundant source of energy in human food. Some other chemical substances, such as glycerol and sugar, alcohols, may have a sweet taste, but are not classified as sugar Ioelovich, et al., (2011).

Sugars are found in the tissues of most plants. Honey and fruit are abundant natural sources of simple sugars. Sucrose is especially concentrated in sugarcane and sugar beet, making them ideal for efficient commercial extraction to make refined sugar. In 2016, the combined world production of those two crops was about two billion tonnes. Maltose may be produced by malting grain. Lactose is the only sugar that cannot be extracted from plants. It can only be found in milk, including human breast milk, and in some dairy products. A cheap source of sugar is corn syrup, industrially produced by converting corn starch into sugars, such as maltose, fructose and glucose, OECD-FAO Agricultural outlook for 2020-2029, (2019).

As sugar consumption grew in the latter part of the 20th century, researchers began to examine whether a diet high in sugar,

especially refined sugar, was damaging to human health. Excessive consumption of sugar has been implicated in the onset of obesity, diabetes, cardiovascular disease, and tooth decay. Numerous studies have tried to clarify those implications, but with varying results, mainly because of the difficulty of finding populations for use as controls that consume little or no sugar. In 2015, the World Health Organization recommended that adults and children reduce their intake of free sugars to less than 10%, and encouraged a reduction to below5%, of their total energy in take, WHO guideline on sugar intake for adults and children (2015)

Scientifically, *sugar* loosely refers to a number of carbohydrates, such as monosaccharides, disaccharides, or oligosaccharides. Monosaccharides are also called "simple sugars", the most important being glucose. Most monosaccharides have a formula that conforms to $C_nH_{2n}O_n$ with n between 3 and 7 (deoxyribose being an exception). Glucose has the molecular formula $C_6H_{12}O_6$. The names of typical sugars end with-*ose*, asin "glucose" and "fructose". Sometimes such words may also refer to any types of carbohydrates soluble in water. The acyclic mono-and disaccharides contain either aldehyde groups or ketone groups. These carbon-oxygen double bonds (C=O) are there active centers. All saccharides with more than one ring in their structure result from two or more monosaccharides joined by glycosidic bonds with the resultant loss of a molecule of water (H₂O) per bond, Pigman, et al., (2002).

Monosaccharides in a closed-chain form can form glycosidic bonds with other monosaccharides, creating disaccharides (such as sucrose) and polysaccharides (such as starch o rcellulose). Enzymes must hydrolyze or otherwise break these glycosidic bonds before such compounds become metabolized. After digestion and absorption, the principal monosaccharides present in the blood and internal tissues include glucose, fructose, and galactose. Many pentoses and hexoses can form ring structures. In these closed-chain forms, the aldehyde or ketone group remains non-free, so many of the reactions typical of these groups cannot occur. Glucose in solution exists mostly in the ring form at equilibrium, with less than 0.1% of the molecules in the open-chainform, Pigman, et al., (2002).

Biopolymers of sugars are common in nature. Through photosynthesis, plants produce glyceraldehyde-3-phosphate (G3P), a phosphated3-carbon sugar that is used by the cell to make monosaccharides such as glucose ($C_6H_{12}O_6$) or (as in cane and beet)

sucrose $(C_{12}H_{22}O_{11})$. Monosaccharides may be further converted into structural polysaccharides such as cellulose and pectin

for cell wall construction or into energy reserves in the form of storage polysaccharides such as starch or inulin. Starch, consisting of two different polymers of glucose, is a readily degradable form of chemical energy stored by cells, and can be converted to other types of energy, Pigman, et al., (2002).

Another polymer of glucose is cellulose, which is a linear chain composed of several hundred or thousand glucose units. It is used by plants as a structural component in their cell walls. Humans can digest cellulose only to a very limited extent, though ruminants can do so with the help of symbiotic bacteria in their gut, Joshi, S; Agte V. (2005). DNA and RNA are built up of the monosaccharides deoxyribose and ribose, respectively. Deoxyribose has the formula $C_5H_{10}O_4$ and ribose theformula $C_5H_{10}O_5$, Much Lude (2002).

Merck Index (2002).

