Journal of Scientific Research & Reports

26(10): 81-92, 2020; Article no.JSRR.64400
ISSN: 2320-0227

Energetic Balance of Biogas and Fertilizing Potential of Digestate from Anaerobic Digestion of Manihot Utilissima

Beaudrique Mboko Nsimba[1,2], Philippe Ngoma Mambanzulua[1,2,3*], Norbert Lolofo Basosila[1], Virginie Alleluya Korangi[4], Mugunda Saidi[4] and Yamba Esole[4]

[1]Department of Chemistry, Faculty of Science, University of Kinshasa, P.O.Box 190, Kinshasa XI, D.R. Congo.
[2]Faculty of Oil, Gas and New Energies, University of Kinshasa, P.O.Box 127, Kinshasa XI, D.R. Congo.
[3]Faculty of Pharmacy, University of Kinshasa, P.O.Box 212, Kinshasa XI, D.R. Congo.
[4]Faculty of Agronomic Sciences, University of Kinshasa, P.O.Box 117, Kinshasa XI, D.R. Congo.

Authors' contributions

This work was carried out in collaboration among all authors. Authors BMN and PNM designed the study and wrote the protocol. Author BMN managed the analyses of the study. Authors BMN and PNM wrote the first draft of the manuscript and managed the study. Authors NLB and VAK performed the statistical analysis. Authors MS and YE managed the literature searches. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JSRR/2020/v26i1030324
Editor(s):
(1) Dr. Magdalena Valšíková, Slovak University of Agriculture, Slovakia.
Reviewers:
(1) P N E Naveen, Nadimpalli Satyanarayana Raju Institute of Technology (NSRIT), India.
(2) Francisco Javier Martínez Valdez, Universidad Tecnológica de Tecámac, México.
Complete Peer review History: http://www.sdiarticle4.com/review-history/64400

Original Research Article

Received 25 October 2020
Accepted 30 December 2020
Published 31 December 2020

ABSTRACT

Optimal scale production of biogas from the anaerobic digestion (AD) of organic wastes (OWs) as substrates has become one of stimulating environment-friendly procedures to foster the replacement of fossil energies by renewable ones. AD of OWs generated from households is desirable as an effective method to fight against environmental pollution effects of the latter in developing countries. Notably in Africa where each year there are more than 600,000 premature

---

**Corresponding author: E-mail: philippe.mambanzulua@unikin.ac.cd;*

deaths following the use of solid biomass energy (especially charcoal). Yet, if assessing the potential of biogas production, that is, biochemical methane potential (BMP) has played a prominent role in the choice of substrates, in the other hand the evaluation of the optimal ratio between the quantity of variety OWs and the amount of energy to be produced has not received several attentions. The latter is valuable not only for energetic productivity but also for profitability. Thus, in this report, an energetic balance between the amount of leaves and stems of Manihot Utilissima (MU) annually produced as well as the energetic potential of their biogas were investigated. Cow manure (CM) was employed as inoculum, under mesophilic conditions of the collection sites: Ngaba and Ndjili in Kinshasa City Province (KCP), Congo DR, where the leaves and stems of MU are among the most generated wastes. Furthermore, we evaluated the fertilizing potential of digestates from the AD of the leaves and stems by their Carbon/Nitrogen (C/N) ratios.

The annual energetic potentials of biogas produced were estimated to be 1.362 ± 0.028 10^9 kWh for the leaves and 0.337 ± 0.006 10^9 kWh for the stems. These were associated to the energy needs for the KCP households corresponding to the use of charcoal. The latter was evaluated to be 166 10^3 tons for leaves and 41 10^3 tons for the stems of MU, respectively.

The fertilizing potential of digestates from the AD of the leaves and stems of MU assessed by their C/N ratios were determined to be 5 and 10, respectively; indicating that they are favorable for the cultivation of vegetables and fruit trees in KCP soils (C/N~5) but also optimal for the organisms, soil conditioning and could improve the soils hydraulic conductivity (C/N~10).

Keywords: Biogas potential; energetic balance; manihot utilissima, digestates; ligneous biomass.

1. INTRODUCTION

Large cities in Sub-Saharan Africa, like KCP, Capital of Democratic Republic of Congo (DRC), are facing a dramatic population explosion [1-3]. This is the result of the increasing of rural-urban migration and the displacement of people from areas of armed conflicts such as the Eastern part of the DRC. Those displacements cause anarchic subdivisions in the peri-urban green spaces and increase crops and energy consumption in KCP [2-4]. Therefore, in KCP there is an increase in the production of improperly managed wastes, deforestation and decreasing areas of arable lands which are otherwise infertile [2-5]. In fact, the KCP produces annually about 3 million tons of wastes whose management is arduous. These wastes contain almost 55% of OWs, 94% of which are essentially vegetal sources [3,6,7].

Fortunately, from OWs, the AD or biomethanation produces biogas that essentially contains methane, and accordingly can solve the above mentioned multiple social-induced environmental issues. The AD of OWs with high energetic potential offers the possibility to recycle nutrients and inherently reduces greenhouse emissions [4,8-11]. AD has shown through its high performance to be an attractive and eco-friendly method for the treatment of the fermentable wastes, through which one can control the inherent pollution and cover energetic needs [12-15]. This energy is renewable and its net CO_2 contribution to the atmosphere is nil. Moreover, it could keep KCP away from deforestation by providing its households with a form of clean energy for cooking; given nearly 80 % of the population of KCP rely on ligneous biomass (mainly charcoal) to satisfy their energy needs [1-4]. The biogas obtained by biomethanation of these wastes, is a second generation biofuel. The latter is more advantageous than the first or third generation biofuels [2,4,8]. As an advantageous energy source, biogas is widely considered as a suitable fuel for heating facilities, power generation, and vehicles [16-18]. The residues or digestates that are generated from the AD are employed as an eco-friendly fertilizer; that could enable to avoid the non-environmental-friendly or traditional one that uses fire in agriculture, and consequently to protect the bio-diversity [2,4,19].

