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Proximate and Ultimate Analysis of *Eichornia natans* (Water Hyacinth), *Pistia stratiotes* (Water Lettuce) and *Nymphaea lotus*(Water Lily) in the Production of Biofuel

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ABSTRACT

The Proximate and Ultimate analyses of some selected freshwater biomasses: Eichornia natans (Water Hyacinth), Pistia stratiotes (Water Lettuce), and Nymphaea lotus (Water Lily) were undertaken with the aim of presenting the analytical results and ascertaining the biomass composition for use as biofuel. Results showed that the amount of fat in the leaf of water hyacinth, water lettuce and water lily was 8.7%, 8.15% and 7.2% while the stem of water hyacinth, water lettuce and water lily recorded 3.65%, 2.2% and 7.25% respectively. The amount of reducing sugar in the leaf of water hyacinth was 0.31 g/l and 0.29 g/l was found in the stem. Similarly, 0.25 g/l was found in water lettuce leaf and 0.27 g/l in its stem, 0.38 g/l in water lily leaf and 0.35 g/l in its stem. The amount of nitrogen in the biomass varied between 2.35% and 7.76% and the amount of sulphur was between 0.27% and 3.24% while the amount of carbon was between 45.25% and 65.04%. The calorific value of the leaf of water hyacinth, water lettuce and water lily was 17.64 MJ/kg, 18.57 MJ/kg and 19.53 MJ/kg respectively, while the stem of water hyacinth, water lettuce and water lily recorded 20.53 MJ/kg, 24.93 MJ/kg and 20.98 MJ/kg respectively. The study concludes that aquatic weeds can be utilized as viable feedstock for the production of biofuel.

Keywords: Biofuel; Biomass; Freshwater; Proximate; Ultimate

INTRODUCTION

Energy consumption has increased steadily over the last century as the world population has grown and more countries have become industrialized [1]. Development of alternative renewable energy continues to grow in recent times due to the fear of energy insecurity in the near future and environmental with sociopolitical issues associated with the use of fossil fuels. Application of lignocellulosic biomass (non-food materials) such as forest residues, agro-wastes, energy grasses, aquatic plants and algae, etc. for bioenergy production seem promising as they are evenly distributed across the globe and have also eliminated initial public perception of food insecurity associated with first generation biofuels which were produced from food materials. In addition, these materials have low levels of sulfur, nitrogen and ash content which make them relatively environmentally friendly [2-3].

The important biomass properties includes: heating value, proximate analysis and ultimate analysis [4]. Any biomass conversion process begins with knowing its energy content in units of MJ/kg and compared with conventional sources like coal. Further, biomass resources may be describe based on its proximate analysis whereby its moisture content (MC) is reported, followed by the volatile combustible matter (VCM) contents, fixed carbon (FC) and ash. Finally, the ultimate analysis is important to illustrate the biomass composition in relation to the top

five elements it contains as follows: carbon (C), hydrogen (H), oxygen (O), nitrogen (N) and sulphur (S) content. Other characterization would include describing its compositional contents such as lignin, cellulose and hemi-celluloses, carbohydrates and fat contents [5].

More research has focused on using non-edible biomass as raw materials including lignocelluloses, celluloses, and marine algae rather than the first generation biomass such as starch and sugar biomass [6].

Freshwater biomass is aquatic weed which interfere with the use of water and constitute a nuisance to the environment and human welfare [7]. These freshwater plants can cause infestations over large areas of water and consequently lead to series of ecological problems including; reduction in biodiversity, blockage of watercourses, depletion of dissolved oxygen, alteration of water chemistry and causing environmental pollution [8]. More recently, attention has been focused on the potentials and constrains of using freshwater biomass for variety of applications. Their application as animal fodder and means of metal remediation has been reported [9]. The prospect of converting aquatic weeds to biogas and bioethanol is ongoing in some developing countries such as India [10].

Global depletion of energy supply due to the unsustainable consumption and the associated environmental problems of fossil fuel utilization have prompted the research on alternative energy sources [11]. The aim of this paper therefore, is to evaluate the proximate and ultimate analysis of *Eichornianatans* (Water Hyacinth), *Pistiastratiotes* (Water Lettuce), and *Nymphaea lotus* (Water Lily) in the production of biofuel.