The potential of lignocellulosic biomass as a renewable energy resource has been evaluated worldwide over many decades. Lignocellulosic sawdust is one of the major wastes resulting from wood exploitation and processing, which stored in uncontrolled conditions may be an important factor of environmental pollution. But at the same time is one of the main sources of biomass for the production of solid fuels for generating heat in both centralized system, in co-generation installations and in a decentralized system for residential use, in classic boilers for thermal energy generation, Zhang, et al., (2018)

Lignocelluloses refer to plant dry matter (biomass), so called lignocellulosic biomass. It is the most abundantly available raw material on the Earth for the production of biofuels. Concerns around greenhouse gas emissions necessitate the development of sustainable processes for the production of chemicals, materials, and fuels from alternative renewable sources, Yang, et al., (2016). The lignocellulosic plant cell walls are one of the most abundant sources of carbon for renewable bioenergy production, Yang, et al., (2016). Certain ionic liquids (ILs) are very effective at disrupting the plant cell walls of lignocellulose, and generate a substrate that is effectively hydrolyzed into fermentable sugars. Conventional ILs are relatively expensive in terms of purchase price, and the most effective imidazolium-based ILs also require energy intensive processing conditions (>140 °C, 3 h) to release >90 % fermentable sugar yields after saccharification, Yang, et al., (2016).

It is composed of two kinds of carbohydrate polymers, cellulose and hemicellulose, and an aromatic-rich polymer called lignin. Each has distinct chemical behaviour. The highly evolved resistance to degradation or even separation is referred to as recalcitrance. Overcoming this recalcitrance to produce useful, high value products requires a combination of heat, chemicals, enzymes, and microorganisms, Santos, et al., (2018).

These carbohydrate-containing polymers contain different sugar monomers (six and five carbon sugars) and they are covalently bonded to lignin. Lignocellulosic biomass can be broadly classified into virgin biomass, waste biomass, and energy crops. Virgin biomass includes all naturally occurring terrestrial plants such as trees, bushes and grass. Waste biomass is produced as a low value byproduct of various industrial sectors such as agriculture (corn stover, sugarcane bagasse, straw etc.) and forestry (saw mill and paper mill discards). Energy crops are crops with high yield of lignocellulosic biomass produced to serve as a raw material for production of second generation biofuel; include switch grass (*Panicum virgatum*) and Elephant_grass, Yang, et al., (2016)

Lignin is a heterogeneous, highly cross linked polymer akin to phenol-formaldehyde-resins. It is derived from 3-4 monomers, the ratio of which varies from species to species. The cross linking is highly extensive, being rich in aromatics; it is hydrophobic and relatively rigid. Lignin confers structural integrity to plants, Santos, et al., (2018). Lignin is so heterogeneous and so recalcitrant that its value is almost exclusively measured as a fuel. Hemicellulose is composed of branched polysaccharides. A particular problem is that hemicellulose is covalently linked to lignin, usually through Ferulic acid component of the lignin, Yoo, et al., (2017). Cellulose is a homopolymer of glucose. It is very poorly soluble in most solvents.

Lignocellulosic biomasses have been considered in the production of bio-composites materials such as particle panels, woodplastic composites, and cement/geopolymer wood composites. Even though the production of bio-composites material rely mostly on wood resources, in less forest-covered countries or in countries where wood resources are already being overused, it is possible to utilize alternative sources of biomass such as invasive plants, agricultural and sawmills residues for the creation of new "green" composites. Bio-composites produced with lignocellulosic biomasses as alternative to conventional materials, are attracting the attention because they are renewable and cheaper but also because they fit perfectly into the policy of the "cascade utilization" of the resources, Tarasov, et al., (2018).

Lignocellulose biomass derived from plant cell walls is a rich source of biopolymers, chemicals, and sugars, besides being a sustainable alternative to petrochemicals. A natural armor protecting living protoplasts, the cell wall is currently the target of intense study because of its crucial importance in plant development, morphogenesis, and resistance to a biotic stress. Beyond the intrinsic relevance related to the overall plant physiology, plant cell walls constitute an exquisite example of a natural composite material that is a constant source of inspiration for biotechnology, biofuel, and biomaterial industries, Tarasov, et al., (2018).

The environment is suffering from climate change, worsened by over-exploitation of resources thus increasing global greenhouse gas emission, Anderson et al., (2019); Hassan et al., (2019). Sustainable and environmentally friendly energy based on renewable resources are required in order to meet the world's future energy needs.