MU is widely cultivated in the world [20,21] and 11% of the global crops is owned by DRC, which is the second African producer after Nigeria [21]. The MU is very cultivated in the world because it contains abundant nutrients and in DRC it rapidly grows in marginal land like the KCP soils [3,4,20,21]. The increase in MU plantation is highly beneficial for economic development [20,21].

The leaves and stems of MU are one of the most abundant among the vegetable wastes produced in the KCP [3,4,6,7]. Mambanzulua et al. [4] previously investigated the energetic potential of the biogas from the leaves of MU. In his report, the AD was achieved by heating the reactor at 30 °C (the ambient temperature in tropical areas like KCP) and using an anaerobic sludge as inoculum. The latter was taken from an anaerobic digester treating agro-food organic wastes. It was inoculated before with a sludge collected from a full-scale anaerobic digester treating an activated sludge from a municipal waste water treatment plant. The reported results showed that leaves of MU were favorable for the AD under mesophilic conditions.

Indeed, the leaves of MU are often accompanied with their stems in the garbage dumps of KCP. However, the AD of stems of the MU at ambient temperature is still not carried out and has remained uninvestigated. The energetic balance of the leaves and stems of MU produced were not evaluated. Furthermore, the characterization of the fertilizing potential of their digestates and the arable land surfaces to be fertilized by these fertilizers were not experimentally assessed.

Thus, the main goal of the present investigation was to assess energetic potential of the biogas from the AD of leaves and stems of MU as well as the energetic balance between the biogas and the quantities these wastes annually produced in KCP. The substrates were sampled in two municipalities of the KCP: Ngaba (Rond point Ngaba Market) and Ndjili (Quarter 9), which are the most densely populated. After determining the BMP of these two substrates owing to their AD, the energetic potentials of these wastes were thereafter evaluated. And knowing the possible annual quantity of these wastes produced in the KCP, we finally had to determine the energetic balance of the biogas from the wastes.

Furthermore, we evaluated the fertilizing potential of the digestates generated and the necessary arable land surface breadths to be fertilized by the latter. Since the nature of the inoculum also has a significant influence on biogas production for a given biomass [4,22-24], the CM was used as inoculum under mesophilic conditions of the KCP. Moreover, in the KCP the CM is easily accessible; it is the most active inoculum in the KCP and more active than activated sludge [12,25-27]. More importantly, this process is economically beneficial to implement in the developing countries of Sub-Saharan Africa like the DRC.

We have empirically demonstrated the broad spectrum of energy needs of the KCP households in charcoal that can be covered by the biomethanization of the stems and leaves of MU and the potential fertilizer of their digestates.

2. MATERIALS AND METHODS

2.1 Substrates and Inoculum Sampling

Stems and leaves of MU were sampled in two municipalities of the KCP, in Ngaba: blue in the map (Rond point Ngaba Market) and in Ndjili: red in the map (Quarter 9) in DRC Fig. 1A. The choice of these two areas is due to the fact that they are the most densely populated where there are major generation of wastes of MU. The stems and leaves Fig. 1B-1C were identified by the Herbarium at the Department of Biology, University of Kinshasa. These samples were washed, dried at the ambient temperature, ground and stored in plastic bags and used for analyses and further tests.

The CM Fig. 1D was collected from Slaughterhouse of Masina in Kinshasa. The collected CM was kept at ambient temperature in sealed plastic bags.

2.2 Characterization of the Substrates and Inoculum

2.2.1 Physical and chemical analysis of the substrates and the inoculum

The substrates and inoculum, that is, leaves and stems of MU, and CM, respectively, were characterized by determining the following parameters: dry matter or dry weight (DW), the ash content and the organic matter or volatile solid (VS) (SI1A-SI1C). The contents of dry matter, ash and organic matter were evaluated according to the standard methods, based on weight loss of heating/or sintering of sample-desiccator, and were operated in the Memmert oven (SI1A-SI1C) [4,28,29]. Prior, the values of dry weight and the ash weight were determined and then followed by the content of organic matter or volatile solid. The DW were determined by drying the samples (substrates and inoculum) at 105 °C in oven (Memmert) and monitoring the weight until the latter became constant (SI1A). Then, the ash content were determined by sintering the dried samples in furnace (Nabertherm) at 600 °C until the weights were constant (SI1B). The content of organic matter or volatile solid were obtained as the difference between the values of dry weight and the ash weight (SI1C). The contents in P and K were determined by the spectrophotometry (SI1D) (spectrophotometer, Perkin Elmer Analyst 200). The principle consisted of a calcination followed by a solubilization of the elements by attack with a strong mineral acid. It was carried out with 1 g of DW of our samples at 450 °C in the oven followed by the transfer of the ash with a mixture of 10 mL of nitric acid in a 100 mL beaker. The mixture was subjected to gentle boiling digestion on a hot plate for 30 minutes. During digestion, care should be taken not to exceed 5 mL of evaporation. After cooling the mixture, 45 mL of distilled water were added to each beaker.

