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A Study on Physicochemical Characteristics of Vetiver Grass (Vetiveria Zizanioides) in Bioenergy Production

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The use of lignocellulose biomass is crucial in biofuel production, to enhance global energy security and avoid food shortages. Feedstock selection of this type of biomass depends on factors such as cellulose, hemicellulose, lignin content, sustainability, quantity, and availability. As such, a good understanding of chemical attributes is crucial for potential energy conversion. Before characterization, the grass was ground and sieved to 0.6 mm for uniformity. The Vetiver grass (VG) was then analyzed through proximate, ultimate, SEM, FTIR, and lignocellulosic characteristics analysis. The VG had 7.63 % moisture content, 4.46 % ash, 69.10 % volatile matter, and 18.81 % fixed carbon. The ultimate analysis showed that the C, H, O, N, and S content was 42.28 %, 5.54 %, 51.54 %, 0.64 %, and 0 %, respectively. Compared to Napier grass, cow dung, and sugar cane bagasse, VG had lower nitrogen and no sulfur with an empirical formula of CH1.6O0.92. The treated VG sample showed increased intensity in the FTIR peak at 1037 cm-1 and additional peaks at 2341 cm-1. Broadening of the peak at 3332 cm-1 indicated improved availability of cellulose and hemicellulose for methane production due to mechanical treatment. The SEM analysis showed the removal of the link between the hemicellulose, cellulose, and lignin cross-linking structure. These results suggest that VG could be a promising bioenergy source.

* 1. Introduction

The increase in energy consumption has depleted fossil fuels, leading to a worldwide energy crisis and climate change issues. The Paris agreement aims to limit global temperature rise to 2 oC by using renewable energy sources like biofuels made from biomass (Rogelj et al., 2019).

Three generations of biomass resources have emerged to alleviate the energy crisis. The first generation involves processing food crops like wheat, corn, rice husks, and sugar cane (Hernández et al., 2019). While it's a potential solution, it's also criticized for its negative impact on food security (Debnath & Das, 2022). Lignocellulose and algae are second and third-generation biomass, respectively (Kaloudas et al., 2021). However, due to the cultivation complexity process of algae, additional land and infrastructure are required (Collins et al., 2022). On the other hand, lignocellulose is the most plentiful renewable resource globally, but its fermentation process can be hindered by its high lignin composition, requiring pretreatment before processing. Due to its complexity and high cost, commercialization of this biomass is considered to be challenging (Broda et al., 2022). Despite the challenges, it's still considered a viable and sustainable resource for biofuel production.

For this study, more focus is on Vetiver grass (VG) which is a second-generation biomass that has been considered a promising bioenergy feedstock. It is a fast-growing and low-cost crop that can tolerate various environmental conditions, such as flood, drought, and submergence, and is resistant to pests and diseases (Chuengcharoenphanich et al., 2023). VG is native to South-East Asia and tropical India and has successful applications in reforestation and erosion control (Huong et al., 2022).

To determine the viability of lignocellulosic biomass as a bioenergy source, its attributes like hemicellulose, lignin, cellulose, volatile solids content, particle size, and C/N ratio need to be identified. These parameters vary depending on their ecotype and origin (Wongwatanapaiboon et al., 2012). Different studies have reported the cellulose, hemicellulose, and lignin content of VG in Thailand ranging between 31.85 - 38.51 %, 37.87 - 42.61 %, and 3.67 - 5.06 %, respectively (Wongwatanapaiboon et al., 2012). Some cellulose, hemicellulose, and lignin characteristics of VG biomass cultivated in India were 39.11 %, 48.57 %, and 11.02 %, respectively (Thakur et al., 2018). In Indonesia the lignocellulosic components were reported to be 31.39 % cellulose, 34.55 % hemicellulose, and 17.58 % lignin (Restiawaty and Dewi, 2017).  These variations highlight the need for proper characterization of South African VG as feedstock for bioenergy production for anaerobic digestion.­­

* 1. Materials and methods

This section outlines the materials and methods used to characterize the physicochemical properties of VG.

2.1 Materials and pre-treatment method for biomass

VG was grown from a farm situated in Bapsfontein in Gauteng. The distinct collected grass was sun dried for 5 days before being analyzed. The grass was mechanically treated using a mortar and pestle and sieved to an appropriate uniform size of 0.6 mm for easy processing.

