Enhancement of biogas potential of primary sludge by co-digestion with cow manure and brewery sludge

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Abstract: Anaerobic digestion (AD) has long been used to treat different types of organic wastes especially in the developed world.   However, organic wastes are still more often considered as a waste instead of a resource in the developing world, which contributes to environmental pollution arising from their disposal.   This study has been conducted at Bugolobi Sewage Treatment Plant (BSTP), where two organic wastes, cow manure and brewery sludge were co-digested with primary sludge in different proportions.   This study was done in lab-scale reactors at mesophilic temperature and sludge retention time of 20 d. The main objective was to evaluate the biodegradability of primary sludge generated at BSTP, Kampala, Uganda and enhance its ability of biogas production.   When the brewery sludge was added to primary STP sludge at all proportions, the biogas production rate increased by a factor of 3.   This was significantly ($p<0.001$) higher than observed gas yield (337 ± 18) mL/(L·d)) in the control treatment containing (only STP sludge).   Co-digesting STP sludge with cow manure did not show different results compared to the control treatment.   In conclusion, Bugolobi STP sludge is poorly anaerobically degradable with low biogas production but co-digestion with brewery sludge enhanced the biogas production rate, while co-digestion with cow manure was not beneficial.

Keywords: wastewater treatment, co-digestion, cow manure, brewery, primary sludge, biogas, energy recovery

1   Introduction

The Bugolobi Sewage Treatment Plant (BSTP) located in Kampala is the largest sewage treatment plant (STP) in Uganda.   It was designed to treat 33 000 m³/d of wastewater but it only receives an average flow of 12 000 m³/d.   The plant treats sewage using a coarse and fine screen, a detritus basin, two settling tanks in parallel, followed by trickling filters and finally by clarifiers. The sludge from the plant is left to stabilize in open semi-anaerobic digesters before being sent to a set of drying beds and later sold as dry organic fertilizer.   The plant, which has been in existence since the late 60 s, is quite dilapidated and releases biogas.   The gas is generated at the open semi-anaerobic tanks where sludge is left to stabilize.   This contributes to greenhouse gas emissions and odor nuisance to the surrounding areas.

Fortunately, the old plant is already in the process of replaced by a new one, which will have similar treatment processes but whose sludge will undergo further treatment by anaerobic digestion. Despite the fact that a new treatment plant will be constructed, information on the performance of Kampala primary sewage sludge with regard to biogas production and potential for co-digestion with other wastes is not available. This provided an opportunity to cover up the information gap. Furthermore, there are a number of abattoirs in Kampala city; the wastes of abattoirs have become an environmental threat because most of them discharge untreated wastewater in the nearby Nakivubo Channel, reaching Lake Victoria. Also, a nearby brewery plant is in need of economical disposal method for brewery waste. Co-digestion of sewage sludge with substrates not only enriches the operation and optimize processes of the new plant, but also could improve the environmental quality of the Northern shores of Lake Victoria.

Anaerobic digestion (AD) has been used for stabilizing organic matter (sewage sludge, cow manure, etc.). AD sludge also has been applied in biogas production increasingly[1,2]. The biogas could be considered as a valuable source of energy and electricity. Substantial research has been optimized the AD process to increase biogas production, which led to studies aim at improving reactor design, optimizing AD process parameters and manipulation of substrates[2-5]. Meanwhile, AD has been expanded into other wastes, such as energy crops, fats and kitchen wastes. Substrate-focused AD optimization considers the selection of suitable substrates and their combinations[6-8] as well as nutrient availability[9], and pre-treatment of the substrates to make them more amendable for AD[10-15]. While substrate manipulation may improve the AD process, some challenges still remain due to the different limitations associated with the properties of different substrate[7,16]. Therefore, continued studies are imperative to further establish the best designs, environment and substrate mixtures to optimise biogas production.

The present study was aimed at evaluating the biodegradability of primary sludge generated at Bugolobi STP. It further sought to explore the possibility of optimizing biogas recovery by means of co-digestion of the primary sludge with cow manure and brewery waste in different proportions.