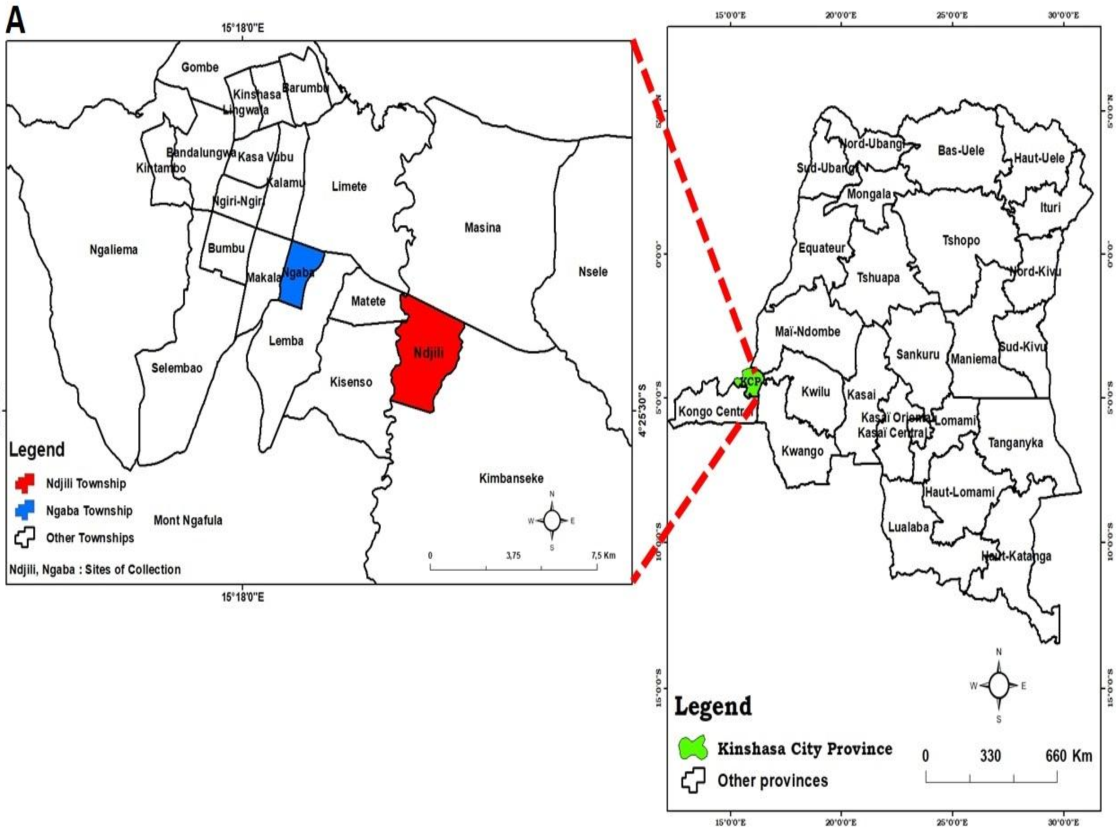
For the analysis of the potassium content, 25 mL of the extract of the mixture were then transferred to a test tube, 6 mL of distilled water and a packet of a specific reagent (potassium 3). It was left to homogenize and react for 1 hour. However, for the analysis of the phosphorus content, 2 mL of the extract of the mixture, 6 mL of distilled water and 2 mL of nitro-vanado-molybdate reagent were transferred to the test tube.

They were allowed to homogenize and react for 1 hour of the time. Finally, it is measured by UV-Vis spectrometry 768 nm for K and 430 nm for P. While the Total Kjeldahl N (TKN) were determined by titrimetry [4] (SI1E) and content in total organic carbon (TOC) were determined according to the method of Walkey and Black [30,31] (SI1F).

2.2.2 Chemical analysis and bioactive substances in leaves and stems of MU

Except saponins, all the active chemical groups in the aqueous extracts of leaves and stems were identified by qualitative colorimetry as describe in literature [4,32] (SI2).

The saponins were determined by a semi quantitative method based on the formation of persistent foam of at least 1 cm height during 15 minutes when vigorously agitating 5 mL of aqueous extracts in a test tube [4] (SI2).



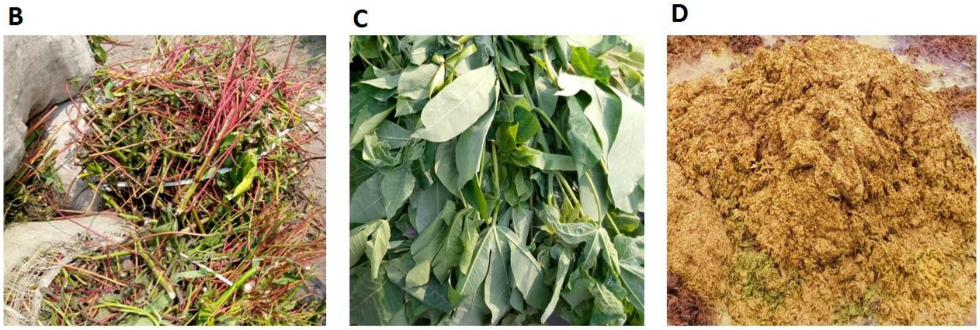


Fig. 1. Cartography of DR Congo, location of the KCP (in green) and the sites of collection (Ngaba: in blue, Ndjili: in red) (A), Stems of MU (B), Leaves of MU (C) and inoculum: Cow manure (D)

2.3 Biochemical Methane Potential (BMP) and Energetic Balance

The Biogas and methane yields assays of stems and leaves of MU were determined by following the procedure described by Rodriguez et al. [33] and Wang et al. [34] (SI3). The tests were carried out in triplicate in 600 mL bottles filled with 500 mL of a mixture. The mixture consisted of 500 mL of phosphate buffer solution (with the pH adjusted to 7.5 with NaOH 3 N), 1250 mg dry weight (DW) of CM as inoculum and 6670 mg DW milled leaves or stems, thereby represent a ratio of 28/1 of SV of the substrate and inoculum (5.6828 g SV of Leaves/ 0.2013 g SV of Inoculum) and a ratio of 30/1 (6.0697 g SV of stems/ 0.2013 g SV of Inoculum), for the leaves and stems of MU, respectively.

As positive control, we utilized 800 mg of cane sugar (CS, saccharose), 500 mL of buffer solution and 1250 mg DW of CM. The negative control sample consisted of 500 mL of buffer solution and 1250 mg DW of anaerobic sludge inoculum. No energetic substrate was added to the blank samples. Each test was performed in triplicate.