Lignocellulosic biomass (LB) continues to attract global interest as a sustainable alternative to fossil carbon resources to produce second-generation biofuels and other bio-based chemicals without compromising global food security, Chandel et al., (2018). These include agricultural wastes such as cereal straw, forest residues such as pine, and dedicated crops and short rotation coppices such as miscanthus, Lewandowski et al., (2000). LB is mainly composed of cellulose, hemicelluloses and lignin, making a complex assembly of polymers naturally recalcitrant to enzymatic conversion. That is why some pre-treatment steps are mandatory to make cellulose more accessible by changing the physical and/or the chemical structure of LB and facilitating the conversion of polysaccharides into fermentable sugars, Zhao et al., (2012), and Kumar, et al., (2009).

Factors affecting LB recalcitrance are strongly interconnected and difficult to dissociate, Zhao et al., (2012); Bichot et al., (2018). They can be divided into structural factors, which mainly refer to cellulose specific surface area, cellulose crystallinity, degree of polymerization, pore size and volume; chemical factors, related to composition and content in lignin, hemicelluloses and acetyl groups. Although many studies have investigated the impact of these factors on recalcitrance by examining different LB feed stocks and operating process conditions, conclusions obtained are not always obvious and even sometimes contradictory. The aim of this study was to produce sugar from lignocellulosic sawdust, and compare its nutritional composition with that of commercial granulated sugar for nutraceutical applications. Thereby, reduced environmental pollution and at the same time sourcing for cheaper natural raw materials with minima side effect for industrialization.

2.0 MATERIALS AND METHODOLOGY2.1 MATERIALS

2.1.1Apparatus

The following apparatus were used; blending machine, beakers, weighing balance, PH meter, two bottle, a tube, water bath, UV spectrophotometer, vortex mixer, glass ware, test tube and it's stand, ice test tube caps, tissue paper, wash bottle, pipette and marble caps.

2.1.2 Reagents

These were the reagents used; hydrochloric acid, distilled water, sodium hydroxide, lime solution, yeast, anthrone reagent, standard glucose, molish reagent and sulphuric acid.

2.1.3 Sample Collection

The lignocellulosic sawdust sample was collected from Oke-Gada Sawmill, Ede, in Ede North Local Government Area of Osun State by basket survey and it was taken to the laboratory for further treatment.

2.1.4 Sample treatment

7.5g of the sample (saw dust) was weighed and air dried. The drying continues until the moisture was completely removed, observed from the constant weight of the sample and then blended. **The sample was then** subjected to acid digestion for instrumental analysis.

2.1.5 Preparation of Analytical Reagents

- 18M Hydrochloric Acid (HCL): Scaling down both acid and distilled water by 10, then 146g of HCL diluted in 100ml of distilled water
- **6M Sodium Hydroxide (NaOH):** 240g of NaOH dissolved in 1000ml of distilled water
- IM Lime Solution: 74.1g of calcium hydroxide was poured in a volumetric flash, 1000ml of distilled water was added, it was shake vigorously for 2 minutes then left for 24 hours, it was carefully ensured not to stir up the sediment, and the clearer solution was poured off the top of flask through a clean filter paper.

Molisch's and Anthrone Reagents

- Molisch's reagents was 1% of α -Naphthol solution in 95% ethanol.
- Anthrone reagent was prepared by dissolve 2g of Anthrone in 1 litre of concentrated H₂SO₄.
 NB: Freshly prepared reagents were used for the assay
- Solution: 1000µg glucose per mL distilled water.

2.1.6 Production of Sugar

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The steps involved in the production of fermentable sugar from lignocellulosic sawdust were in two stages; the first stage was the acid hydrolysis of the blended sawdust, while the second stage was anaerobic fermentation of the acid hydrolyzed blended sawdust, after which it was crystallized.