2 Materials and methods

2.1 Substrates for co-digestion

Three different feed stocks, primary STP sludge (STP sludge), cow manure (CM) and brewery waste (BW) were mixed in different proportions and used for AD. STP sludge was collected from the primary settling tanks at Bugolobi STP in Kampala, Uganda. Fresh cow manure was collected from the Makerere University farm in Kampala. Deionized water was diluted to the cow manure to reduce its dry matter content, made it easier to pour. Brewery waste was collected from East African Brewery Limited (EABL). The substrate was prepared such that primary STP sludge was mixed with cow manure and brewery sludge in different proportions, and were labelled as follows; S_0 (100% STP sludge), S_1 (75% STP sludge and 25% CM), S_2 (50% STP sludge and 50% CM), S_3 (75% STP sludge and 25% BW), S_4 (50% STP sludge and 50% BW), S_5 (50% STP sludge, 25% CM and 25% BW) and S_6 (100% BW). The ratios were selected to have at least 50% STP sludge in each substrate mixture since in normal operations of the digester; priority would be given to STP sludge treatment.

2.2 Experimental set-up

At laboratory scale, the experiments to determine the biodegradability and digestibility of STP sludge, brewery sludge and cow manure mixtures, which were set up at using glass bottles with a total volume of 1 L as anaerobic reactors. Seven anaerobic reactors, each filled with 700 mL of anaerobic inoculum sludge obtained from the EABL UASB wastewater treatment plant in Kampala (Uganda), were incubated at mesophilic conditions (36±1)°C. The inoculum sludge was initially diluted in a ratio of 1:1. Each of continuously stirred tank reactors (CSTR) was fed with seven different substrates (S_0, S_1, S_2, S_3, S_4, S_5 and S_6). The anaerobic reactors were operated for 72 d. During the start-up period, the daily organic loading rate (OLR) was started at 0.71 g COD/(L·d) and it was gradually increased until the desired sludge

retention time (SRT) of 20 d was reached. Each reactor was performed in duplicate and the average results were reported.

2.3   Analytical techniques

2.3.1   Characteristics of the inoculum sludge and substrate

Samples were taken from the substrates and inoculum, total phosphates (TP), chemical oxygen demand (COD) and total ammonium nitrogen (TAN) were determined using a HACH DR 5 000 Spectrometer as described in standard methods[17].   The pH value was measured with a Toledo pH meter.   Volatile solids (VS) and total solids (TS) were also analyzed according to standard methods[17].

2.3.2   Gas and pH monitoring

The biogas was captured in 2 000 mL plastic transparent measuring cylinders.   The cylinders were inverted in a basin with water and HCl (pH<4.3) to avoid the dissolution of CO_2.   Air tight plastic tubing from each reactor was connected to an inverted cylinder.   To enable direct measurement of the gas produced, the columns were graduated with volume markings and the volume of gas produced deduced from the displaced liquid volume within the columns.   To enable a quick identification of potential changes in the acidic condition of the solution within the columns, this solution was treated with methyl-orange indicator.   Biogas production and pH in the reactors were monitored on a daily basis for 72 d.   To determine the biogas composition, the gas was collected in gas bags from each reactor on two different days after SRT of 20 d was reached.   The samples were then taken to the College of Engineering, Design, Art, and Technology, Makerere University for analysis.   The gas analyzer (Model GC 2000 PLUS) was then used to determine the CH_4 and CO_2 percentage in the biogas. The average of the two measurements is reported.

2.3.3   Statistical methods

Analysis of variance was performed using SPSS, originally a parametric test was tried but the normality assumptions were not fulfilled even after performing a square root, inverse and logarithmic transformations. Therefore, the Kruskal-Wallis Ranks non parametric test was used to verify if there was no difference between the measured gas yield and production rates from the different substrates.   A significance level of 0.05 was used.

2.3.4   Effluent sludge characteristics

Samples of the effluent from the anaerobic reactors were collected and analyzed on a weekly basis for TS, VS, COD, TP and TAN.

2.4   Energy equivalents and conversion factors

During anaerobic digestion, the biodegradable organics are transformed into CH_4 and CO_2 and new microbial biomass.   It is estimated that from the AD of 1 kg sludge COD, 0.5 kg COD is converted to biogas, while the residual non-biodegradable matter (0.4 kg) and the new anaerobic biomass (0.1 kg) are exported with the effluent slurry[18].   As a rule of thumb, 1 kg of COD converted yields about 0.5 m^3 of biogas, and the latter, when converted in a combined heat and power module yields 1 kW·h of electricity (el) and 3 kW·h of heat energy.