When the sample bottles were filled, they were capped tightly with rubber septa [4,35]. The bottles were then incubated at 25°C, and the composition and volume of biogas produced were periodically measured during 128 days, in according to the method of CO_2 absorption by KOH [4,36] (SI1).

Biogas or methane yield was calculated by dividing the biogas or methane volume (m^3) by the weight (kg) of sample VS added to each bottle [4,34,37]. The balance between the amount of stems or leaves produced annually in the KCP and the annual energetic potential of the biogas resulting from their AD was calculated from their BMP and the Lower Heating Value (LHV) of methane [16,38-40] (SI3).

2.4 Analysis of Digestates

After 128 days of AD, the digestates were separated in liquid and solid residues by centrifugation and filtration on 0.2 µm cellulose acetate membrane for TOC, TKN, P and K analyses [4] (SI1D-SI1F). The TOC and TKN were expressed relatively to the quantity of the digestates resulting from the inoculum, or from the total digestates (inoculum and substrates), or from the substrates alone by considering that the inoculum DW did not change after the digestion.

3. RESULTS AND DISCUSSION

3.1 Biogas and Methane Production

The evolution of the biogas production was monitored in different digesters in order to assess the AD of stems and leaves of MU. Fig. 2 shows the total cumulative volumes of biogas Fig. 2A produced and that of methane Fig. 2B obtained after purification of the biogas by the absorption of CO_2 by the KOH (SI1) versus time. Obviously, it is shown that no biogas production was detected for the blank samples or negative control (T**-**). After 128 days of AD, the total cumulative volume of biogas produced was observed to be 501.5 ± 8.5 mL for the positive control (T**+**), with a total volume of methane of 421.0 ± 13.0 mL. This corresponded to 84.0 % of the biogas produced. The detection/or non-detection of biogas for (T**+**)/or (T**-**), respectively, enabled us to highlight the methanogenic activity of inoculum as well as the methanogenic potential of substrate. More importantly the fact that the biogas was not detected in (T**-**) make us sure and confident that all the biogas were produced from the AD of substrates. The total cumulative volume of biogas produced were evaluated to be 1144.5 ± 40.0 mL and 921.0 ± 31.0 mL for stems and leaves of MU, respectively. The cumulative amount of methane was 946.0 ± 18.0 mL for the stems of MU. That represented about 83.0 % of methane in biogas. By contrast the amount of methane was 770.0 ± 16.0 mL for leaves of MU, representing about 84.0 % of methane in the biogas.

The delay observed before the biogas production starts could be attributed to the concentration of organic matter in the substrates (VS). Indeed, the biogas production in the digester containing higher concentration of organic matter like the stems rapidly produced (10 days) the biogas after inoculation, whereas the digester with lower content of organic matter lately produced (20-30 days) the biogas after incubation Fig. 2A, B; Table 1. Although the pH conditions were suitable in the different digesters, that is, between 6.5 and 7.2 [41-44].

The physical and chemical analysis of the substrates Table 1 showed that the MU stems contained lower amount of nitrogen, and high contents of carbon and mineral elements (K and P) compared to MU leaves. Moreover, the C/N ratio was in the optimal range (C/N: 20-30) for a good AD [37,38,45,46]. Biogas production containing 84% from cane sugar, which is free of nitrogen revealed that the inoculum contained nutrients capable to trigger the methanization.

Although the C/N ratio of the substrate has high influence on the AD process [37,38,45,46], we also noticed that the proportion of methane in the biogas depended not only on the C/N ratio of the substrates, but also on that of the total composition in the digester. This was proved by the fact that the methane yield recorded by cane sugar (0.526 m^3 of CH_4/ kg.VS) was around that theoretically expected (0.692 m^3 of CH_4/kg.VS) (SI3). This methane yield was achieved after 50 days of AD and stayed nearly steady after 128 days like that of the MU leaves Fig. 2.

The range of concentration of K required for its stimulating effect of the AD under mesophilic conditions is 200-400 mg/L [41,42]. The methanization of leaves of MU could be stimulated by the K because its concentration was determined to be 287 mg/L. The half maximal inhibitory concentration of K of the AD is IC_{50} ~ 2900 mg/L [41,42]. However, with a K content of about 1900 mg/L for the stems of MU, which is by far lower than the IC_{50}, the methanation of this substrate was not affected by the concentration this mineral. The same effect was observed for phosphorus.

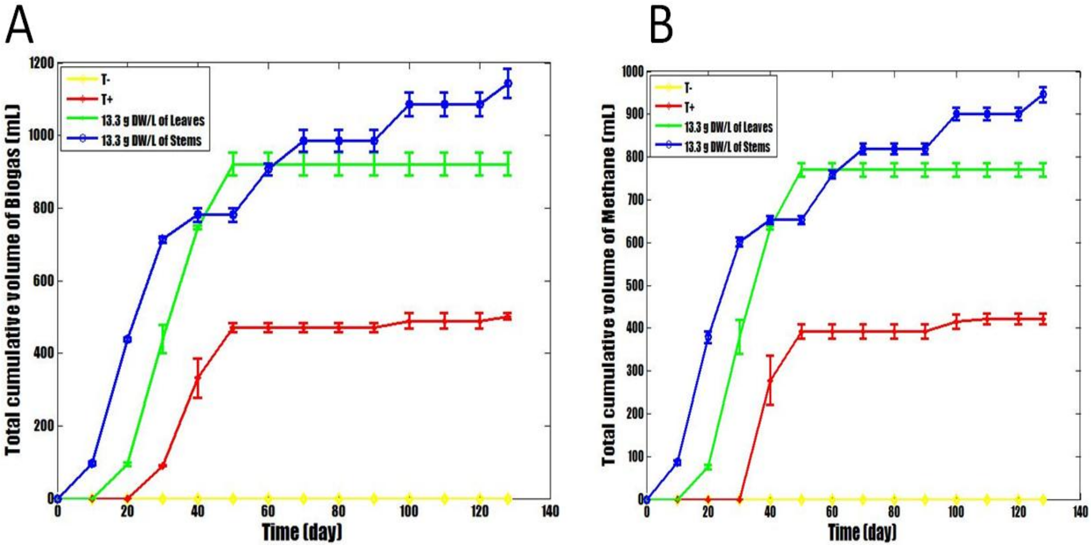


Fig. 2. Total cumulative volumes of biogas and methane produced from the AD of the leaves and stems of MU for different rectors versus time. (A) Produced biogas, and (B) Produced methane

