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Saccharification of Different Delignified Sawdust Masses from Various Trees Along the Lagos Lagoon in Nigeria

N. A. Ndukwe*, J. B. M. Seeletse and J. P. H. van Wyk**†**

*Department of Chemical Sciences, College of Basic and Applied Sciences, Mountain Top University, Magoki, Ogun State, Nigeria

**Department of Pharmacology and Therapeutics, Sefako Makgatho Health Sciences University, South Africa

†Corresponding author: J. P. H. van Wyk: bioenergy.res@gmail.com

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ABSTRACT

Sawdust, a major waste product of the forestry industry, is accumulating along the Lagos Lagoon in Lagos, Nigeria, without it being effectively managed. Besides its use in manufacturing sound-absorbing boards to reinforce concrete beams and for energy purposes, its potential as a renewable energy source and feedstock for bio-product development has not yet been realized. Cellulose, a glucose biopolymer and structural component of cellulose can be hydrolyzed by a hydrolytic enzyme known as cellulase. During the process, the enzyme breaks the B-1,4-glucosidic bond, which keeps the glucose units together, and by acting on this bond, numerous glucose units are released. As part of sawdust, the cellulose molecule is not freely available for the degradation action of the cellulase enzyme as it is strongly associated with lignin, which acts as bio-glue, keeping cellulose and hemicellulose together. Delignification is an effective technique that was used to make the sawdust from ten different trees along the Lagos Lagoon in Nigeria more susceptible to saccharification by cellulase isolated from the fungus *Aspergillus niger*. Delignified and non-delignified sawdust masses between 2 mg and 10 mg were incubated with the *A. niger* cellulase solution (2 $mg.mL^{-1}$), whereafter, the amount of sugar produced by the cellulase action was determined. The percentage saccharification of each sawdust material was also linked with the amount of sugar produced during cellulase action. From these investigations was concluded that delignification increased sugar production when almost all the masses of different sawdust materials were degraded. It was also observed that the ratio of sawdust mass to enzyme concentration is an important variable that influences the effectiveness of the saccharification process. The percentage saccharification of the various sawdust materials was also determined, and it indicated that the highest percentage of saccharification was not obtained when the highest amount of sawdust was degraded, producing the highest amount of sugar. The saccharification of sawdust could contribute to the development of renewable energy sources and feedstock for bioproduct development. The process is, however, not that straightforward as variables such as the type of cellulase enzyme, pretreatment of the cellulose substrate, and optimizing of cellulase to cellulose ratio are a few that need to be optimized for the process to be effective in terms of glucose production.

INTRODUCTION

Environmental pollution is increasing at an alarming rate as a result of rapid population growth and industrialization with the production of solid waste, the most visualized form of waste. This type of waste is not only present in cities and towns because of human activities but is also a major pollutant in the forestry industry, where numerous volumes of sawdust are produced daily. Such is the case in Nigeria along the Lagos Lagoon, where thousands of trees are felled annually (Akhator et al. 2017). Simultaneously is the production of sawdust, which not only occupies land but has a negative effect on the water quality of the Lagos Lagoon and has also been identified as a major pollutant of air, causing a threat to the quality life of many people living along the Lagoon (Martin et al. 2020). Another concerning reality of these huge volumes of sawdust is its flammability, and due to the small particle size of this material, the spread of fire will be quick and unstoppable (Przybysz et al. 2023).

Also of environmental concern is the process of fossil fuel consumption not only as a source of energy but also as feedstock for the synthesis of many chemical-related

substances (Kapsalyamova & Paltsev 2020). The search for alternative and renewable energy resources that could also be developed as a feedstock for the biosynthesis of chemicals, biochemicals, and chemical substances with medicinal properties is topical, as many research efforts are focused on this approach (Mishra et al. 2021). Many cities are already using solid waste, especially organic parts such as kitchen waste, garden waste, and agriculture waste, as a resource to produce electricity by burning these materials, and the development of this type of waste as a resource for chemical synthetic procedures is well researched (Phiri et al. 2024). Biomass, including wood waste such as sawdust, has been identified as a renewable substance with a good potential to be developed as a resource for the synthesis of many bioproducts such as organic acids, polyhydroxyalkanoates, and bioplastics (Ashokkumar et al. 2022). The chemical substance in wood which makes sawdust attractive as a resource for bioproduct development is cellulose, a structural component, and glucose-based bio-polymer (Wan et al. 2010), also described as a complex chemical substance with the glucose units linked through B-1,4-glucosidic bonds (Andberg et al. 2015) to form chains which are interacting with each other through hydrogen bonds (Wang et al. 2023a).

When resolved into individual glucose units, the released glucose, which is a fermentable sugar, can be bio-converted into an environmentally friendly fuel known as bio-ethanol (Liu & Bao 2019), and the degradation of cellulose into glucose can be done employing acid or alkaline hydrolysis (Abeer et al. 2010). This process of degrading cellulose is not environmentally benign due to the negative effect of these acids and alkaline substances on the environment when regarded as a waste product after performing the hydrolysis process. Alternative to acidic and alkaline degradation of cellulose into glucose is a process that is catalyzed by cellulase, a multi-component enzyme system, and which is environmentally friendly (Wang et al. 2023b). This enzyme originates mostly from bacterial (Ali et al. 2013) and fungal (Yang et al. 2023) sources and has been described in the saccharification of many cellulose-containing substances such as bagasse (Hemansi & Saini 2023), wood rice straw (Pal et al. 2022) and wastepaper (Ndlovu & Van Wyk 2023). Besides the environmental benefit of using cellulase to degrade cellulose into fermentable sugars is the fact that the enzyme has components that can act specifically on the two structural sections of cellulose, namely the amorphous (Ciolacu et al. 2011) and crystalline (Cheng et al. 2011) sections which are essential for the effective degradation of cellulose. These sections show different susceptibilities towards the cellulase enzymes and need specific components of the cellulase complex to be degraded into sugars.

The saccharification of cellulose into fermentable sugars by the cellulase enzyme is a complicated process and many variables of the process need to be optimized for the process to be effective in terms of sugar production. A major stumbling block in the bioconversion process is the presence of lignin in the wood material. This bio-polymer acts as a biological glue, keeping cellulose and hemicellulose strongly together, and destroying this interaction will make cellulose more susceptible to cellulase-catalyzed degradation. Many variables regarding the saccharification of cellulose, such as cellulase concentration (Kaschuk et al. 2019), incubation temperature (Bhati et al. 2019), incubation pH (Bellaouchi et al. 2021), and product inhibition (Zou et al. 2021), could influence the outcome of this glucose producing process.

The current investigation reveals information regarding the *Aspergillus niger* cellulase-catalyzed saccharification of sawdust from different trees along the Lagos Lagoon in Nigeria. To increase the susceptibility of the cellulose to cellulase action, the various sawdust materials have been delignified with the Kraft process (Gustafson et al. 1983) as well as hydrogen peroxide treatment (Ndukwe et al. 2009). This investigation focused on the effect which different masses of the non-delignified and delignified sawdust materials will have on the saccharification thereof by the *A. niger* cellulase enzyme.

MATERIALS AND METHODS

Sawdust Substrate and Cellulase Enzyme

Non-delignified and delignified sawdust samples from ten different trees were transferred in triplicate into test tubes. Names of these sawdust samples are *Erythropleum suaveolens, Symphona globulifera, Ricindendron heudelotii, Pterygota macrocarpa, Milicia excels, Ipomoeu asarifolia, Hallelea ciliate, Sacoglottis gabonensis, Pycnanthus angolensis*, and *Terminalia superb*. Commercially obtained *Aspergillus niger* cellulase enzyme (0.1g) was dissolved in 0.005 mol.dm^{-3} pH 5.0 tris buffer resulting in an enzyme solution concentration of 2.0 mg.mL^{-1}.