- Acid Hydrolysis: Enough blended sawdust was poured into 400ml beaker to reach 100ml mark and weighed. 100ml of 18M hydrochloric acid was added and it was ensured that the acid cover the dust, 400ml of distilled water was poured inside 1000ml beaker; the mixture of acid and sawdust was poured into the distilled water in 1000ml of beaker. Then the acidity of the mixture was checked using PH meter. The PH was regularized between the range of 5.0-6.0 (distill water added was not more than 800ml), 6M of sodium hydroxide was then added with a dropper bottle until PH required was obtained.
- Fermentation: 250ml of hydrolyzed mixture was poured into a bottle with a cover, for anaerobic condition, then 250ml of 1M lime solution was poured into another bottle, a hole was created and a tube was passed into the hole, and other end of the tube was connected to the second bottle with hydrolyzed blended sawdust. 0.5g of yeast was added to 20ml of warm water and 2ml of the hydrolyzed mixture was shake together for 5minutes to activate the yeast and then it was poured into 250ml of hydrolyzed mixture inside the bottle, the whole mixture was kept for 4 days, after which it was filtered, the filtrate was the fermented sugar solution, which was later crystallized. The procedure was repeated for 1.0g, 3.0g, 5.0g, and 7.0g of yeast respectively.

Reactions involved

Reactions: HydrolysisMonosaccharide's + Disaccharide + MonosaccharideDehydrationEurfural Monosaccharide + FurfuralReaction of furfural with anthrone Furfural + Anthrone reagentBlue greencomplex

2.2 METHODOLOGY

2.2.1 Proximate Analysis

The following proximate parameters were investigated: Moisture Content, Ash Content, Crude Fibre, Crude Protein. Among others were; Crude Fat, and Carbohydrate Content.

2.2.2 Mineral Analysis

For the determination of the mineral contents of this sample, two stages were involved:

- > Sample Digestion
- Instrumental Analysis

The digested sample was then introduced into a well calibrated UV spectrophotometer for the reading. The final concentration was then calculated.

3.0 RESULTS AND DISCUSSION

3.1 Results

3.1.1 Proximate Analysis

Table 3.1 present the mean value of the result obtained from triplicate determination of proximate analysis of the samples. It was observed that to achieve unit increment in the proximate composition it requires more than 4g increase in the sample. The moisture content was observed to reduce significantly as the weight of the sugar increases. There was significant increase in the carbohydrate composition of the sample only when there was at least 4g increment in the weight, and same characteristics was

observed in crude protein as well. Other than these two, there was no significant increase in any other parameters measured, all other remain of constant proportion throughout the increase in weigh of the sample.

Samples	Moisture content	Crude fat	Fibre content	Crude Protein	Ash Content	Carbohydrate
А	41.47±0.21	0.001±0.01	0.99±0.02	6.25±0.00	0.21±0.01	51.07±0.19
В	39.57±0.23	0.02±0.00	0.98±0.01	7.29±0.36	0.22±0.01	51.92±0.33
С	38.60±0.52	0.02±0.0	0.68±0.58	7.29±0.36	0.23±0.01	53.19±034
D	3.11	0	0	0	1.01	95.58

Table 3.1: Proximate Analysis of the samples

KEY: A = 3g, B = 5g, C = 7g, and D = Commercial granulated sugar

There are parameters that vary alongside increment in weight of lignocellulosic sugar which included crude protein, carbohydrate proportion and moisture content. Moisture content was observed to decrease as the weight of sugar increase while Crude protein and Carbohydrate composition of the sugar had unit increase in 4g weight increment.

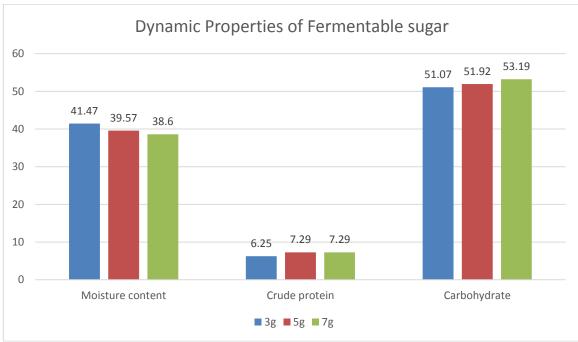


Figure 3.1: Dynamic properties of Lignocellulosic Sugar

3.1.2 Mineral Analysis

Mineral composition of the lignocellulosic sugar was presented in Table 3.2 and it revealed that there was very low concentration of micro element like Sodium, Manganese, Iron and Zinc, but rich in macro element like Potassium, Chromium, Phosphorus, and magnesium. There was statistically significant difference between the composition of Zn, Fe, Mg, P, Cr, Na and K as the sample increases in weight while such was not observed in Manganese.