Table 1. Physical and chemical characterization of the substrates and the inoculums

Components	Substrates		Inoculum
	MU leaves	MU stems	CM
Dry weight (%)	80.8 ± 0.0	17.1 ± 0.4	21.6 ± 0.2
Organic matter (% DW)	85.2 ± 0.3	91.0 ± 0.2	16.1 ± 0.4
TOC (mg/g DW)	357.5 ± 14.3	530.5 ± 15.4	486.4
TKN (mg/g DW)	50.5 ± 0.8	23.8 ± 03	26.1
P (mg/g DW)	2.2	180.2	27.6
K (mg/g DW)	21.5	142.3	10.5
C/N	7	22	19
C/N/P	163/23/1	227/10/75	180/9/10

The C/N or C/N/P ratio in the leaves of MU alone and in the overall composition of digester before AD were not in the range recommended (C/N: 20-30; C/P/N: 150/4/1) [37,38,45,47]. For the stems of MU, only the C/N ratio was in the range commonly recommended but not their C/N/P Tables 1, 2.

Due to their difficulty to solubilize induced by their composition rich in cellulosic fiber [20,48,49], the production of methane of the stems of MU continued to increase after 128 days of AD Fig. 2. This is justified that the TOC concentration of 10.9 mg/g for stems in their liquid residues compared to 73.3 mg/g for leaves Table 2.

The analysis of secondary bioactive metabolites or bioactive substances in the leaves and stems of MU showed the presence of saponins and catechic tannins Table 3. However, their presences did not affect the methanization of the leaves and stems. Indeed, it is known that some plants or their extracts with high concentrations of secondary bioactive metabolites such as saponins, tannins, essential oil, organosulphur compounds, flavonoids and many other metabolites have potential to inhibit methane production [4,50-52]. The methonogenesis is inhibited at concentrations of secondary bioactive metabolites above 0.3 g/L [4,51]. This behavior indicated that that concentrations of saponins and catechic tannins in the leaves and stems would be less than 0.3g/L.

The methane yields of our two substrates were therefore 0.156 ± 0.003 m^3 CH_4/kg VS and 0.136 ± 0.003 m^3 CH_4/kg VS for stems and leaves of MU respectively (SI3). These methane yields are in the range of those found by Amon Thomas et al. [35] and stipulated by Gunaseelam [53] for the vegetable wastes, whose the range is between 0.12 to 0.43 m^3 of CH_4/kg VS depending in the chemical composition of the substrate.

Table 2. C/N/P ratio and K quantity of different samples in the digester before and after AD

Substrates and inoculum	Before AD		After AD	
	C/N/P	K(mg)	C/N/P	K(mg)
Inoculum alone	186/10/11	13	186/10/11	13
Positive control	293/10/10	13	218/10/10	13
MU leaves	600/75/10	156	517/75/10	156
MU stems	216/10/65	962	187/10/65	962

Table 3. Bioactive substances in leaves and stems of MU

Components	leaves	stems
Saponins	+	+
Flavonoids	-	-
Alkaloids	-	-
Anthraquinones (bound quinones)	-	-
Catechic tannins	+	+
Gallic tannins	-	-
Anthocyanins	-	-
Leuco-anthocyanins	-	-

+ : sustrate contains the component - : sustrate do not contain the component

This difference of the biogas or methane yields for the stems and leaves could be due to the difference between their organic matter and C/N ratio.

The yields of the methane obtained par Mambanzulua et al. [4] after 100 days of AD of the leaves at the concentration of 13 g/L was 0.023 m^3 CH_4/g VS at 30°C. In this study, after 100 days of AD, the methane yield obtained in the same concentration leaves at 25°C was about 6-fold higher. That could be explained by the fact that methanogenic activity of CM used in this work is approximately 6-fold superior than that of the anaerobic sludge which came from in origin activated sludge. It is reported in the literature that the CM is more active than activated sludge which is inoculum from anaerobic reactor. The methanogenic activity of the CM is 3-13 mg CDO-CH_4/g VS by contrast that of the activated sludge is not exactly known. However, the latter is classified in the range of 1-2 mg CDO-CH_4/g VS and it depends on the features of the granular sludge, type of the substrate, environmental conditions, and the test procedure [25-27].

3.2 Energetic Potentials of Biogas

The calorific value of biogas is proportional to its methane content. This can be determined thanks to Lower Heating value (LHV) and the proportion of methane in the biogas produced [38-40]. The proportions of methane in the biogas produced from the stems and leaves of MU were 83% and 84%, respectively. These correspond to the calorific values evaluated at 7.820 kWh/m^3 and 7.914 kWh/m^3, respectively. These energy values found are within the range of the biogas calorific value reported in the literature (4.726 kWh/m^3 - 9.452 kWh/m^3) [16,38-40].