3   Results and analyses

3.1   Feed characteristics

Selected characteristics of the raw STP sludge, CM, BW and the inoculum are shown in Table 1.   BW was slightly acidic with pH of 4.4, while the pH in the STP sludge, CM and the inoculum was at neutral values of 7.2, 6.8 and 7.0, respectively.   In the feed mixtures S_1, S_2, S_3, S_4 and S_5, the pH values were 7.1, 7.0, 6.5, 5.5 and 6.2, respectively.   TAN was the highest in the CM while COD and TP were the highest in the brewery waste.

Table 1   Parameters of the primary STP sludge, brewery waste, cow manure and the inoculum

Parameter	Inoculum	STP-sludge	BW	CM
COD/g·kg^{-1} (w.b.)	10	48	150	61
TS/g·kg^{-1} (w.b.)	14	31	62	40
VS/g·kg^{-1} (w.b.)	12	16	48	29
TAN/mg·kg^{-1} (w.b.)	48	92	67	160
TP/mg·kg^{-} (w.b.)	238	299	655	346
pH	7.0	7.2	4.4	6.8

Note: w.b.: wet base.

3.2   Operational parameters of the different reactors during stable operation at SRT of 20 d

The operational parameters measured at SRT of 20 d are shown in Table 2.   The average pH ranged between 7.0±0.2 and 7.4±0.1 for the reactors.   On a few

occasions, the pH values of digesters with substrates S_3, S_4 and S_5 decreased below 7.0, reaching minimum pH values of 6.5, 6.3 and 6.9 respectively. In such occurrences, 0.1 N mol NaOH was used to correct the pH value to a range of 7.0-7.6. The digester with substrate S_4 required more frequent pH adjustment than the other reactors. The pH value in the rector that received 100% BW, was maintained between 6.3-7.3, until the OLR exceeded 5.3 g COD/(L·d) and it subsequently reached a value of 5.5. It was not possible to maintain the pH value above 7, in this reactor after that, even with the addition of 0.1 N mol NaOH. Hence, it failed at SRT of 28 d.

The average pH at SRT of 20 d for all digesters (except 100% BW was used) was in the proper range required for efficient AD as indicated in Table 2. The generally accepted range for good process efficiency is 6.5-7.6[19]. This indicated an adequate buffering capacity, as well as stable operation for the anaerobic reactors receiving substrates S_3, S_4 and S_5 that had an initial pH value below 7.0. The reactor with S_6 also had an initial pH value below 7.0 but failed before reaching SRT of 20 d, due to organic overloading. The other three digesters (S_0, S_1 and S_2) had a constant pH value ranging between 7.0-7.6 throughout the entire experimental period of 72 d.

The loading rate was increased slowly from 0.71 g COD/(L·d), and was maintained at a value of 2.0 for S_0, 2.5 for S_1, 2.7 for S_2, 3.7 for S_3, 4.9 for S_4 and 3.8 g COD/(L·d) for S_5 at SRT of 20 d. At an organic loading rate of 5.3 g COD/(L·d) and SRT of 28 d, the reactor with 100% BW completely failed (data not shown). Overloading during anaerobic digestion can disrupt the operational stability of the digester. Increased loading rates may cause an accumulation of fatty acids which consequently causes the pH to drop to conditions which can inhibit methanogenic activity[2,20]. This implied that the loading rates at a STR of 20 d in the digesters with S_1, S_2, S_3, S_4 and S_5 did not generate residual levels of VFA that could limit the methanogenic activity.

Table 2   Operational parameters at SRT of 20 d for the 6 digesters (S_0 to S_5), that reached a stable performance, S_6 is not shown as it failed before reaching SRT of 20 d