MATERIALS AND METHODS

The plant samples were obtained from different locations within Kaduna metropolis. They were collected manually using gloved hands and placed in polythene bags, then transported to the laboratory for further preparation. *Eichornianatans* (Water Hyacinth) was obtained at Ungwanboro in Chikun Local Government, Kaduna within longitude 10^{o}30.332'N and latitude 007^{o}27.230'E, *Pistiastratiotes* (Water Lettuce) and *Nymphaea lotus* (Water Lily) were obtained at a farmland in Zangon Aya, Igabi Local Government, Kaduna within longitude 10^{o}55.117'N and latitude 007^{o}39.733'E; 10^{o}55.131'N and 007^{o}39.577'E respectively. The plant samples were authenticated and a voucher number was given at the herbarium section in the botany department of Ahamdu Bello University, Zaria.

The aquatic plants were thoroughly washed with tap water to remove adhering dirt and were chopped into separate pieces of leaves and stems using sharp knife. The leaves and stems were air dried, then grinded using a blender and sieved with a 0.4mm mesh and kept in a labelled container for further use [12].

2.1 Determination of Moisture Content

The moisture content of the leaves and stems were determined in triplicate by weighing 5 g of each samples in a labelled pre-weighed petri dish then placed in an oven to dry at 105^{o}C for 3 hours. After drying, the petri dish and dry sample was transferred to a desiccator to cool before being weighed again. The percentage moisture was calculated as percentage weight loss moisture content [13].

2.2 Determination of Volatile Matter

The volatile matter of the leaves and stems were determined in triplicate by weighing 2 g of each samples in a crucible, then covered with a lid and placing it in a muffle furnace at a temperature of 550°C for 10 minutes and then weighed after cooling in a desiccator [14].

2.3 Determination of Ash Content

The ash content was determined in triplicate by weighing 2 g of the sample into a crucible, then placed in a muffle furnace and heated at 550^{o}C for 8 hours. Then the crucibles was taken out from the furnace and put into the desiccator to cool. After cooling the weight was taken to determine the ash content [13].

2.4 Percentage fixed carbon

The percentage fixed carbon was computed by subtracting the sum of percentage volatile matter (PVM) and percentage ash content (PAC) from 100 as shown in the Equation below:

Fixed Carbon = 100% − $(PVM + PAC)$, [14].

2.5 Determination of Fat Content

The fat content was determined using soxhlet fat extraction method. 2g of the dried sample was placed in an extraction thimble plugged lightly with cotton wool. A round bottom flask was filled with 200 cm^3 of petroleum ether and boiled at 50^oC. The boiling flask containing the petroleum ether placed in the extraction thimble and the soxhlet apparatus was allowed to reflux till extraction was complete. The extracted solvent in the flask was transferred into an already weighed oven dried beaker, then evaporated to dryness at 50^oC using a water bath. The beaker was then transferred into a desiccator to cool before weighing[15].

2.6 Determination of Sugar Concentration

A portion (3 g) of the leaves and stems of each dried pulverized plant sample was weighed using an analytical balance and placed into a 250 cm^3 conical flask, then 4% sulfuric acid was added to make a solution. The mixture was autoclaved at 121^oC for 15 minutes and then filtered to remove the unhydrolysed materials [16].The filtrate from the pretreatment was collected and analyzed for the reducing sugar content by using DNS test.

Total sugar content was estimated using dinitrosalicylic acid (DNS) reagent. 3 cm^3 of DNS reagent was added to 3 cm^3 of hydrolyzed sample in a lightly capped test tube. The mixture was heated on a hot plate at 90ºC for 5-15 min to develop the red brown color. Further 1 cm^3 of 40% Potassium tartrate (Rochelle salt) solution was added to stabilize the color. After cooling at room temperature, the absorbance was taken with a UV spectrophotometer at 540nm. The concentration of reducing sugar was determined by making reference to a standard curve of known glucose concentration [17].

2.7 Nitrogen Content Determination

The amount of nitrogen in the sample was determined by weighing 0.2 g of the sample into a Kjeldahl digestion tube. 20 cm^3 of H_2SO_4 and 3 g of a mixed catalyst consisting of $CuSO_4$, Na_2SO_4 and selenium powder was added into the tube. The tube was heated gently until boiling for about 3 hours, then allowed to cool. The cooled mixture was then diluted with 100 ml of distilled water in a volumetric flask. To an aliquot (10 cm^3) of the digest, 10 cm^3 of 40% NaOH solution was added to the connected Kjeldahl distillation apparatus and heated to boiling. The distillate was condensed into the conical flask containing 10 cm^3 of 2% boric acid and 2 drops of methyl blue indicator was added and the alkaline distillate was titrated against 0.01 M sulphuric acid acid[13].