However, in KCP, the MU is the first most consumed vegetable and the second most planted [3,4,7,21]. KCP produces annually about 3 million tons of wastes. These wastes contain 1.65 million of organic wastes, which represents 55% [6]. Previous research has shown that these organic wastes are essentially composed (94%) of vegetable wastes of which the majority is leaves [3,4,7]. Considering that the latter consisted mainly of leaves or stems, the annual energetic potential of vegetable wastes produced in the KCP will therefore be estimated at 1.362 ± 0.028 10^9 kWh for leaves (SI4A) and 0.337 ± 0.006 10^9 kWh for the stems (SI4B). Knowing that the LHV of charcoal is about 8.229 kWh/kg [47,54,55], the annual energetic potential obtained would cover the energy needs for the KCP households corresponding to the use of charcoal, evaluated with 166 10^3 tons for the leaves, and with 41 10^3 tons of charcoal for the stems (SI4). The substitution of the use of charcoal by biogas in KCP households, will allow them to spare from the problems of air pollution which causes 4 million premature deaths each year in the world, with more than 600 000 in African countries following the use of solid biomass energy [56,57]. The Table 4 reports the energy amounts in the resulting biogas production from the AD of leaves and stems of MU during 100 and 128 days.

3.3 Fertilizing Potential of the Digestates

The fertilizing pontential of the digestates of leaves and stems of MU is primarily due to the availability of the mineral elements (N, P, K) previously retained in the complex structures of these wastes, owing to their mineralization through AD. The N, P and K concentrations in the liquid digestate for the stems were determined to be 2.1, 85.0 and 117.3 mg/L respectively. By contrast, the N, P and K concentrations in the liquid digestate for leaves were 13.7, 4.4 and 110.0 mg/L respectively.

We evaluated the fertilizing potential of the digestates utilizing their C/N ratio. We noticed that the C/N ratio of Leaves digestates has fallen to 5. The digestate with a C/N ratio of 5 is recognized to be favorable for vegetable crops and fruit trees soils [4,7,58,59]. The C/N ratio of the digestats resulting from the AD of stems was 10. A C/N ratio of 10 is considered optimal for the organisms, soil conditioning and could improve the soils hydraulic conductivity [4,58,60-62].

It has been shown that one can spread until 30 tons of dry matters of digestate by hectare per year on an acidic poor soil [3,59,60,62]. Therefore, with the 222 10^3 and 1.039 10^6 tons of digestates that we can produce from methanization of 1.55 10^6 tons of stems and leaves of MU, respectively, produced in Kinshasa per year, one could fertilize 7,400 hectares for stems and 39,966 hectares for leaves (SI5A, SI5B).

Table 4. Energy amounts in the resulting biogas production from the anaerobic digestion of leaves and stems of MU during 100 and 128 days

Samples concentrations	Methane yields for 100 days (L/g VS)	Methane yields for 128 days (L/g VS)	Energies for 100 days (kWh/g VS)	Energies for 128 days (kWh/g VS)
1.6 g CS/L	0.519 ± 0.020	0.526 ± 0.016	$4.888 \pm 0.188\ 10^{-3}$	$4.958 \pm 0.153\ 10^{-3}$
13.3 g leaves MU/L	0.136 ± 0.003	0.136 ± 0.003	$1.277 \pm 0.027\ 10^{-3}$	$1.277 \pm 0.027\ 10^{-3}$
13.3 g stems MU/L	0.148 ± 0.002	0.156 ± 0.003	$1.399 \pm 0.023\ 10^{-3}$	$1.468 \pm 0.028\ 10^{-3}$

4. CONCLUSION

In summary the present report investigated the energetic balance between the amount of leaves and stems of Manihot Utilissima (MU) annually produced in the KCP as well as the energetic potential of their biogas, by utilizing the CM as inoculum. This work was carried out at the ambient temperature of 25°C (under mesophilic conditions) favorable to tropical regions such as KCP, where leaves and stems of MU are among the most generated vegetable wastes.

The annual energetic potential of biogas produced from vegetable wastes of MU from the KCP would cover the energy needs for the KCP households corresponding to the use of charcoal. This energy is renewable and its net CO_2 contribution to the atmosphere is nil. Moreover, it could keep KCP away from deforestation by providing its households with a form of clean energy for cooking. The substitution of the use of charcoal by biogas in KCP households, will allow them to spare from the problems of air pollution which causes 4 million premature deaths each year in the world, with more than 600 000 in African countries following the use of solid biomass energy. In addition the fertilizing potential of digestates resulting from the anaerobic digestion of leaves and stems of MU were evaluated by their C/N ratios. The characteristics of this fertilizer show that not only they are favorable for the cultivation of vegetables and fruit trees on the KCP soils but also optimal for the organisms and the conditioning of the soil.

ACKNOWLEDGEMENTS

BM Nsimba thanks the laboratoire de bioprocedes and the Faculty of Pharmacy at the University of Kinshasa. BM Nsimba also thanks the Ministry of the environment of the KCP for providing the statistical data of OWs.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Khennas S. Understanding the political economy and key drivers of energy access in addressing national energy access priorities and policies: African Perspective. Energy Policy 2012;47:21-26.
2. Hanif I. Impact of economic growth, nonrenewable and renewable energy consumption, and urbanization on carbon emissions in Sub-Saharan Africa. Environmental Science and Pollution Research, Springer-Verlag GmbH, Germany; 2018. Available:https://doi.org/10.1007/s11356-018-1753-4
3. Mambanzulua PN, Hiligsmann S, Sumbu EZ, Ongena M, Thonart P. Potentiel d'élimination des déchets végétaux (feuilles de Mangifera Indicat et de Manihot Utilissima) par méthanisation à Kinshasa (République Démocratique du Congo). Vertigo : La revue électronique en sciences de l'environnement. 2015a;15(1).
4. Mambanzulua PN, Hiligsman S, Sumbu EZ, Culot M, Fievez T. Comparative study of the methane production based on the chemical composition of *Mangifera Indica* and manihot utilissima leaves. Springer. 2015b;4:75. Available:https://doi.org/10.1186/s40064-015-082-y
5. Barrios S, Bertinelli L, Strobl E. Climatic change and rural-urban migration: The case of sub-Saharan Africa. J Urban Econ. 2006;60(3):357-371.
6. Ministère Provincial d'Environnement. 2ième table ronde sur l'assainissement

dans la Ville de Kinshasa. Kinshasa, DR. Congo; 2019.
Available:https://www.7sur7.cd/index.php/2019/10/17/rdc-kinshasa-produit-3-millions-de-tonnes-des-dechets-par-soit-plus-de-8-mille-tjr.
(Accessed 20.06.2020)