Delignification of Sawdust- Kraft Pulping and Hydrogen Peroxide Treatment of the Wood Sawdust

To ensure a maximum cellulose exposure to the cellulase enzyme, the various sawdust materials were delignified by subjecting 2kg of each of the different sawdust materials (2.8-5.0 mm particle size) to 350g of NaOH and 140g NaS_2 during the Kraft pulping process. The Kraft pulping chemicals were dissolved in 8 L water. The delignification of the lignocellulosic materials (sawdust) was carried out in a rotary steel digester at 170°C and a pressure of 200 kPa for

1 h 45 min at cooking liquor to the wood ratio of 4:1. After the Kraft pretreatment, the extracted cellulose fibers were washed in turns with deionized water until they were free of the Kraft reagents (Ndukwe et al. 2009).

To remove residual lignin from these Kraft-treated cellulose, all these sawdust materials (10 g) were treated with 30% hydrogen peroxide (60 mL) at 40°C for 25-30 min.

Cellulase Incubation and Sugar Analyses

The delignified and non-delignified sawdust materials (2 mg, 4 mg, 6 mg, 8 mg, and 10 mg) were transferred in triplicate in test tubes and incubated with the *A. niger* cellulase enzyme solution (200 uL) and Tris buffer solution pH 5.0 (800 uL) for 2h at an incubation temperature of 50°C. The concentration of sugars released from the sawdust materials during cellulase-catalyzed degradation was determined from a standard glucose calibration curve constructed with glucose standard solutions at concentrations of 0.50 $mg.mL^{-1}$, 2.00 $mg.mL^{-1}$, 4.00 $mg.mL^{-1}$, 6.00 $mg.mL^{-1}$ and 8.00 $mg.mL^{-1}$. The DNS method, as described by Miller (1959), was used to calculate the concentration of the sugar produced during *A. niger* action on the waste sawdust.

Calculation of Resultant Amount of Sugar Produced and Percentage Saccharification

The resultant amount of sugar produced from the delignified and non-delignified sawdust was calculated by subtracting the amount of sugar released from each type of sawdust in the absence of cellulase action from the amount of sugar released when the sawdust was treated with the cellulase enzyme. This amount of sugar, known as the resultant amount of sugar, was released because of the cellulase action on each type of sawdust material.

The percentage saccharification of each sawdust material was calculated by dividing the resultant mass of sugars produced through cellulase action by the total mass of the sawdust incubated multiplied by a hundred. These values indicate to what extent the sawdust was bioconverted into sugars and can also be used to conclude the relative saccharification of the various sawdust materials.

Statistical Analysis

All the experimental analyses were performed in triplicate, and the mean values with standard deviations were determined with Microsoft Excel.

RESULTS AND DISCUSSION

The physical interaction between an enzyme and a substrate is an important prerequisite for the action of the bio-catalyst in converting the substrates into products. In the case of cellulase technology, the cellulase enzyme attacks a cellulose molecule, and during the interaction, the reaction product, glucose, is produced. During the cellulolytic action, the cellulase enzyme performs an adsorption-desorption action on the cellulose materials while degrading cellulose into either shorter cellulose chains or in glucose (Wang et al. 2020). One of the factors which control the rate and magnitude of cellulose hydrolysis is the availability and accessibility of cellulose to the cellulase enzyme which will determine the adsorption of the enzyme onto the cellulose surface. The interaction and binding of cellulase to cellulose is a mandatory step for cellulose degradation (Kim et al. 2005). The three-dimensional structure, size, and shape of the substrate determine the enzyme accessibility to the substrate, which has more ordered internal and less ordered external surfaces (Chang et al. 1981). The adsorption has also been attributed to other features including the irreversibility of the cellulase adsorption (Palonen et al. 1999), interaction among the adsorbing components of enzyme in high concentrations (Jeon et al. 2002), multiple adsorbing sites for a single cellulase molecule (Linder & Teeri 1997, Carrard & Linder 1999), enzymes being entrapped by cellulose pores (Lee et al. 1988) and adsorption of multiple components having different constants (Beldman et al. 1987).

Described above are all variables that could affect the outcome of the cellulase-catalyzed saccharification reaction of cellulose, and during this investigation, the mass of various insoluble sawdust samples was increased while treated with a fixed amount (concentration) of the *A. niger* cellulase enzyme. Delignified as well as non-delignified sawdust samples with masses varied between 2 mg to 10 mg were treated with the cellulase enzyme, and the amount of sugar produced from each material, as well as the percentage saccharification of each sawdust material, were determined. The sugar-producing and saccharification profiles are illustrated on various graphs and illustrated the increase in the sugar concentration produced from the delignified sawdust and the non-delignified sawdust when degraded by the cellulase enzyme.

The sugar production profiles, as well as percentage saccharification and the resultant amount of sugar produced from the different non-delignified and delignified sawdust samples from *E. suaveolens,* are represented in Fig. 1. The bioconversion of non-delignified sawdust from *E. suaveolens* by *A. niger* cellulase resulted in an increased amount of sugar produced that varied between a concentration of 0.78 $mg.mL^{-1}$ when 2 mg of the sawdust was degraded to a concentration of 1.12 $mg.mL^{-1}$ during the degradation of the highest masses of 8 mg and 10 mg. These results showed

a 143% increase in sugar production from the lowest mass of sawdust to the highest mass of sawdust at an increasing rate of 0.034 mg sugar produced for a 1 mg increase in sawdust degraded. When the delignified sawdust was degraded, the relative amount of sugar produced from all the masses exposed to the cellulase enzyme was higher than the sugar produced from the corresponding non-delignified sawdust masses. The sugar concentration produced from delignified sawdust increased from 1.16 $mg.mL^{-1}$ when 2 mg was degraded to a concentration of 2.26 $mg.mL^{-1}$ during degradation of the highest mass of 10 mg. This degradation pattern shows an increase of 195% in sugar formation from the lowest mass to the highest mass bioconverted by the cellulase enzyme.

The difference in sugar production between the non-delignified and delignified sawdust materials at different masses showed an increase in difference, which varied from 0.38 $mg.mL^{-1}$ when 2 mg of the material was degraded to 1.14 $mg.mL^{-1}$ when the highest mass of 10 mg was degraded. The rate of sugar production when the delignified sawdust was degraded was calculated at 0.11 mg sugar produced for an increase of 1 mg delignified sawdust degraded. The general trend of saccharification for both the non-delignified as well as delignified sawdust showed a decrease in percentage saccharification as the mass degraded is increased. In the case of the non-delignified sawdust, the percentage of saccharification decreased from 39% when the lowest mass was degraded to 12% when the highest mass of 10 mg was degraded. The degradation of the delignified sawdust resulted in a 58% degradation when the lowest mass of 2 mg was saccharified. At the same time, a 22% saccharification was obtained when the highest mass of 10 mg was bioconverted into sugars.

The sugar production profiles, as well as percentage saccharification and the resultant amount of sugar produced from the different non-delignified and delignified sawdust samples from *S. globulifera*, are represented in Fig. 2. The bioconversion of non-delignified sawdust from *S. globulifera* by *A. niger* cellulase resulted in an increased amount of sugar produced that varied between a concentration of 1.21 $mg.mL^{-1}$ when 2 mg of the sawdust was degraded to a concentration of 1.95 $mg.mL^{-1}$ during the degradation of the highest masses of 10 mg. These results showed a 161% increase in sugar production from the lowest mass of sawdust to the highest mass of sawdust at an increasing rate of 0.074 mg sugar produced for a 1 mg increase in sawdust degraded. When the delignified sawdust was degraded, the relative amount of sugar produced from all the masses exposed to the cellulase enzyme was higher than the sugar produced from the corresponding non-delignified sawdust masses. The sugar concentration produced from delignified sawdust increased from 1.21 $mg.mL^{-1}$ when 2 mg was degraded to a concentration of 2.18 $mg.mL^{-1}$ during degradation of the highest mass of 10 mg. This degradation pattern shows an increase of 180% in sugar formation from the lowest mass to the highest mass bioconverted by the cellulase enzyme.