2.2 Analytical methods

2.2.1 Proximate analysis and ultimate analysis

The biomass was evaluated using proximate analysis to determine moisture content (MC), ash content (AC), volatile matter (VM), and fixed carbon (FC) by a modified procedure followed by Fajobi et al., (2022). The analyses were performed in triplicates. For the MC test, the samples were placed in the preheated oven at a rate of 105 °C for 90 min. For the AC tests, samples were combusted for 2 h, while for VM test, they were subjected to 7 min at 600 °C. The values of MC, AC and VM were computed using the eq. (1), (2), and (3), respectively.

$MC=\frac{W\_{cb}-W\_{cba}}{W\_{cb}-W\_{c}}×100\%$ (1) $AC=\frac{W\_{cbc}-W\_{c}}{W\_{cb}-W\_{c}}×100\%$ (2) $VM=\frac{W\_{cb}-W\_{cbi}}{W\_{b}}×100\%$ (3)

where Wc = weight of empty crucible, Wcb = weight of crucible + raw VG sample, Wcba = weight crucible + VG sample after drying, Wcbc = weight crucible + VG sample after combustion, Wcbi = weight crucible + VG sample after incineration, and Wb = weight of raw VG sample.

The FC value was calculated by subtracting the sum of mass percentages of MC, VM, and AC from the total mass (100 wt.%) as shown in eq. (4). The elemental analysis of the sample was carried out to determine the C, N, O, S, and H contents using the PerkinElmer® 2400 Series II CHNS/O Elemental Analyzer. The C/N ratio was then calculated using eq. (5).

$FC=100-\left(\%MC +\%AC+\%VM\right)$ (4) $ C:N ratio=\frac{\% Carbon}{\% Nitrogen }$ (5)

2.2.2 Lignocellulosic properties

The lignocellulosic properties were determined using a procedure adopted from Ayeni et al. (2015). To measure extractives, 60 ml of acetone per 1 g of dried biomass sample was used and heated to 90 °C for 2 h. The difference in weight before and after the extraction process is the amount of extractive present in the sample. To obtain hemicellulose content, 1 g of biomass is boiled in 150 mL of 500 mol/m3 sodium hydroxide (NaOH) solution for 3.5 h, filtered and washed until neutral pH, and dried at 105 °C. The difference in weight is the hemicellulose content (%w/w). To determine lignin content, biomass is treated with 72 % sulfuric acid (H2SO4) and autoclaved. The hydrolysates are filtered and dried to determine acid-insoluble lignin. Acid-soluble lignin is measured at 320 nm absorbance. Acid insoluble and soluble lignin are measured and added to calculate the total lignin content. Cellulose content is estimated by subtracting extractives, hemicellulose, lignin, ash, and other components from the total biomass weight.

2.2.3 Higher Heating value (HHV) and lower heating value (LHV)

A CAL3k-F bomb calorimeter was used to determine the heating value of VG. The analyses were performed in triplicate and yielded average values. Using the results of the ultimate analysis, the models reported by Thakur et al., (2017) were used to forecast the higher heating value (HHV) and lower heating value (LHV) of the biomass theoretically using eq. (8) and (9). The heating value was calculated in MJ/kg.

$HHV \left(\frac{MJ}{kg}\right)=0.341C+1.323H+0.0685-0.0153A-0.1194 \left(O+N\right)$ (8)

$LHV \left(\frac{MJ}{kg}\right)=HHV-H\_{g}(\frac{9H}{100}+\frac{M}{100})$ (9)

2.3 Characterization methods

The raw VG sample and mechanically treated samples were analyzed using FTIR spectroscopy and SEM to identify the functional groups and analyze the morphology, respectively.

2.3.1 Fourier transform infrared spectroscopy (FTIR) analysis.

The Perkin Elmer FTIR spectrum was utilized in this study. The samples were pressed onto a disc and scanned over a range of 600-4000 cm-1 with a resolution of 4 cm-1. The spectrum was then recorded.

**2.3.2 Scanning electron microscopy (SEM) analysis.**

The SEM micrographs were generated through the JEOL JSM-IT300 at a voltage of 20.0 Kv. To prepare the sample for SEM, it was coated with carbon using the coating machine Quorum model with plate number 1 to minimize sample charging and enhance visibility.

* 1. Results and discussions

3.1 Analytical results (proximate analysis, ultimate and heating value analysis)

This study compared the characteristics of VG with other biomass like Napier grass (NG), cow dung (CD), and sugar cane bagasse (SCB). It focused on the proximate, ultimate, lignocellulosic characteristics, HHV, and LHV analysis. Table 1 summarizes the results of the physiochemical characteristics of the biomasses.