Parameter	STP-sludge (S_0)	75% STP : 25% CM mix (S_1)	50% STP : 50% CM mix (S_2)	75% STP : 25% BW mix (S_3)	50 % STP : 50% BW mix (S_4)	50 % STP : 25% BW : 25% CM mix (S_5)
Weight influent/(g·L^{-1}·d^{-1})	50	50	50	50	50	50
SRT=HRT /d	20	20	20	20	20	20
OLR/g COD·(L·d)$^{-1}$	2	2.5	2.7	3.7	4.9	3.8
OLR/g VS·(L·d)$^{-1}$	0.8	1.0	1.1	1.2	1.6	1.4
Average Biogas yield ± SD /(mL·g^{-1} COD)	169±10	174±12	153±10	316±19	398±32	331±23
Average Biogas yield ± SD /(mL·g^{-1} VS)	297±17	304±21	264±15	677±50	851±71	629±44
Average Biogas production rate ± SD /(mL·L^{-1}·d^{-1})	337±18	435±12	414±24	1169±70	1952±155	1259±88
pH ± SD	7.4±0.1	7.3±0.1	7.2±0.0	7.3±0.3	7.0±0.2	7.3±0.2

3.3   Biogas yield and biogas production rate

The daily biogas production was monitored by keeping record of the increase in the gas columns on a two day basis. The biogas yield (Figure 1a) and the biogas production rate (Figure 1b) were derived from the daily gas readings as established from each digester.

From these results, it can be noted that 100% STP sludge has a low biogas yield and biogas production rate. The average biogas yield and production rate in the control digester of S_0 after a steady state SRT of 20 d was reached, were (160±10) mL/g COD and (337±18) mL/(L·d), indicated that biodegradability was quite low. The STP sludge had a CH_4 yield of 0.12 m^3/kg VS fed, which is less than the range estimated by Zhao and Viraraghavan[21] for primary and secondary sludge (0.24-1.01 m^3/kg VS fed) and those reported by Sommer et al.[22], for sewage sludge (0.28-0.32 m^3/kg VS fed). Also, Parkin and Owen[13] estimated the standard CH_4 yield from primary sludge at SRT of 20 d at a value of 643 mL/g VS fed. This is much higher than the CH_4 yield of 122 mL/g VS fed observed from Bugolobi STP sludge.

Primary sludge is usually composed of natural fibres, fats and other solids that settle in the primary clarifier of a wastewater treatment plant, and in contrast to waste activated sludge (WAS), it normally displays a relatively high biodegradability[19]. The results from our study

indicate that the primary sewage sludge at Bugolobi STP is poorly anaerobically digestible. The reason for the poor digestibility was not determined in this study, but it is suspected to be due to factors, such as long travel times to the treatment plant. The long sewage pipe distance (average of 12 km and 100 m manhole spacings) and the high temperatures (about 24°C), favor growth of sulphate reducing bacteria (SRB). Otherwise, the SRB consume the organic matter which could be converted to biogas[2]. The long travel time also encourage degradation before digestion given the high temperatures. Another factor could be due to heavy metal contamination that may originate from illegal disposal of industrial wastewater into the domestic sewer network.