The difference in sugar production between the non-delignified and delignified sawdust materials at different

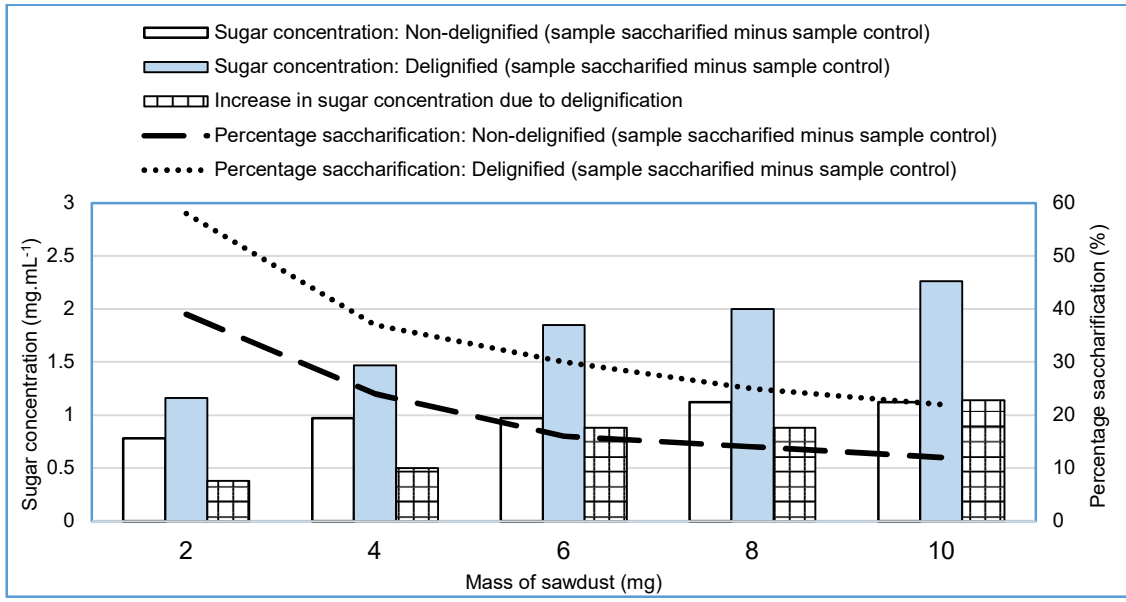

Fig. 1: *A. niger* cellulase catalyzed degradation of delignified as well as non-delignified sawdust from *Erythropleum suaveolens*.

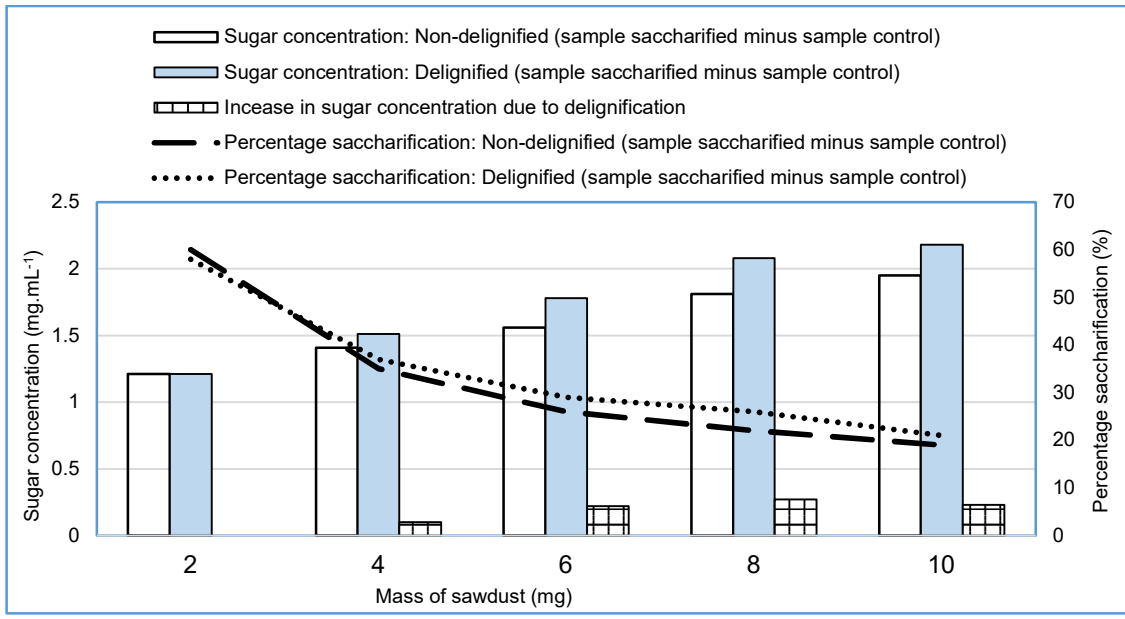

Fig. 2: *A. niger* cellulase catalyzed degradation of delignified as well as non-delignified sawdust from *Symphonia globulifera*.

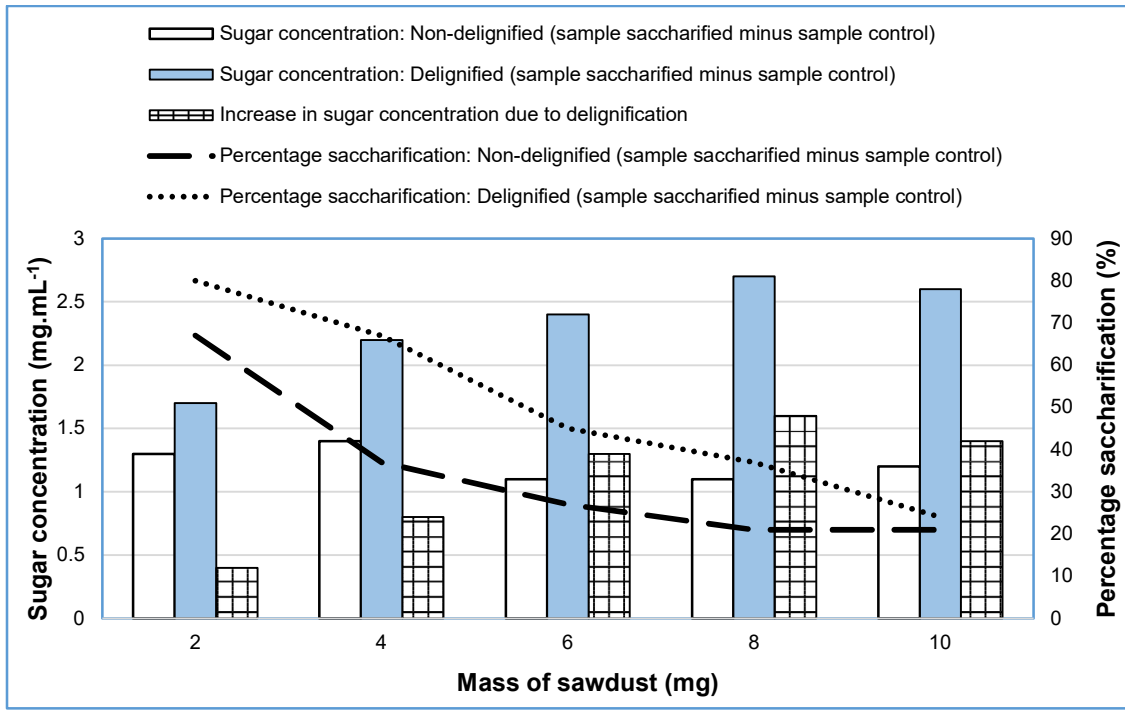

Fig. 3: *A. niger* cellulase catalyzed degradation of delignified as well as non-delignified sawdust from *Ricindendron heudelotti*.