Table 1: Physiochemical characteristics of VG, NG, CD and SCB

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Biomass characteristics  | VG  | NGa,b | CDc,d | SCBe |
| Proximate analysis (dry basis) [%] |  |  |  |  |
| Moisture content  | 7.63 | 4.5 | 8.1 | 4.68 |
| Ash content  | 4.46 | 6.31 | 14 | 9.75 |
| Volatile matter  | 69.10 | 85.52 | 62.7 | 79.30 |
| Fixed carbon  | 18.81 | 8.17 | 15.2 | 10.95 |
| Ultimate analysis |  |  |  |  |
| Carbon [%] | 43.69 | 45.1 | 34.3 | 43.56 |
| Hydrogen [%] | 5.95 | 5.94 | 8.05 | 6.03 |
| Nitrogen [%] | 0.74 | 0.45 | 4.1 | 0.20 |
| Oxygen [%] | 49.62 | 48.52 | 53.3 | 40.46 |
| Sulphur [%] | 0 | 0 | 0.21 | 0 |
| C/N | 59.04 | 100.2 | 8.37 | 217.8 |
| Lignocellulosic characteristics [%] |  |  |  |  |
| Cellulose | 41.9 | 35.2 | 16.6 | 39.4 |
| Hemicellulose | 38.91 | 26.6 | 15.3 | 27.9 |
| Lignin | 11.06 | 35 |  0 | 20.9 |
| Extractives | 3.67 | - |  - | 1.78 |
| Heating value [MJ/kg] |  |  |  |  |
| HHV (experimental) | 15.91 | 16.7 | 16.4 | 18.2 |
| HHV (theoretical) | 15.41 | 17.3 | 12.6 | 17.9 |
| LHV (theoretical) | 14.16 | 16.1 | 10.74 | 16.6 |

\*(Md Said et al., 2019)a, (Holder et al., 2018)b, (Ashraf et al., 2021)c, (Yan et al., 2018)d, (Najafi et al., 2023)e

The moisture content of biomass used in anaerobic digestion is crucial. As such, it is important to monitor the moisture content to avoid resistance to mass diffusion which can ultimately decrease methane production. In this study, the average moisture content of VG was 7.63 % which was higher than that of NG, SCB but lower than CW. VG also has a VM content of 69.10 %, making it suitable for anaerobic digestion. Although having higher VM content is beneficial, extremely high concentrations may lead to an increase in acidification while low VM concentrations can decrease microbial activity (Induchoodan et al., 2022). The present VG sample is further characterized by a low AC of 4.46 %, acceptable for anaerobic digestion. NG, CW, and SCB have higher AC of 6.31 %, 14 % and 9.75 %, respectively, which can result in reduced fuel quality (Dahunsi et al., 2019). The results also show FC content of 18.81 %, 8.17 %, 15.2 % and 10.95 % for VG, NG, CD and SCB, respectively.

Additionally, Table 1 features the elemental analysis (CHNSO) for the selected biomasses. VG has a C, H, O, N and S content of 42.28 %, 5.54 %, 51.54 %, 0.64 % and 0 % respectively. NG and SCB have a similar composition, while CD has sulfur (0.21 %) and nitrogen (4.1 %). N and S in biomass can emit NOx and SOx during anaerobic digestion which raises environmental concerns, but VG has low N and no S content. Therefore, VG is an attractive energy source with an empirical formula of CH1.6O0.92.

In anaerobic digestion, C/N ratio quantifies carbon and nitrogen in biomass, providing energy and supporting cell structure formation. Acceptable ratios range from 20-35 % (Kainthola et al., 2019). C/N ratio of 59.04 % was found in the study which is relatively high and indicates lower gas production due to the fast consumption of nitrogen by methanogens (Zheng et al., 2021). Therefore, co-digestion of VG with other biomasses to balance the high C/N ratio can be crucial for efficient biogas production.

VG has an HHV of 15.91 MJ/kg, similar to its theoretical HHV of 15.41 MJ/kg and comparable to NG and CD. Its LHV is estimated at 14.16 MJ/kg. Meanwhile, SCB has a higher HHV of 18.2 MJ/kg compared to others.

According to Table 1, the percentages of cellulose, hemicellulose, and lignin in different types of biomasses were as follows: VG (41.9, 38.91, 11.06 %), NG (35.2, 26.6, 35 %), CD (16.6, 15.3, 0 %) and SCB (39.4, 27.9, 20.9 %). Biomass with low lignin content, such as VG, is recommended for an efficient anaerobic digestion process. This is because the hard structure of high lignin content biomass makes it difficult to be converted by the anaerobic consortium, resulting in decreased biogas output (Ma et al., 2019).