7. Mulaji CK. Composts et sols acides en milieu tropical humide (Kinshasa, R.D. Congo). Editions Universitaires Européennes, Sarrebruck, Allemagne; 2017.
8. Chandra R, Takeuchi H, Hasegawa T. Methane production from lignocellulosic agricultural crop wastes. Renew Sust Energ Rev. 2012;16:1462-1476.
9. Surendra KC, Takara D, Hashimoto AG, Khanal SK. Biogas as a sustainable energy source for developing countries: Opportunities and challenges. Renew Sust Energ Rev. 2014;31: 846-859.
10. Barakat A, Monlau F, Steyer JP, Carrere H. Effect of lignin derived and furan compounds found in lignocellulosic hydrolysates on biomethane production. Biores Technol. 2012;104:90-99.
11. Chaaban MA. Hazardous waste source reduction in materials and processing technologies. J Mater Process. 2001;119:336-343.
12. Oumar A, Benhabyles L, Igoud S. Energetic valorization of biomethane produced from cow-dung. Procedia Eng. 2012;33:330-334.
13. Malta–alvarez J, Macé S, Llabrés P. Anaerobic digestion of solid wastes-an overview of research achievements and perspectives. Biores Technol. 2000;74:3-16.
14. Ward AJ, Hobbs PJ, Holliman PJ, Jones DL. Optimization of the anaerobic digestion of cultural resources. Biores Technol. 2008;99:7928-7940.
15. Wyman CE, Goodman BJ. Biotechnology for production of fuels, chemicals, and materials from biomass. Appl Biochem Biotechnol. 1993;39:41.
16. Li J, Huang H, Huhetaoli, Osaka Y, Bai Y, Kobayashi N. Combustion and heat release characteristics of biogas under hydrogen-and oxygen-enriched condition. Energies. 2017;10(8):1200.
17. Galvagno A, Chiodo V, Urbani F, Ferni F. Biogas as hydrogen source for fuel cell applications. Int J Hydrog Energy. 2007;38:3913-3920.
18. Shan X, Qian Y, Zhu L, Lu X. Effects of EGR rate and hydrogen/carbon monoxide ratio on combustion and emission characteristics of biogas/diesel dual fuel combustion engine. Fuel. 2016;181:1050-1057.
19. Koszel M, Lorencowicz E. Agriculture use of biogas digestate as replacement fertilizers. Agric Agric Sci Procedia. 2015;7:119-124.
20. Han M, Kim Y, Kim Y, Chung B, Choi GW. Biomethanol production from optimized pretreatment of cassava stem. Korean J Chem Eng. 2011;28 (1):119-125.
21. Bell A, Mück O, Schuler B. Les richesses du sol, les plantes à racines et tubercules en Afrique: une contribution au développement des technologies de récoltes et d'après récolte. Deutsche Stiftung fur internationale Entwicklung (DSE), Deutsche Gesellschaft für Technische Zusammenarbeit (GTZ), Feldafing, Allemagne, ISBN. 2000;3-934068-17-0.
22. Chynoweth DP, Turick CE, Owens JM, Jerger DE. Biochemical methane potential of biomass and waste feedstocks. Biomass Bioenergy. 1993;5(1):95-111.
23. Rodriguez-Chiang ML, Dahl OP. Effect of inoculum to substrate ratio on the methane potential of microcrystalline cellulose production wastewater. BioResources. 2015;10(1):898-911.
24. Lawal AA, Dzivama AU, Wasinda MK. Effect of inoculum to the substrate ratio on biogas production of sheep paunch manure. Res Agr Eng. 2016;62(1):8-14.
25. Hussain A, Dubey SK. Specific methanogenic activity test for anaerobic degradation of influents. Applied Water Sci. 2017;7(2):535-542.
26. Ashekuzzaman SM, Poulsen GT. Optimizing feed composition for improved methane yield during anaerobic digestion of cow manure based waste mixtures, Biores Technol. 2011;102(3):2213-2218.
27. Shin HS, Han SK, Song YC, Lee CY. Performance of UASB reactor treating leachate from acidogenic fermenter in the two-phase anaerobic digestion of food waste. Water Res. 2001;35(14);3441-3447.
28. APHA. Standard methods for the examination of water and waste water: method 4500 P-E- Heteropoly blue Method, 18th ed., Washington; 1992.
29. Liu K. Effect of sample size, dry ashing temperature and duration on