masses showed an increase in difference, which varied from 0.00 $mg.mL^{-1}$ when 2 mg of the material was degraded to 0.23 $mg.mL^{-1}$ when the highest mass of 10 mg was degraded. The rate of sugar production when the delignified sawdust was degraded was calculated at 0.097 mg sugar produced for an increase of 1 mg delignified sawdust degraded. The general trend of saccharification for both the non-delignified as well as delignified sawdust showed a decrease in percentage saccharification as the mass degraded is increased. In the case of the non-delignified sawdust, the

percentage of saccharification decreased from 60% when the lowest mass was degraded to 19% when the highest mass of 10 mg was degraded. The degradation of the delignified sawdust resulted in a 58% degradation when the lowest mass of 2 mg was saccharified, while a 21% saccharification was obtained when the highest mass of 10 mg was bioconverted into sugars.

The sugar production profiles, as well as percentage saccharification and the resultant amount of sugar produced from the different non-delignified and delignified sawdust samples from *R. heudelotti,* are represented in Fig. 3. The bioconversion of non-delignified sawdust from *R. heudelotti* by *A. niger* cellulase resulted in an amount of sugar produced that varied between a concentration of 1.1 mg.mL^{-1} and 1.4 mg.mL^{-1} during the degradation of the masses starting from 2 mg and 10 mg. These results showed a 161% increase in sugar production from the lowest mass of sawdust to the highest mass of sawdust at an increasing rate of 0.074 mg sugar produced for a 1 mg increase in sawdust degraded. When the delignified sawdust was degraded, the relative amount of sugar produced from all the masses exposed to the cellulase enzyme was higher than the sugar produced from the corresponding non-delignified sawdust masses. The sugar concentration produced from delignified sawdust increased from 1.7 mg.mL^{-1} when 2 mg was degraded to a concentration of 2.7 mg.mL^{-1} and 2.6 mg.mL^{-1} during degradation of the highest mass of 8 mg and 10 mg. This degradation pattern shows an increase of 158% in sugar formation from the lowest mass to the highest mass bioconverted by the cellulase enzyme.

The difference in sugar production between the non-delignified and delignified sawdust materials at different masses showed an increase in difference, which varied from 0.4 mg.mL^{-1} when 2 mg of the material was degraded to 1.4 mg.mL^{-1} when the highest mass of 10 mg was degraded. The rate of sugar production when the delignified sawdust was degraded was calculated at 0.1 mg sugar produced for an increase of 1 mg delignified sawdust degraded. The general trend of saccharification for both the non-delignified as well as delignified sawdust showed a decrease in percentage saccharification as the mass degraded is increased. In the case of the non-delignified sawdust, the percentage of saccharification decreased from 80% when the lowest mass was degraded to 24% when the highest mass of 10 mg was degraded. The degradation of the delignified sawdust resulted in a 67% degradation when the lowest mass of 2 mg was saccharified. At the same time, a 21% saccharification was obtained when the highest mass of 10 mg was bioconverted into sugars.

The sugar production profiles, as well as percentage saccharification and the resultant amount of sugar produced from the different non-delignified and delignified sawdust samples from *P. macrocarpa,* are represented in Fig. 4. The bioconversion of non-delignified sawdust from *P. macrocarpa* by *A. niger* cellulase resulted in an increased amount of sugar produced that varied between a concentration

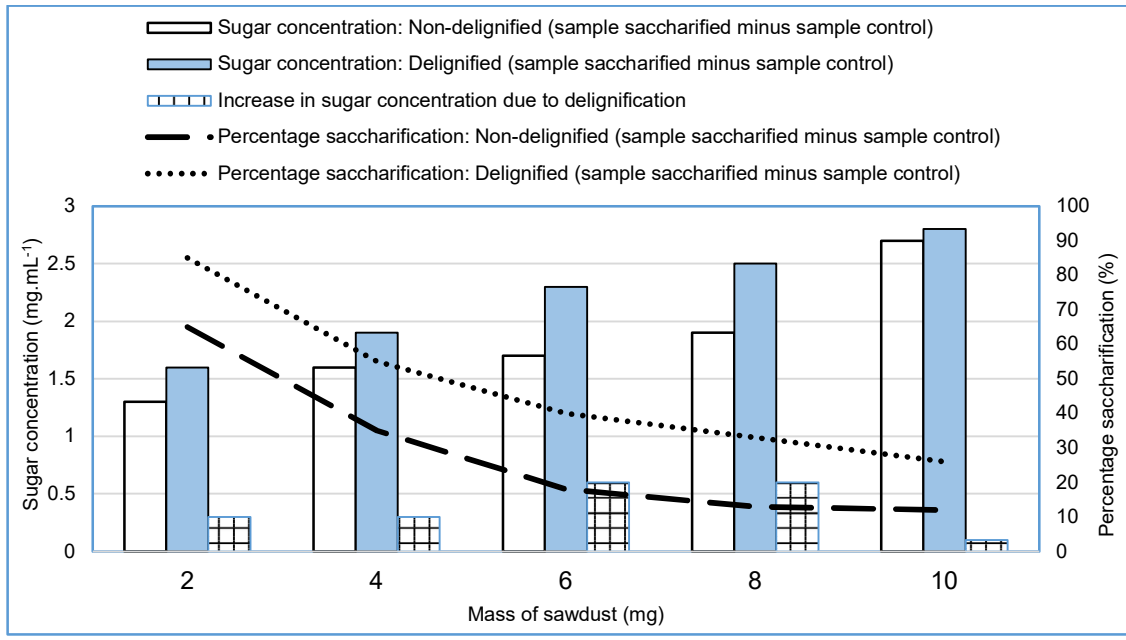

Fig. 4: *A. niger* cellulase catalyzed degradation of delignified as well as non-delignified sawdust from *Pterygota macrocarpa*.

of 1.3 mg.mL^{-1} when 2 mg of the sawdust was degraded to a concentration of 2.7 mg.mL^{-1} during the degradation of the highest masses of 10 mg. These results showed a 140% increase in sugar production from the lowest mass of sawdust to the highest mass of sawdust at an increasing rate of 0.14 mg sugar produces for a 1 mg increase in sawdust degraded. When the delignified sawdust was degraded, the relative amount of sugar produced from all the masses exposed to the cellulase enzyme was higher than the sugar produced from the corresponding non-delignified sawdust masses. The sugar concentration produced from delignified sawdust increased from 1.6 mg.mL^{-1} when 2 mg was degraded to a concentration of 2.8 mg.mL^{-1} during degradation of the highest mass of 10 mg. This degradation pattern shows an increase of 175% in sugar formation from the lowest mass to the highest mass bioconverted by the cellulase enzyme.

The difference in sugar production between the non-delignified and delignified sawdust materials at different masses showed an increase in difference, which varied from 0.3 mg.mL^{-1} when 2 mg of the material was degraded to 0.1 mg.mL^{-1} when the highest mass of 10 mg was degraded. The rate of sugar production when the delignified sawdust was degraded was calculated at 0.12 mg sugar produced for an increase of 1 mg delignified sawdust degraded. The general trend of saccharification for both the non-delignified as well as delignified sawdust showed a decrease in percentage saccharification as the mass degraded is increased. In the case of the non-delignified sawdust, the percentage of saccharification decreased from 65% when the lowest mass was degraded to 12% when the highest mass of 10 mg was degraded. The degradation of the delignified sawdust resulted in an 85% degradation when the lowest mass of 2 mg was saccharified, while a 26% saccharification was obtained when the highest mass of 10 mg was bioconverted into sugars.