a. Biogas yield

b. Biogas production rate
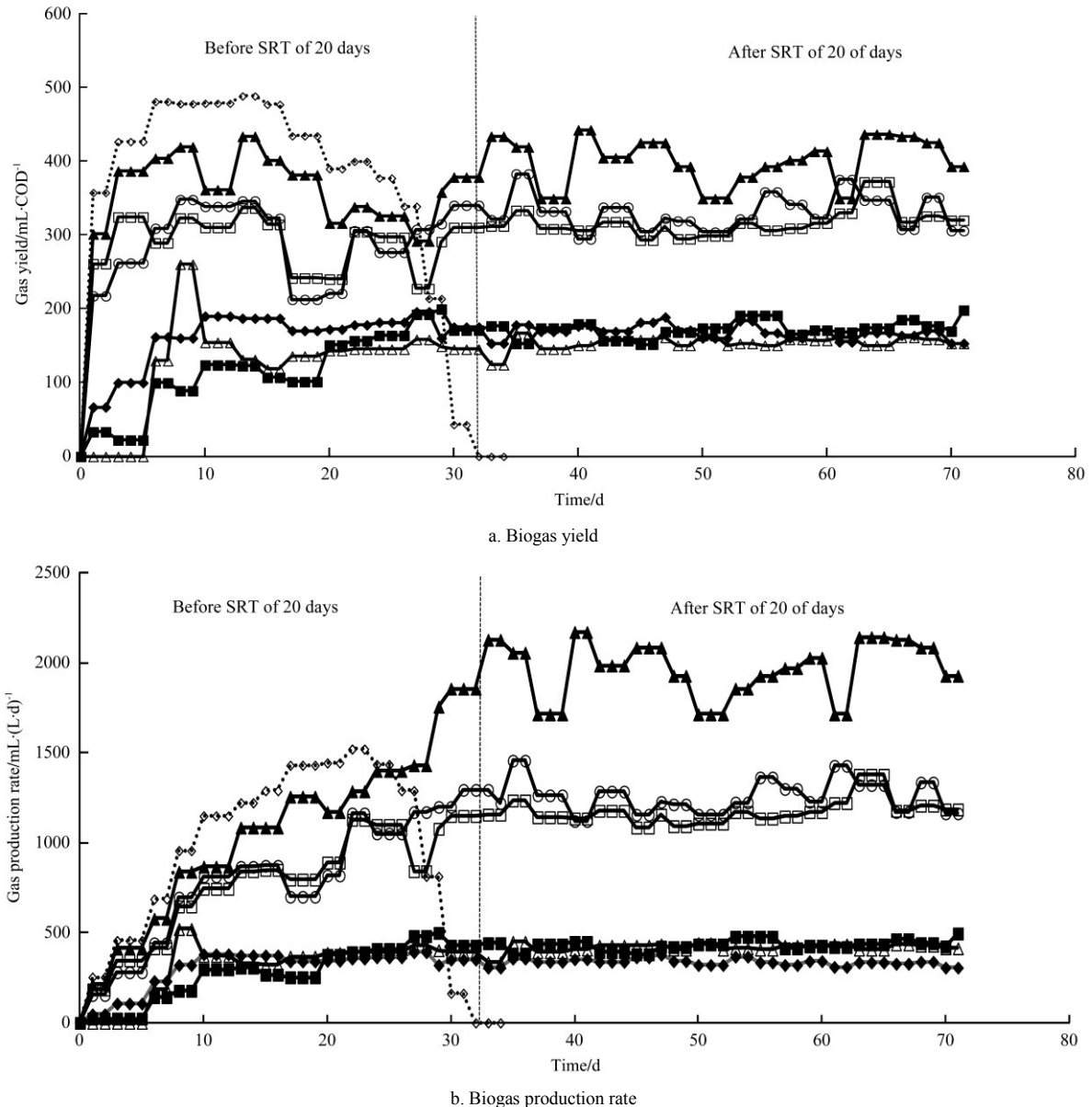
Note: (◆) 100% STP sludge, (■) 75% STP sludge and 25% CM, (△) 50% STP sludge and 50% CM, (□) 75% STP and 25% BW, (▲) 50% STP sludge and 50% BW, (○) 50 % STP sludge, 25% CM and 25% BW, (◇) 100% BW

Figure 1   Biogas yield and biogas production rate during the entire digestion period

This study further showed that co-digesting STP sludge with BW under mesophilic conditions enhanced both biogas production rate and biogas yield. The biogas production rate as well as the biogas yield, increased significantly ($p<0.001$) when BW was mixed with STP sludge. In general, both the biogas production rate and yields were observed to increase with an increasing ratio of BW/STP sludge. However, when the ratio was increased to 100% BW, the digester failed due to organic overloading (data not shown). The biogas yield for S_4 (50% STP sludge and 50% BW) showed a significantly higher ($p<0.001$) average biogas yield of

(398±32) mL/g COD compared to (316±19) mL/g COD for S_3 (75% STP sludge and 25% BW) and of (331±23) mL/g COD for S_5 (50% STP sludge, 25% BW and 25% CM). Similarly, the highest average biogas production rate was in S_4 at (1952±155) L/(L·d), followed by S_5 and S_3 at (259±88) L/(L·d) and (1169±70) mL/(L·d), respectively. Our results showed similar trends with those reported by Barbel et al.[5] who observed higher biogas production with an increasing brewery: sewage sludge ratio in the substrate during co-digestion. Meanwhile, Li et al.[23] observed increased biogas production when BW was co-digested with cattle slurry compared to cattle slurry alone, which is similar to our study. In the substrate with 25% CM, 50% STP sludge and 25% BW, the biogas yield was significantly higher ($p<0.001$) than when STP sludge was digested with CM alone (Table 2). In general, organic components in BW are easily biodegradable since they largely consist of sugars, soluble starch, ethanol and volatile fatty acids, which explain the observed increased biogas production when brewery was added as a co-substrate.