determination of ash content in algae and other biomass. Algal Res. 2019;40:101486.
Available:https://doi.org/10.1016/j.alga.2019.101486
30. Black CA, Evan DD. Methods of soil analysis, American Society of Agronomy Part 2. Agronomy. 1965;9:917-918.
31. De Vos B, Lettens S, Muys B, Deckers JA. Walkley-Black analysis of forest soil organic carbon: Recovery, limitations and uncertainty. Soil Use Manag. 2007;23:221-229.
32. Wagner H, Bladt S. Plant drug analysis. Springler Verglag, Berlin; 1966.
33. Rodriguez C, Hiligsmann S, Ongena M, Charlier R, Thonart P. Development of an enzymatic assay for the determination of cellulose bioavailability in municipal solid waste. Biodegradation 2005;16:415-422.
34. Wang YS, Byrd CS, Barlaz MA. Anaerobic biodegradability of cellulose and hemicellulose in excavated refuse samples using a biochemical methane potential assay. J Ind Microbiol. 1994;13:147-153.
35. Amon T, Amon B, Kryvoruchko V, Zollitsch W, Mayer K, Gruber L. Biogas production from maize and dairy cattle manure-Influence of biomass composition on the methane yield. Agric Ecosyst Environ. 2006;118(1):173-182.
36. Hiligsmann S, Masset J, Hamilton C, Beckers L, Thonart P. Comparative study of biological hydrogen production by pure strains and consortia of facultative and strict anaerobic bacteria. Biores Technol. 2011;102:3810–3812.
37. Banks JC, Heaven S. Optimisation of biogas yields from anaerobic digestion by feedstock type: The Biogas Handbook. Woodhead Publishing Limited; 2013.
Available:https://doi.org/10.1533/9780857097415.1.131
38. Borja R, Rincón B. Biogas production. Reference Module in Life Sciences; 2017.
Available:https://doi.org/10.1016/B978-0-12-809633-8.09105-6
39. Chen Y, Chen JJ, Creamer KS. Inhibition of anaerobic digestion process: A review. Biores Technol. 2008;99(10):4044-4064.
40. Abbasi T, Tauseef SM, Abbasi SA. Biogas energy. SpringerBriefs in Environmental Science 2; 2012.
Available:https://doi.org/10.1007/978-1-4614-1040-9
41. Appels L, Baeyens J, Degrève J, Dewil R. Principles and potential of the anaerobic digestion of waste-activated sluge. Prog Energy Combust Sci. 2008;34:755-781.
42. Turovskiy IS, Mathai PK. Wastewater sludge processing. Wiley, New York; 2006.
43. Zhang C, Su H, Tan T. Batch and semi-continuous anaerobic digestion of food waste in a dual solid-liquid system. Biores Technol. 2013;145:10-16.
44. Peng X, Börner RA, Nges IA, Liu J. Impact of bioaugmentation on biochemical methane potential for wheat straw with addition of *Clostridium cellulolyticum*. Biores Technol. 2014;152:567-571.
45. Yen HW, Brune DE. Anaerobic co-digestion of algal sludge and waste paper to produce methane. Biores Technol. 2007;98:130-134.
46. Akunna JC. Anaerobic treatment of brewery wastes. Brewing Microbiology; 2015.
Available:https://doi.org/10.1016/B978-1-78242-331-7.00019-8
47. Solarte-Toro JC, Chacon-Pérez Y, Cardona-Alzate CA. Evaluation of biogas and syngas as energy vector for heat and power generation using lignocellulosic biomass as raw material. Electron J Biotechnol. 2018;33:52-62.
48. Reed JD, McDowell RTE, Van Soest PJ, Horvath PRJ. Condensed tannins: A factor limiting the use of cassava forage. Journal of the Science of Food and Agriculture. 1982;36(6):433-441.
49. Klinpratoom B, Ontanee A, Ruangviriyachai C. Improvement of cassava stems hydrolysis by two-satge chemical pretreatment for high yield cellulosic ethanol production. Korean J Chem Eng. 2015;32(3):413-423.
50. Patra AK, Saxena J. A new perspective on the use of plant secondary metabolites to inhibit methanogenesis in the rumen. Phytochemistry. 2010;71:1198-1222.
51. Mambanzulua PN, Hiligsmann S, Sumbu EZ, Ongena M, Thonart P. Impact of different plant secondary metabolites addition: saponin, tannic acid, salicin and aloin an glucose co-digestion. Ferment Technol. 2015c;4:113.
Available :https://doi.org/10.4172/2167-7972.1000113
52. Beauchemin KA, Kreuzer M, O'Mara F, McAllister TA. Nutritional management for enteric methane abatement: a review. Aust J Exp Agric. 2008;48:21-27.

53. Gunaseelan VN. Biochemical methane potential of fruits and vegetable solid waste feedstocks. Biomass Bioenerg. 2004;26(4):389-399.
54. Jenkins RG. Thermal gasification of biomass- A primer. Bioenergy. 2015;261-286.
55. Gary CY. Municipal solid waste to energy conversion process. Willey & Sons. 2010.
56. Ifegbesan AP, Rampeli IT, Annegarn HJ. Nigerian households' cooking energy use, determinants of choice, and some implications for human health and environmental sustainability. Habits International. 2016;1-8.
    Available:https://doi.org/10.1016/j.habitatint.2016.02.001
57. World Health Organisation (WHO). Household air pollution and health; 2018.
    Available:http://www.who.int/news-room/fact-sheets/detail/household-air-pollution and health (10/07/2020)
58. Hawke RM, Summers SA. Land application of farm dairy effluent: Results from a case study, Wairarapa, New Zealand. New Zeal J Agr Res. 2003;46:339-346.
59. Glowacka A, Szostak B, Klebaniuk R. Effect of biogas digestate and mineral fertilization on the soil properties and yield and nutritional value of switchgrass forage. Agronomy. 2020;10:490.
    Available:https://doi.org/10.3390/agronomy10040490
60. Wrap. Digestate and compost use in agriculture: Good practice guidance. United Kingdom; 2016.
    Available:https://www.wrap.org.uk/using-renewable-fertilisers
    (Accessed 02.06.2020).
61. Lessard P, Bihan YL. Fixed film processes. Handbook of Water and Wastewater Microbiology. 2003;317-336.
62. Sparling GP, Williamson JC, Magesan GN, Schipper LA, Lloyd-jones RH. Hydraulic conductivity in soils irrigated with wastewaters of differing strengths: Field and laboratory studies. Aust J Soil Res. 1999;37:391-402.

Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sdiarticle4.com/review-history/64400