The sugar production profiles, as well as percentage saccharification and the resultant amount of sugar produced from the different non-delignified and delignified sawdust samples from *M. Excels,* are represented in Fig. 5. The bioconversion of non-delignified sawdust from *M. excels* by *A. niger* cellulase resulted in an increased amount of sugar produced that varied between a concentration of 1.34 mg.mL^{-1} when 2 mg of the sawdust was degraded to a concentration of 2.17 mg.mL^{-1} during the degradation of

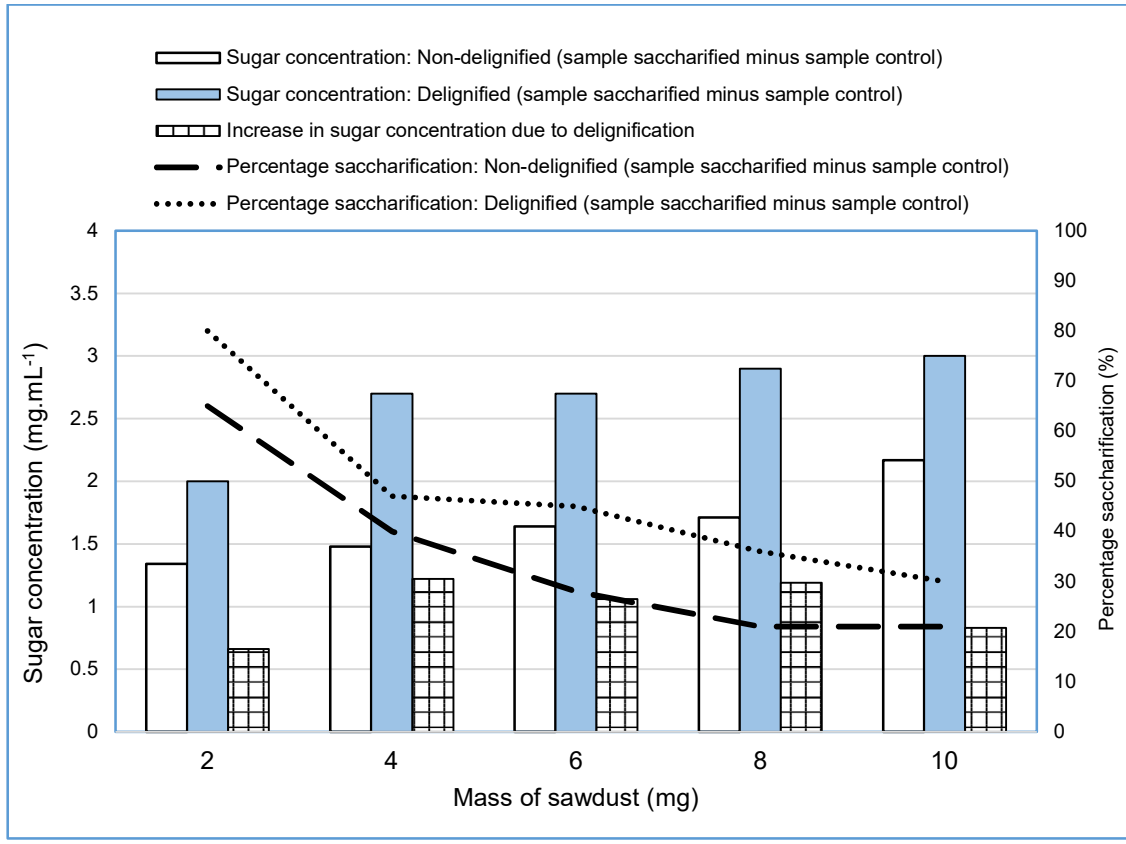

Fig. 5: *A. niger* cellulase catalyzed degradation of delignified as well as non-delignified sawdust from *Milicia excels*.

the highest masses of 10 mg. These results showed a 161% increase in sugar production from the lowest mass of sawdust to the highest mass of sawdust at an increasing rate of 0.083 mg sugar produced for a 1 mg increase in sawdust degraded. When the delignified sawdust was degraded, the relative amount of sugar produced from all the masses exposed to the cellulase enzyme was higher than the sugar produced from the corresponding non-delignified sawdust masses. The sugar concentration produced from delignified sawdust increased from 1.16 $mg.mL^{-1}$ when 2 mg was degraded to a concentration of 2.26 $mg.mL^{-1}$ during degradation of the highest mass of 10 mg. This degradation pattern shows an increase of 195% in sugar formation from the lowest mass to the highest mass bioconverted by the cellulase enzyme.

The difference in sugar production between the non-delignified and delignified sawdust materials at different masses showed an increase in difference, which varied from 0.66 $mg.mL^{-1}$ when 2 mg of the material was degraded to 1.22 $mg.mL^{-1}$ when the mass of 4 mg was degraded. The rate of sugar production when the delignified sawdust was degraded was calculated at 0.1 mg sugar produced for an increase of 1 mg delignified sawdust degraded. The general trend of saccharification for both the non-delignified as well as delignified sawdust showed a decrease in percentage saccharification as the mass degraded is increased. In the case of the non-delignified sawdust, the percentage of saccharification decreased from 65% when the lowest mass was degraded to 21% when the highest mass of 10 mg was degraded. The degradation of the delignified sawdust resulted in 80% degradation when the lowest mass of 2 mg was saccharified, while a 30% saccharification was obtained when the highest mass of 10 mg was bioconverted into sugars.

The sugar production profiles, as well as percentage saccharification and resultant amount of sugar produced from the different non-delignified and delignified sawdust samples from *I. asarifolia*, are represented in Fig. 6. The bioconversion of non-delignified sawdust from *I. asarifolia* by *A. niger* cellulase resulted in an increased amount of sugar produced that varied between a concentration of 0.85 mgmL when 2 mg of the sawdust was degraded to a concentration of 1.49 $mg.mL^{-1}$ during the degradation of the highest masses of 10 mg. These results showed a 175% increase in sugar production from the lowest mass of sawdust to the highest mass of sawdust at an increasing rate of 0.064 mg sugar produced for a 1 mg increase in sawdust degraded. When the delignified sawdust was degraded, the relative amount of sugar produced from all the masses exposed to the cellulase enzyme was higher than the sugar produced from the corresponding non-delignified sawdust masses. The sugar concentration produced from delignified sawdust increased from 0.83 $mg.mL^{-1}$ when 2 mg was degraded to a concentration of 1.75 $mg.mL^{-1}$ during degradation of the highest mass of 10 mg. This degradation pattern shows an increase of 210% sugar formation from the lowest mass to the highest mass bioconverted by the cellulase enzyme.