Moreover, co-digestion of STP with CM alone could not improve biogas production. The biogas yield for S_1 and S_2 were (174±12) mL/g COD and (153±10) mL/g COD, respectively. Statistical tests show that the biogas yields between S_0, S_1 and S_2 were not significantly different ($p=0.05$). CH_4 yields showed similar trends, a CH_4 yield of 0.1 m³/kg VS fed was observed in both digesters which substrates consisted of CM and STP sludge. This is within the range of the lower limit of 0.11-0.24 m³/kg VS fed, as observed by Hansen et al.[24] and Sommer et al.[22] when CM was digested. CM is more difficult to digest as compared to other animal manure (e.g. swine manure). Its low digestibility could be attributed to the presence of recalcitrant compounds, such as cellulose and hemicelluloses complexes with lignin[21]. Since CM originates from the rumen where it is already partially digested[21], it is likely to lead to lower biogas yields, compared to other wastes that are directly generated without prior digestion. However, Li et al.[23] have reported values up to 0.328 m³/kg VS fed of CH_4 when dry cow manure was co-digested with wastewater in batch experiments. This may be due to the manure characteristics which may vary depending on the animal species or difference in the animal feed as well as difference in manure management practices[25]. This variability consequently leads to variation of CH_4 production during AD.

3.4 Biogas quality

The average CH_4 content in biogas in the reactors treating substrates with BW was higher, i.e. 64.1%, 58.3% and 52.6% for S_3, S_4 and S_5, respectively. The biogas production in S_0 (100% STP sludge) showed the lowest quality with only 40.9% of CH_4, followed by S_1 and S_2 were STP sludge was mixed with CM. The biogas from S_1 and S_2 had CH_4 content of 44.7% and 47.5%, respectively. The CO_2 content in the samples was in the range of 30%-48%. Traces of CO and H_2S were also measured. H_2S is produced during hydrolysis when certain organisms break down the essential amino acid methionine[21].

The CH_4 content observed in this study was generally quite low compared to other studies[24,26,27]. CH_4 percentages above 70% were reported when sewage sludge was co-digested with brewery sludge ratios similar to our study at SRT of 20 d during biochemical methane potential (BMP) tests[26]. However, the same study reported CH_4 percentages below 30% for sewage sludge alone at SRT of 20 d, which was attributed to existence of heavy metals in the sewage sludge. Davidson et al.[28], Li et al.[23] and Martinez et al.[29] observed CH_4 content of 60% and more at SRT of 21 d for sewage sludge. Li et al.[23] also reported a CH_4 content of at least 50% for cow manure co-digested with sewage sludge. In CSTR systems, SRT of 20 d or more are recommended in order to avoid washout of the methanogens, which are responsible for CH_4 production[2]. While the above mentioned studies achieved higher CH_4 contents at SRT of 20 d, it is still possible that the same SRT of 20 d in our study was not sufficient to avoid washout of some methanogens. The low CH_4 level observed when sewage sludge alone was digested could also be due to the inoculum sludge was BW. This may not be favourable for digestion of sewage sludge and may require a longer SRT.

3.5   TAN concentration in the digesters

The concentration of TAN increased slightly in all digesters over the experimental period of 72 d.   The concentrations of TAN in the control digester with S_0 increased from an initial value of 230 to 253 mg/L, for S_1 from 205 to 238 mg/L, for S_2 from 215 to 248 mg/L, for S_3 from 253 to 305 mg/L, for S_4 from 300 to 365 mg/L and for S_5 from 260 to 320 mg/L.   Ammonium (NH_4^+) and free ammonia (NH_3), were produced during anaerobic digestion, mainly from proteins and amino acids.   Free ammonia was the most toxic even at low levels[2], but methanogenesis could be severely inhibited at concentrations (TAN) exceeding 3 000-4 000 mg/L[20,28].   The concentrations of TAN in all digesters increased during the experimental period, but none of the reactors reached inhibiting values.   Therefore, the TAN concentrations were not likely to have contributed to CH_4 yield inhibition in any of the digesters.