3.2 Characterization results

3.2.1 FTIR

Figure 1 shows spectra with peaks at 3431, 2917, 1649, 1388, and 1037 cm-1 for raw VG and a spectrum with peaks at 3332, 2912, 2341, 1616, 1245, and 1037 cm-1 for pre-treated VG.



Figure 1: FT-IR analysis of vetiver grass, raw vetiver grass (R-VG) and pre-treated vetiver grass (P-VG)

The presence of the C-O-C stretching is responsible for the strong peak at 1037 cm-1, representing the ether functional group. The peak at 1037 cm-1 intensifies in the pre-treated VG sample, indicating the availability of cellulose and hemicellulose polymers. The increased intensity can be attributed to the grass' rigid structure disintegrating, which exposes the cellulose and hemicellulose content. Hemicellulose's acetyl groups form acetic acid, used by methanogenic bacteria for methane production (Eswari et al., 2023). The sample shows peaks between 1245 to 1388 cm-1 indicating the presence of C-O stretching, aromatic ring vibrations, and phenol (O-H) hydroxyl groups. These peaks suggest the existence of lignin and hemicellulose.

Dual peaks are seen at 1616 and 1649 cm-1 after pre-treatment. The peak at 1616 cm-1 is broader due to increased presence of aromatic C=C rings stretching. The double sharp peak at 2917 cm-1 reduced in intensity in the untreated sample and was observed at 2912 cm-1 in the treated sample. The peaks are due to C-H stretching of carboxylic acid group caused by lignin. The pre-treated sample showed decreased peak intensity due to lignin structure breakdown linked to intense peak of 2917 cm-1 in the raw sample. The change in peaks at 2912 cm-1 affected the shape of the peaks at 3332 cm-1, indicating the presence of O-H stretching vibration in hemicellulose, cellulose, and lignin with an intensified broad peak. This peak also indicated the increased presence of cellulose and hemicellulose for microbial attack.

3.2.2 SEM

The SEM analysis is used to visualize the features and degradation of lignocellulose biomass at a cellular and nano-resolution level. In this study, both untreated and pre-treated VG samples were analyzed. The surface of untreated VG is shown in Figure 2(a) at a WD of 11.5 mm, HFW of 10 μm and magnification of x1000. The figure displays stomata and a highly rigid structure consisting of cellulose, hemicellulose, and lignin, which are organized into macro-fibrils format. The surface of the pre-treated VG shows significant disintegration features in Figure 2(b) at a WD of 11.9 mm, HFW of 20 μm and magnification of x700. The macro-fibrils are disrupted, revealing the removal of the cross-linking structure between cellulose, hemicellulose, and lignin, exposing cellulose fibrils. The disruption of the lignin-hemicellulose complex increases the surface area, providing more sites for enzymatic hydrolysis by microorganisms (Zhao et al., 2012).

(a)

(b)

Figure 2: SEM images of (a) raw vetiver grass and (b) pre-treated vetiver grass sample

* 1. Conclusions

This study analyzed the potential of VG for bioenergy production by evaluating its biomass characteristics, including proximate, ultimate, and heating value analysis. The morphology and functional groups of the grass were also analyzed to determine its chemical and physical structure. The VG had 7.63 % MC, 4.46 % AC, 69.10 % VM, and 18.81 % FC. It has potential for bioenergy through anaerobic digestion due to its high volatile matter, low moisture, and ash content. The ultimate analysis shows it has 42.28 % C, 5.54 % H, 51.54 % O, 0.64 % N, and 0 % S, with an empirical formula of CH1.6O0.92, making it less of an environmental concern than other biomass. The lignocellulosic characteristic shows that VG consists of 41.9 % of cellulose, 38.9 % of hemicellulose and 11.06 % of lignin. The VG sample showed improved availability of cellulose and hemicellulose for methane production due to mechanical treatment, as indicated by increased intensity in the FTIR peak at 1037 cm-1, additional peaks at 2341 cm-1 and broadening of the peak at 3332 cm-1. SEM analysis showed the removal of the link between hemicellulose, cellulose, and lignin cross-linking structure, increasing surface area for enzymatic hydrolysis. Therefore, VG could be a promising bioenergy source, aligning with the trend toward exploring low-cost biomass alternatives.

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