Table 3.2. Mineral Composition of the Fermented sugar of different weights

Sa mpl es	Mn(ppm)	Zn(ppm)	Fe(ppm)	Mg(ppm)	P(ppm)	Cr(ppm)	Na(ppm)	K(ppm)
А	0.15±0.01	2.49±0.01	0.05±0.00	2.43±0.01	2.52±0.02	2.43±0.01	0.44±0.02	3.11±0.01
В	0.22±0.00	2.37±0.01	0.07±0.00	2.31±0.01	2.48±0.00	2.66±0.02	0.53±0.03	2.98±0.02
C	0.13±0.01	2.54±0.00	0.05±0.00	2.25±0.01	2.73±0.01	2.59±0.02	0.55±0.01	2.79±0.01
D	0	0	35	124.7	1104	0	112	5150

KEY: A = 3g, B = 5g, C = 7g, D = Commercial granulated sugar

3.2 DISCUSSION

The thought of eliminating lignocellulose biomass produce plant as sawdust was the beginning of eliminating both pollution and disposal problem that may come in with it in the industries.

3.2.1. Proximate Analysis

The proximate analysis shown that more than 50% of the samples were carbohydrates base as expected of sugar, but there was a significant difference between the sample's carbohydrate compositions which is in accordance with the work of Schmer, et al., (2008). Secondly, the moisture content was high; this makes the sugar to be liable in forming syrup in storage while standing. Lignocellulosic sugar had relatively higher proportion of crude protein, although there was significant increase in the proportion of crude protein present in the samples 3g and 7g. This was enough to undergo a Maillard reaction with any sugar including fructose in the fermented sugar and result in a decrease in protein quality which may be due to the loss of amino acid residues and decreased protein digestibility which is in accordance with the work of Harman-Ware, et al., (2017). Hence the concentration of protein available in the Lignocellulosic sugar was not enough to be considered a good source of protein even though it is present in a very low concentration. Although a very low proportion of the sample was observed as ash content this was an evidence of low mineral and fiber content, in accordance with the work of Finke, (2002).

3.2.2. Mineral Analysis

With a high concentration of macronutrients in the samples, there was optimum concentration of trace elements, as they had an average of less than 4ppm in Mn, Zn, Fe, Mg, P, Cr, Na and K. However, the concentration observed was considerably higher than that of the commercialized sugar in accordance with the work of Ademolu, et al., (2004). The concentration of Zinc (2.37ppm), Manganese (0.22ppm), and Iron (0.07 pm) were lower than the commercial sugar (Zn=11.33ppm, Mn=7.33ppm, Fe=3.33ppm,) in accordance with the work Banjo, et al., (2006). The presence of Zinc increases alongside with the weight of the sample, it was observed that the little concentration of zinc observed from this Lignocellulosic sugar will be of help in DNA creation, enzyme creation, Cell growth, building protein, and healing of damaged cells as it improves the healthy immune system. Another trace element with a very low concentration was Manganese which has an average of 0.22g in a 5g sample. The presence of manganese made the sugar beneficial in forming connective tissue, bones, blood-clotting factors, and sex hormones while it helps carbohydrate metabolism as well as blood sugar regulation.

From the same table above, it was observed that sodium and phosphate concentration of the two samples increases significantly (P<0.05) while Potassium centration in 5g of this sample (2.98 ± 0.02 ppm) was significantly higher than that of 5g of commercialized sugar (1.88ppm), Magnesium concentration in the 5g of fermented sugar (2.31 ± 0.01) was higher than that of commercial sugar (0.07ppm) in accordance with the work of Sales-Campos et al., (2009). It could be inferred that Lignocellulosic sugar was more nutritious in supplying of macronutrient as well as macro-element in accordance with the work of Banjo, et al., (2006).

The concentration of potassium was higher than that of commercial sugar and with this, it will help in maintaining normal level of fluids inside cells, and aid muscle contraction and support normal blood pressure.

4.0 CONCLUSION AND RECOMMENDATIONS

4.1. Conclusion

This research work showed that, the sugar produced was rich in carbohydrates, and crude protein, but containing a disadvantageous volume of moisture content. It was as very rich in macronutrients but not as much as found in commercial sample.

4.2. Recommendation

With the outcome of this work, the product could be recommended to be used in place of the imported commercial sugar cubes that compose mainly of carbohydrate. In addition, the acceptability of the fermented sugar in the society should also be investigated based on its organoleptic properties as further research work should emphasis on financial fitness of the production of sugar from lignocellulose biomass (saw dust).

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