3.6   Optimization strategies towards highest energy production

The primary sludge production rate at STP, Kampala (Uganda) was estimated at 40 m^3/d while the brewery plant had an average daily production of 10 m^3/d.   Table 3 presents the calculated energy potential of different options of using the substrates to which BW was added, compared to the control with 100% STP sludge.   Option C could give the highest energy output with 11 times more than the control.   However, this required a volume 40 m^3 of BW.   The current volume of BW produced at the plant was lower than that, hence the option was not considered practical for application.   This was followed by Option D and B with energy outputs that were seven and four times more than the control, respectively.   However, it is important to note that the tank volume required by option D is 1.5 times more than the Option B, which increases its capital cost.   Operational costs may also slightly be higher in option D, considering that three different waste streams need to be handled.   However, the increased costs may easily be covered in a short time given the fact that the energy production in option D is almost double that of option B.   Moreover, option D is a better scenario at solving problems of abattoir wastes which are increasingly polluting the fresh water sources

nearby.   Therefore, option D is proposed as the optimal co-digestion option in this study.

Table 3   Electricity and heat energy potential of options that brewery sludge was added compared to 100% STP sludge was added

Option	STP:BW: CM ratio	Digester volume /($m^3 \cdot d^{-1}$)	Biogas production rate/($m^3 \cdot d^{-1}$)	Electricity /(kW·h)	Heat energy /(kW·h)
A	100:0:0	800	280	560	1680
B	75:25:0	1060	1272	2544	7620
C	50:50:0	1600	3200	6400	19200
D	50:25:25	1600	2080	4160	12480

Note: The tank volume is calculated based on complete digestion of STP sludge produced at the plant at SRT of 20 d.

The energy is calculated based on a rule of 0.5 m^3 biogas≈1 kW·h electricity + 3 kW·h heat energy in a combined heat and power module.

4   Discussion

National Water and Sewerage Corporation (NWSC) is in charge of the Bugolobi sewage treatment plant and already planed to build an anaerobic digester for the STP sludge.   They would benefit from the increased energy generation.   The annual electricity production estimated from option A is 204 400 kW·h/a, which barely sustains the current plant electricity requirement, estimated at 230 000 kW·h/a. Adapting option D will increase the electricity by a factor 7.   For the new plant, whose sludge volume is estimated to be 10 times than the current one, option D would fully cater for its higher mechanized energy requirements.   In addition, it will provide surplus electricity, which can be sold off to the National Grid, then generate extra income for NWCS with time.

For EABL, the option of co-digesting STP sludge with BW provides a short term optimal solution for save BW disposal.   Otherwise, this would remain a concern, since it is currently quite costly for EABL to treat and get rid of this waste.   The brewery plant will easily be relieved of this cost if their waste is directly fed into the AD process proposed.   Furthermore, on the long term, if EABL decided to adopt AD for BW alone, it will be more costly as the reactor has to be designed to be operated at a higher SRT, of more than 28 d for a stable process. Adopting co-digestion of BW with STP sludge provides good buffering for the process.   This ensures the stability of the reactor at a lower SRT to provide a beneficial option.

Moreover, the proposed optimal substrates with STPS:BW:CM ratios of 50:25:25 represents a scenario which will contribute to decreased pollution to Lake Victoria, since it caters for the safe disposal of CM as well. One of Kampala's biggest abattoirs owned by Uganda meat packers is a few kilometres away from Bugolobi STP. This abattoir lacks waste treatment and disposal facilities. The abattoir waste is damped on an open nearby site and decomposes into manure, which is sometimes collected by farmers. This persistently contributes to greenhouse gas emissions and odour nuisance to the surrounding environment. Furthermore, the runoff through the decomposing waste pile is discharged into the nearby Nakivubo channel and then drains into Lake Victoria ultimately. Therefore, utilizing the CM during co-digestion will make a great contribution towards minimizing pollution to the nearby environment, especially the region's largest fresh water lake.

5　Conclusions

The results in this study have shown that the biodegradability of Bugolobi STP sludge is limited with a biogas yield of (169±10) mL/g COD. Co-digesting STP sludge with BW increased the biogas production rates by a factor of 3, while CM alone did not improve biogas production. Substrate S_4 (50% STP sludge and 50% BW) showed the highest biogas yield and production rate, but S_5 (50% STP sludge, 25% BW and 25% CM) was selected as the optimal mixture for practical application.

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