The difference in sugar production between the non-delignified and delignified sawdust materials at different masses showed an increase in difference, which varied from 0.67 $mg.mL^{-1}$ when 2 mg of the material was degraded to 0.35 $mg.mL^{-1}$ when the highest mass of 10 mg was degraded. The rate of sugar production when the delignified sawdust

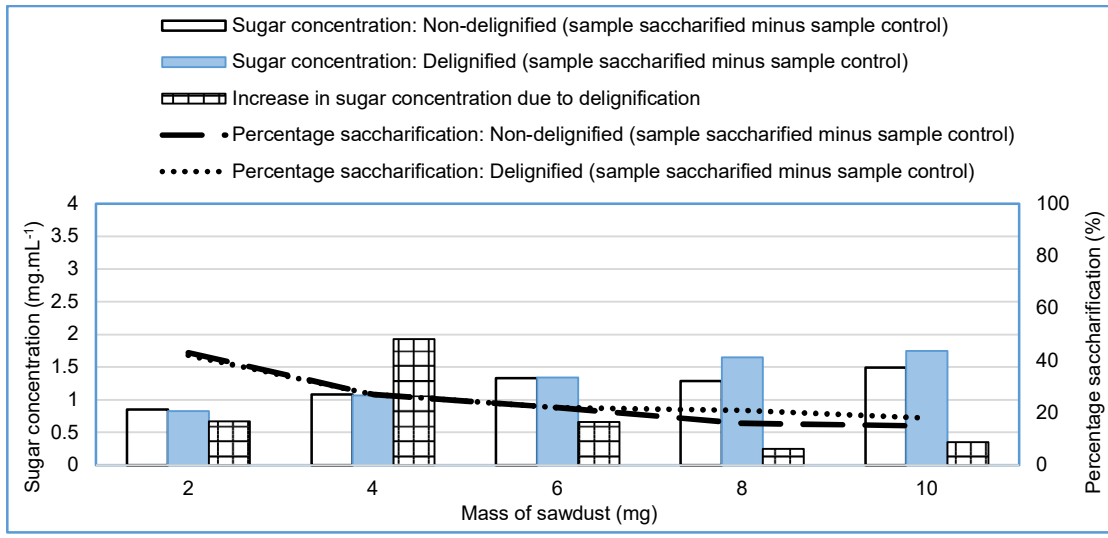

Fig. 6: *A. niger* cellulase catalyzed degradation of delignified as well as non-delignified sawdust from *Ipomoea asarifolia*.

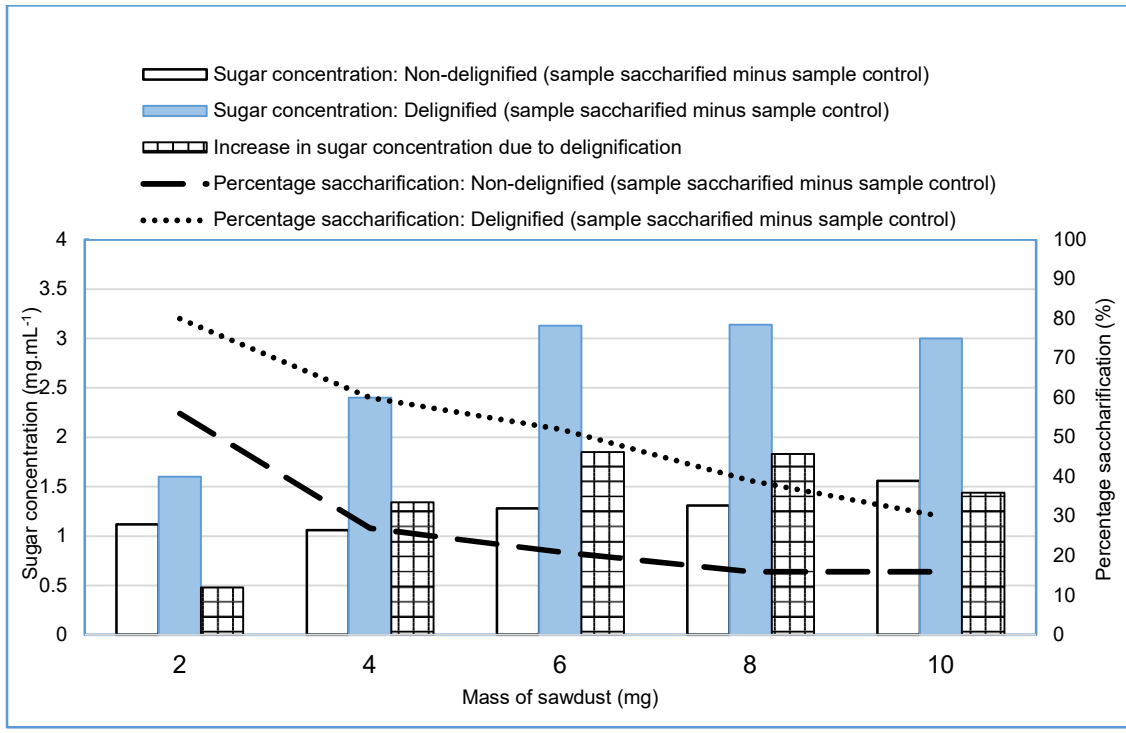

Fig. 7: *A. niger* cellulase catalyzed degradation of delignified as well as non-delignified sawdust from *Hallelea ciliate*.

was degraded was calculated at 0.092 mg sugar produced for an increase of 1 mg delignified sawdust degraded. The general trend of saccharification for both the non-delignified as well as delignified sawdust showed a decrease in percentage saccharification as the mass degraded is increased. In the case of the non-delignified sawdust, the percentage of saccharification decreased from 43% when the lowest mass was degraded to 15% when the highest mass of 10 mg was degraded. The degradation of the delignified sawdust resulted in 42% degradation when the lowest mass of 2 mg was saccharified, while an 18% saccharification was obtained when the highest mass of 10 mg was bioconverted into sugars.

The sugar production profiles, as well as percentage saccharification and the resultant amount of sugar produced from the different non-delignified and delignified sawdust samples from *H. ciliate*, are represented in Fig. 7. The bioconversion of non-delignified sawdust from *H. ciliate* by *A. niger* cellulase resulted in an increased amount of sugar produced that varied between a concentration of 1.12 $mg.mL^{-1}$ when 2 mg of the sawdust was degraded to a concentration of 1.56 $mg.mL^{-1}$ during the degradation of the highest mass of 10 mg. These results showed a 139% increase in sugar production from the lowest mass of sawdust to the highest mass of sawdust at an increasing rate of 0.044 mg sugar produced for a 1 mg increase in sawdust degraded. When the delignified sawdust was degraded, the relative amount of sugar produced from all the masses exposed to the cellulase enzyme was higher than the sugar produced from the corresponding non-delignified sawdust masses. The sugar concentration produced from delignified sawdust increased from 1.6 $mg.mL^{-1}$ when 2 mg was degraded to a concentration of 3.13 $mg.mL^{-1}$, 3.14 $mg.mL^{-1}$, and 3.0 $mg.mL^{-1}$ during degradation of the high masses of 6 mg, 8 mg, and 10 mg. This degradation pattern shows an increase of 196% in sugar formation from the lowest mass to the highest mass bioconverted by the cellulase enzyme.