2.8 Determination of Sulphur

The amount of sulphur in the biomasses was determined by digesting 0.2 g of with 3 cm^3 of HNO_3 and 2 cm^3 of $HClO_4$, then transferred into a 100 cm^3 volumetric flask and made up to the mark with distill water. $BaCl_2$ was added to 10 cm^3 of an aliquot to get a resulting turbid solution. The absorbance of the turbid solution was determined using a colorimeter at wavelength of 420nm [18].

2.9 Determination of Carbon

Carbon was determined using the procedure described in [19] by weighing 1 g of the sample into a 250 cm^3 conical flask. 10 cm^3 of 0.1 M $K_2Cr_2O_7$ was added, then 20 cm^3 of conc H_2SO_4 was added to the solution in the conical flask, then allowed to cool for about 30 minutes. After cooling 100 cm^3 of distilled water was added and 3 drops of O-phenanthroline ferrous complex was added as an indicator and titrated with 0.5 M $FeSO_4$.

2.10 Determination of Calorific Value

The determinations of calorific values was made using a bomb calorimeter (model 6100 series manufactured by Parr Instrument Company) in the chemical engineering department, Ahmadu Bello University, Zaria by weighing 0.5 g of the sample into an ignition cup, then wrapped with a fuse wire and sealed inside the bomb. The bomb was then filled with 30 atmosphere of oxygen, then placed in a static jacket filled with 2 liters of water. After combustion has taken place in about 6 minutes, the calorific value was read out on the screen of the calorimeter in MJ/Kg.

RESULTS AND DISCUSSION

The result of the proximate analysis for *Eichornianatans* (Water Hyacinth), *Pistiastratiotes* (Water Lettuce), and *Nymphaea lotus* (Water Lily) is presented in Table 1. Analysis of Variance (ANOVA) was used to test the significance difference in the values obtained and Fisher Least Significant Difference (LSD) was used to separate the means.

Table 1 shows that the stem of water lily has the highest percentage of moisture content of 93.05% while the leaf of water hyacinth has the lowest with 82.96%. The moisture contents of other biomasses that is, water hyacinth stem, water lettuce leaf, water lettuce stem and water lily leaf are respectively 84.21%, 91.99%, 90.11% and 84.06%. The comparatively high moisture contents of these biomasses indicates that they would have to be dried so that they could easily burn off when used as sources of heat.

Table 1: Proximate Analysis of Water Hyacinth, Water Lettuce and Water Lily

Biomass	Moisture Content (As Received) (%)	Moisture Content (Dry Basis) (%)	Volatile Matter (%)	Ash Content (%)	Fixed Carbon (%)	Dry Matter (%)	Fat (%)
Water Hyacinth Leaf	82.96±0.53^{c}	2.96±0.24	67.08±0.40^{a}	13.93±0.83^{d}	18.98±0.86^{d}	17.04±0.53^{a}	9.27±0.81^{a}
Water Hyacinth Stem	84.21±0.41^{c}	3.48±0.26	57.67±2.28^{b}	19.80±1.25^{bc}	22.53±2.33^{cd}	15.79±0.41^{a}	3.65±0.24^{c}
Water Lettuce Leaf	91.99±0.48^{ab}	4.21±0.18	57.23±2.45^{b}	23.20±0.20^{a}	19.57±2.63^{d}	8.01±0.48^{bc}	8.17±1.15^{ab}
Water Lettuce Stem	90.11±0.90^{b}	3.01±0.05	39.20±2.34^{d}	23.87±1.53^{a}	36.93±3.86^{a}	9.89±0.90^{b}	2.21±0.19^{d}
Water Lily Leaf	84.06±1.81^{c}	5.53±0.56	57.94±0.77^{b}	17.53±1.03^{c}	24.53±1.12^{bc}	15.94±1.81^{a}	7.64±0.55^{b}
Water Lily Stem	93.05±2.85^{a}	6.77±0.24	51.02±3.48^{c}	22.00±2.12^{ab}	26.98±1.85^{b}	6.95±2.85^{c}	7.48±0.21^{b}
LSD	3.13	0.13	7.17	2.46	7.92	3.13	0.59

**Means that do not share a letter are significantly different (P>0.05)*

The amount of fat extracted from the biomasses varied from 2.21% for the stem of water lettuce to 9.27% for the leaf of water hyacinth. Similarly, 8.17%, 7.48%, 3.65% and 7.64% was recorded for water lettuce leaf, water lily stem, water hyacinth stem and water lily leaf respectively. These results show that except for the stem of water hyacinth and water lettuce with fat contents less than 5%, other biomasses have values higher than 5%. These fall above values obtained for groundnut shells (3.42%) and corn cub (3.42%) and much lower than those of well-known biomass for biodiesel production like neem seed (39%), moringa (31%), jatropha (60%) and castor seed (54%) [20-21].

The results of ash content show the amount of inorganic substance that would remain after complete combustion of the biomass. The ash contents of the biomasses varied from 13.93% for leaf of water hyacinth to 23.87% for the stem of water lettuce. The ash contents of the other selected biomasses are 23.20%, 22.00%, 19.80% and 17.53%, for water lettuce leaf, water lily stem, water hyacinth stem and water lily leaf respectively. These results show that the biomasses have ash content between 15% and 25% except for the leaf of water hyacinth that is less than 15%. This falls below values obtained for rice husk briquette (16.10%) and melon shell (19.57%) and higher than those of well-known biomass fuel like grape pomace (2.7%), apple pomace (4.0%), and wood (0.1%) [21-23].

Volatile matter refers to the part of the biomass that is released when the biomass is heated (up to 400 - 900°C). During this heating process the biomass decomposes into volatile gases and solid char. Biomass typically has a high volatile matter content (up to 80 percent) [23]. The leaves of water hyacinth, water lettuce and water lily recorded a volatile content of 67.08%, 57.23% and 57.94% respectively while their stems recorded 57.67%, 39.20% and 51.02% respectively. The values obtained are a little lower than some other used biomass like rice husk briquettes which has 68.20% [23] but higher than the value obtained for fossil fuel such as coal (32.56%) [24].The values obtained is high and signifies easy ignition of the biomass and proportionate increase in flame length as suggested by Loo and Koppejan[25]. The high volatile matter content indicates that during combustion, most of the biomass will volatilize and burn as gas in combustion chambers [23].

The relatively low percent dry matter in all the selected biomasses as compared to rice husk briquette (87.33%), groundnut shell (70.50%) and coal (94.01) [21,24] indicate that a lower percentage (varying from 8.01% for the leaf

of water lettuce to 17.04% for the leaf of water hyacinth) would be available for combustion. This implies that the biomass would have to be dried so that a higher percentage will be available for use.

Fixed carbon is a measure of the solid combustible material in solid fuel after the expulsion of volatile matter; its content is used as an estimate of the amount of coke that will be obtained on carbonization [26]. The fixed carbon of a fuel is the percentage of carbon available for char combustion. For the selected biomasses, it was found to be 18.98, 22.53, 19.57, 36.93, 24.53 and 26.98% for water hyacinth leaf, water hyacinth stem, water lettuce leaf, water lettuce stem, water lily leaf and water lily stem respectively. The fixed carbon gives a rough estimate of the heating value of a fuel [26].

The concentration of reducing sugar present in the freshwater biomass after 4% sulfuric acid hydrolysis pretreatment is shown in Table 2. The amount of reducing sugar varied between 0.25 g/l and 0.38 g/l for the leaf of water lettuce and water lily respectively. Similarly, 0.31 g/l and 0.29 g/l was found in the leaf and stem of water hyacinth, while 0.27 g/l and 0.35 g/l was found in the stem of water lettuce and water lily respectively. These values obtained are in agreement with the work ofNamadi and Awasthi [12, 27]. The reducing sugar content is essential in the fermentation of biomass into bioethanol.

Table 2: Reducing Sugar Concentration of the Selected Freshwater Biomass

Biomass	Absorbance at 540nm	Reducing Sugar concentration (g/l)
Water Hyacinth Leaf	0.197	0.31
Water Hyacinth Stem	0.176	0.29
Water Lettuce Leaf	0.156	0.25
Water Lettuce Stem	0.163	0.27
Water Lily Leaf	0.234	0.38
Water Lily Stem	0.217	0.35

Ultimate Analysis involves the estimation of important chemical elements that makes up the biomass, namely carbon, nitrogen and sulphur. Table 3 shows the result of the ultimate analysis of the freshwater biomasses.