The difference in sugar production between the non-delignified and delignified sawdust materials at different masses showed an increase in difference, which varied from 0.48 $mg.mL^{-1}$ when 2 mg of the material was degraded to 1.44 $mg.mL^{-1}$ when the highest mass of 10 mg was degraded. The rate of sugar production when the delignified sawdust was degraded was calculated at 0.154 mg of sugar produced for an increase of 1 mg delignified sawdust degraded. The general trend of saccharification for both the non-delignified as well as delignified sawdust showed a decrease in percentage saccharification as the mass degraded is increased. In the case of the non-delignified sawdust, the percentage of saccharification decreased from

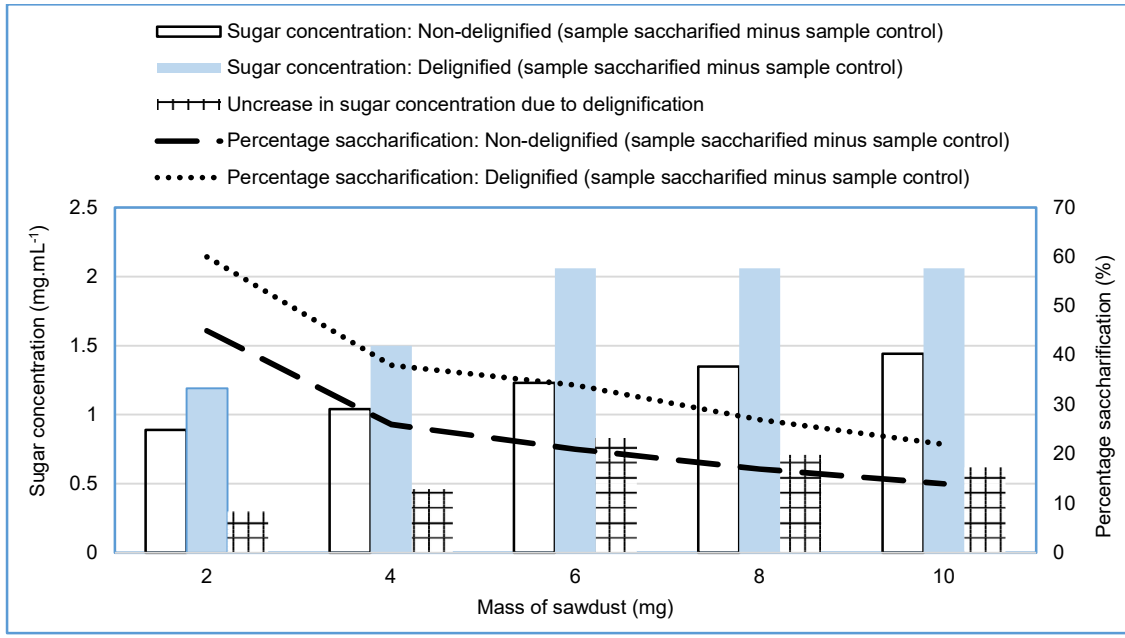

Fig. 8: *A. niger* cellulase catalyzed degradation of delignified as well as non-delignified sawdust from *Sacoglottis gabonensis*.

56% when the lowest mass was degraded to 16% when the highest mass of 10 mg was degraded. The degradation of the delignified sawdust resulted in an 80% degradation when the lowest mass of 2 mg was saccharified, while a 30% saccharification was obtained when the highest mass of 10 mg was bioconverted into sugars.

The sugar production profiles, as well as percentage saccharification and resultant amount of sugar produced from the different non-delignified and delignified sawdust samples from *S. gabonensis*, are represented in Fig. 8. The bioconversion of non-delignified sawdust from *S. gabonensis* by *A. niger* cellulase resulted in an increased amount of sugar produced that varied between a concentration of 0.89 mg.mL^{-1} when 2 mg of the sawdust was degraded to a concentration of 1.44 mg.mL^{-1} during the degradation of the highest masses of 10 mg. These results showed a 161% increase in sugar production from the lowest mass of sawdust to the highest mass of sawdust at an increasing rate of 0.055 mg sugar produced for a 1 mg increase in sawdust degraded. When the delignified sawdust was degraded, the relative amount of sugar produced from all the masses exposed to the cellulase enzyme was higher than the sugar produced from the corresponding non-delignified sawdust masses. The sugar concentration produced from delignified sawdust increased from 1.19 mg.mL^{-1} when 2 mg was degraded to a concentration of 2.06 mg.mL^{-1} during degradation of the masses 6 mg, 8 mg, and 10 mg. This degradation pattern shows an increase of 231% in sugar formation from the lowest mass to the highest mass bioconverted by the cellulase enzyme.

The difference in sugar production between the non-delignified and delignified sawdust materials at different masses showed an increase, varying from 0.3 mg.mL^{-1} when 2 mg of the material was degraded to 0.62 mg/mL when the highest mass of 10 mg was degraded. The rate of sugar production for the delignified sawdust was calculated at 0.087 mg of sugar produced per 1 mg increase in degraded delignified sawdust. The general trend of saccharification for both non-delignified and delignified sawdust showed a decrease in percentage saccharification as the mass degradation increased. For the non-delignified sawdust, the percentage of saccharification decreased from 45% at the lowest mass to 14% at the highest mass of 10 mg. The degradation of the delignified sawdust resulted in 60% saccharification at the lowest mass of 2 mg, while 22% saccharification was obtained at the highest mass of 10 mg.

The sugar production profile, as well as percentage saccharification and resultant amount of sugar produced from the different non-delignified and delignified sawdust samples from *P. angolensis*, are represented in Fig. 9. The bioconversion of non-delignified sawdust from *P. angolensis* by *A. niger* cellulase resulted in an increased amount of sugar produced that varied between a concentration of 0.52 mg.mL^{-1} when 2 mg of the sawdust was degraded to a concentration of 1.15 mg.mL^{-1} during the degradation of the highest masses of 10 mg. These results showed a 221%

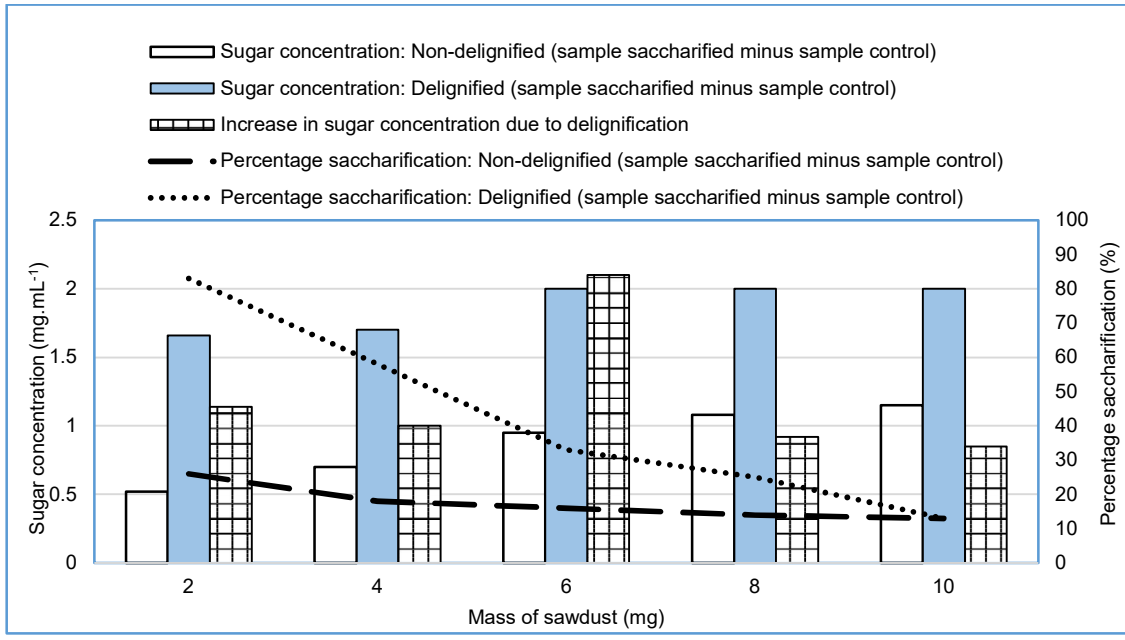

Fig. 9: *A. niger* cellulase catalyzed degradation of delignified as well as non-delignified sawdust from *Pycnanthus angolensis*.