The percentage of carbon in the biomass are 45.25%, 50.75% and 56.42% for the leaves of water hyacinth, water lettuce and water lily respectively and 52.45%, 65.04%, and 53.78 for the stems of water hyacinth, water lettuce and water lily respectively. The values obtained are lower than the values for coal (82.8%) as reported byAdekunle [24], Number 2 fuel oil (87.3%) and natural gas (74.9%) shown in Table 3. However, the values obtained in this study are higher than some other biomasses in other literatures: rice husk briquette (45.2%), groundnut shell (14.99%), corn cob (19.73%) and melon shell (21.61%) [21, 23]. A good biomass should have high amount of carbon. The higher the carbon content, the higher is the calorific value and the better is the quality of the biomass [24].

The sulphur contents of the biomass varied between 0.27% for the stem of water hyacinth and 3.24% for water lettuce leaf as shown in Table 3. Other results include 2.94% and 2.76% for the leaves of water lily and water hyacinth respectively, 2.29% and 1.71% for the stem of water lettuce and water lily respectively. The sulphur content was observed to fall below 4% in all the samples, which would mitigate the emission of sulphur dioxide (SO_2) into the atmosphere causing acid rain. Sulphur contents are higher for the freshwater biomass than for Number 2 fuel oil (0.22%) as shown in table 4.2, coal has about 0.6% beacuse calcium hydroxide was added as a desulphurizer to reduce its sulphur content. Natural gas (methane) produces virtually no sulphur [24, 28,29].

From the analysis, nitrogen content is less than 5% for the stems of water hyacinth and water lily with 2.35% and 2.24% respectively. These results are comparable to results obtained for conventional fuels like coal (2.2%) and non-conventional fuels like agricultural wastes such as yam peels (2.67%) and mango peels (2.40%) [21,24]. The other samples: the leaves of water lily, water hyacinth and water lettuce have nitrogen content of 4.13%, 3.89%, 4.76% respectively while the stem of water lettuce stem had 3.33%. The amount of nitrogen is a little higher due to the likely effect of fertilizer runoff from farmlands into the water body where the samples were collected. Fuel bound nitrogen is an important contributor to oxides of nitrogen (NOx) emission from biomass combustion system [30, 31]. Both nitrogen and sulphur content are not important in biofuel production. They tend to increase the release of toxic gases that are either irritants (NOx, SO2, aldehydes and acrolein) or asphyxiants (HCN) which may cause adverse effect to living organisms.

The calorific values obtained from for the leaf of water hyacinth, water lettuce and water lily was 17.64 MJ/kg, 24.93 MJ/kg and 20.98 MJ/kg while the stem of water hyacinth, water lettuce and water lily recorded 20.53 MJ/kg, 18.57 MJ/kg and 19.53 MJ/kg respectively. This energy value is sufficient enough to produce heat required for household cooking and small scale industrial applications. The values obtained are higher than some biomass energy such as groundnut shell briquette 12.60 MJ/kg [32], cowpea 14.37 MJ/kg, and soybeans 12.95 MJ/kg [33].

Table 3: Comparison of Ultimate Analysis for the freshwater biomass and fossil fuels

Biomass	N (%)	S (%)	C (%)	HHV (MJ/kg)
Water Hyacinth Leaf	3.89	2.76	45.25	17.64
Water Hyacinth Stem	2.35	0.27	52.45	20.53
Water Lettuce Leaf	4.76	3.24	50.75	18.57
Water Lettuce Stem	3.33	2.29	65.04	24.93
Water Lily Leaf	4.13	2.94	56.42	19.53
Water Lily Stem	2.24	1.71	53.78	20.98
Coal **[24]**	2.4	0.6	82.8	32.93
Number2 Fuel Oil **[30]**	0.006	0.22	87.3	43.43
Natural Gas (95% Methane) **[31]**	-	-	74.9	55.56

CONCLUSION

The result obtained suggests that freshwater biomass can be used in the production of biofuel. However they might not be viable resource for biodiesel production. Further studies should be carried out on how to increase their fat contents through genetically modified biotechnology. The high heating value (calorific value) of 17.64 – 20.98 MJ/kg obtained for the biomasses indicates that they can produce heat required for household cooking and small scale industrial applications. All the freshwater biomasses considered have heat values about the same values of some well-known biomass-fuels and fall within the limit for the production of steam in electricity generation.

The low composition of nitrogen and sulphur in virtually all the sample selected will result in low emission of oxides of nitrogen and sulphur into the atmosphere and there may not be need for equipment for the removal of oxides of nitrogen in the design of equipment for the conversion of these freshwater biomasses to energy. The low percentage of sulphur is good for combustion since good fuels are known to have low sulphur contents.

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