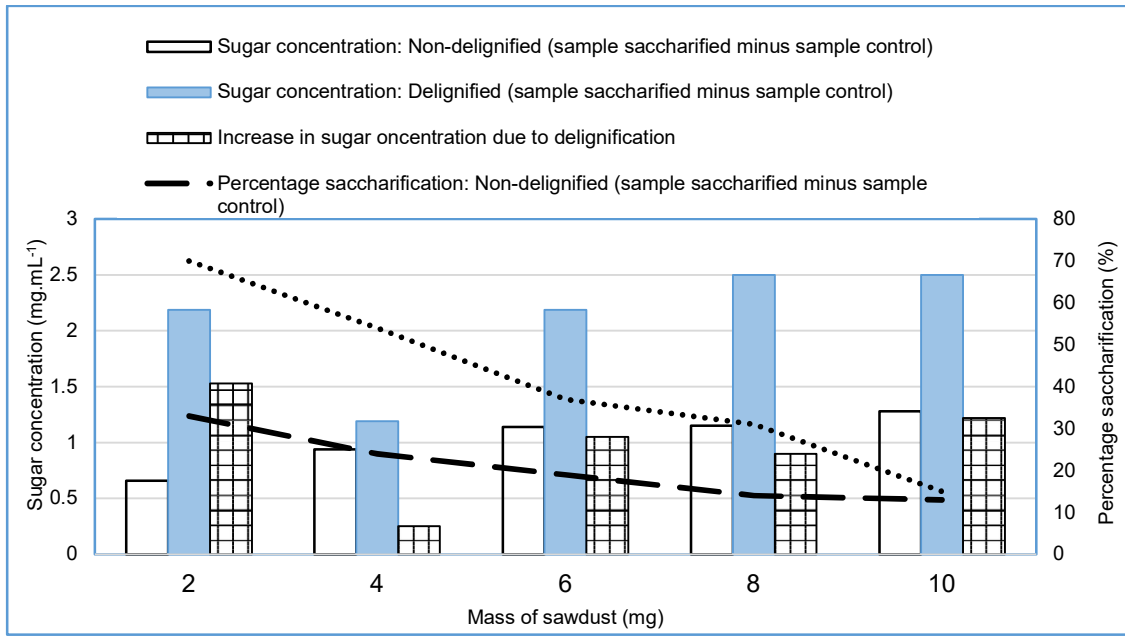

Fig. 10: *A. niger* cellulase catalyzed degradation of delignified as well as non-delignified sawdust from *Terminalia superb*.

increase in sugar production from the lowest mass of sawdust to the highest mass of sawdust at an increasing rate of 0.063 mg sugar produced for a 1 mg increase in sawdust degraded. When the delignified sawdust was degraded, the relative amount of sugar produced from all the masses exposed to the cellulase enzyme was higher than the sugar produced from the corresponding non-delignified sawdust masses. The sugar concentration produced from delignified sawdust increased from 1.66 $mg.mL^{-1}$ when 2 mg was degraded to a concentration of 2.0 $mg.mL^{-1}$ during degradation of the masses of 6 mg, 8 mg, and 10 mg. This degradation pattern shows an increase of 120% in sugar formation from

the lowest mass to the highest mass bioconverted by the cellulase enzyme.

The rate of sugar production when the delignified sawdust was degraded was calculated at 0.034 mg sugar produced for an increase of 1 mg delignified sawdust degraded. The general trend of saccharification for both the non-delignified as well as delignified sawdust showed a decrease in percentage saccharification as the mass degraded is increased. In the case of the non-delignified sawdust, the percentage of saccharification decreased from 26% when the lowest mass was degraded to 13% when the highest mass of 10 mg was degraded. The degradation of the delignified sawdust resulted in an 83% degradation when the lowest mass of 2 mg was saccharified, while a 13% saccharification was obtained when the highest mass of 10 mg was bioconverted into sugars.

The sugar production profiles, as well as percentage saccharification and the resultant amount of sugar produced from the different non-delignified and delignified sawdust samples from *T. superb,* are represented in Fig. 10. The bioconversion of non-delignified sawdust from *T. superb* in *A. niger* cellulase resulted in an increased amount of sugar produced that varied between a concentration of 0.66 $mg.mL^{-1}$ when 2 mg of the sawdust was degraded to a concentration of 1.28 $mg.mL^{-1}$ during the degradation of the highest masses of 10 mg. These results showed a 193% increase in sugar production from the lowest mass of sawdust to the highest mass of sawdust at an increasing rate of 0.062 mg sugar produced for a 1 mg increase in sawdust degraded. When the delignified sawdust was degraded, the relative amount of sugar produced from all the masses exposed to the cellulase enzyme was higher than that produced from the corresponding non-delignified sawdust masses. The sugar concentration produced from delignified sawdust increased from 2.19 $mg.mL^{-1}$ when 2 mg was degraded to 2.5 mg/mL during the degradation of the highest mass of 10 mg. This degradation pattern shows an 86% increase in sugar formation from the lowest mass to the highest mass bioconverted by the cellulase enzyme.

The difference in sugar production between the non-delignified and delignified sawdust materials at different masses showed an increase in variance, ranging from 1.53 $mg.mL^{-1}$ when 2 mg of the material was degraded to 1.22 $mg.mL^{-1}$ when the highest mass of 10 mg was degraded. The rate of sugar production for the delignified sawdust was calculated at 0.031 mg sugar produced per 1 mg increase in degraded delignified sawdust. The general trend of saccharification for both non-delignified and delignified sawdust showed a decrease in percentage saccharification as the mass degradation increased. For the non-delignified sawdust, the percentage of saccharification decreased from 33% at the lowest mass to 13% at the highest mass of 10 mg. The degradation of the delignified sawdust resulted in 70% saccharification at the lowest mass of 2 mg, while a 15% saccharification was obtained at the highest mass of 10 mg.

The development of alternative and renewable energy resources would become a crucial phenomenon as the use of petrochemical-derived substances results in the intensification of climate change and global warming. What is also of immense importance is to introduce procedures for the effective management of solid waste which negative effects on the environment are visible and realized. Biomass has already been proven to fulfill the requirements of a clean resource for the energy industries and feedstock for chemical-related industries (Long et al. 2013). An effective dual action would be to consider organic solid waste such as sawdust as a potential resource for bio-energy as well as bio-product development. Such an action would not only limit the amount of solid waste, but it could also decrease the global dependence on fossil fuels for energy and feedstock purposes. Certain pretreatment procedures, such as delignification before cellulase-catalyzed degradation, should be a standard procedure as it increases the bioconversion of cellulose into fermentable sugars such as glucose (Soleimanzadeh et al. 2023). It should also be advisable to grow cellulase systems that are more active on the cellulose structure by focusing on the production of cellulase systems with more active endoglucanase, exoglucanase, and B-glucosidase enzyme components (Ejaz et al. 2021). Sawdust is not the only organic waste material that is susceptible to saccharification by the cellulase enzyme system, as other materials like wastepaper, kitchen waste, and garden waste are also potential resources due to their relatively high cellulose content.

CONCLUSIONS

Cellulose, one of the structural components of organic waste materials such as sawdust, has the potential to be developed as a resource for alternative and renewable fuel production and bioproduct synthesis. Glucose, the major saccharification product produced during cellulase-catalyzed degradation of cellulose, is a fermentable sugar with a huge potential as a renewable feedstock for many synthetic procedures. Concluded from this study is not only that cellulose from different trees has different susceptibilities for a specific cellulase enzyme system, such as that from *A. niger*, but that delignification resulted in an increase in saccharification compared to the amount of sugar released from nom-pretreated sawdust. The positive effect of increasing masses of the non-delignified and lignified materials on the sugar-

releasing ability of the cellulase enzyme acting on cellulose is also well concluded. Delignification proved to be an effective pretreatment procedure, resulting in an increase in the degradation of sawdust compared to the non-delignified material.

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ORCID DETAILS OF THE AUTHORS

J. P. H. van Wyk: https://orcid.org/0000-0003-3067-6231