BIOENERGY RECOVERY FROM THE ANAEROBIC DIGESTION OF MIXED ANIMAL WASTES CO-DIGESTED WITH MIXED FRUIT WASTES

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ABSTRACT

This work evaluated the effects of agitation, feed-inoculum ratio (F/I) and total solid content (TSC) on the anaerobic digestion of mixed animal wastes (cattle dung, pig dung, poultry droppings) co-digested with mixed fruit wastes (of mango, orange, and pineapple) as co-substrate and contents of chicken-gizzard as inoculums for efficient and high recovery of bioenergy (biogas/biomethane). A F/I ratio of 1:1, 1:2, 1:3, 2:1 and 3:1; TSC of 2, 4, 6, 8 and 10% and agitation speed of 0 (i.e. no agitation) and 30 rpm were studied. The anaerobic digestion experiments were carried out in several 20 kg capacity batch anaerobic biodigesters operated at an agitation speed of 30 rpm and incubated at ambient temperature (28 ± 2°C) for 10 weeks. The results showed that the cumulative biogas/biomethane yield obtained from the anaerobic digestion of mixed animal wastes co-digested with mixed fruit wastes and contents of chicken-gizzard as inoculums generally increased with increase in F/I ratio, TSC, and minimum agitation of 30 rpm, respectively. F/I ratio of 1:3 and 3:1 as well as TSC of 8% resulted in a maximum cumulative biogas/biomethane yield of 6.2 dm^3/g/63.3%, 6.1 dm^3/g/62.4% .and 5.8 dm^3/g/60%, with an energy content (EC) of 22.6 MJ/m^3, 22.1 MJ/m^3 and 22.3 MJ/m^3, respectively. A minimum agitation of 30 rpm exhibited a cumulative biogas/biomethane yield of 5.8 dm^3/g/60% with an EC of 21.4 MJ/m^3 higher than 5.2 dm^3/g/55% (EC of 19.6 MJ/m^3) obtained without agitation. The biogas/biomethane production data were fitted to three kinetic growth models of Logistic, Exponential Rise to Maximum and Modified Gompertz, respectively. Modified Gompertz and Exponential Rise to Maximum growth models fitted very well to the data ($R^2 > 0.90$) and thus showed a better correlation of cumulative biomethane production than the Logistic model.

KEYWORDS: *Anaerobic digestion; Animal waste; Biogas; Biomethane; Chicken rumen; Kinetic model*

1. INTRODUCTION

Bioenergy recovery and control of solid waste pollution through anaerobic digestion of biodegradable organic wastes is a promising and more sustainable waste mitigation alternative (Rajagopal *et al*., 2013; Chen *et al*., 2014). Methane rich biogas being one of the renewable fuel or energy substitute is the main product of anaerobic digestion Anaerobic digestion is a multi-step biochemical process where organic materials of different origin or source serially undergoes the following metabolic steps: hydrolysis, acidogenesis, and methanogenesis and each metabolic step is a function of various types of microorganisms (Guo *et al*., 2014; Chen *et al*., 2014). These microorganisms are present in a mixed culture or consortia but differ in their temperature, pH and nutritional requirement, their ability to tolerate environment stresses and growth kinetics (Yi *et al*., 2014). Since anaerobic digestion process is driven by a complex and diverse population of microbial organisms, its performance is affected by various forms of operational variables, such as pre-treatment of feedstock or substrates, pH, total solid content (i.e. concentration of substrate/nutrients), temperature, agitation (i.e. digester mixing), type of substrate, hydraulic retention time, feed/inoculum ratio and the carbon: nitrogen ratio (Sreenivas *et al*., 2010; Umar *et al*, 2013; Chen *et al*., 2014). The mechanism and process of methane rich biogas generation or production is normally slow and takes about 30-50 days (Ogunleye *et al*., 2016). Attempts have therefore been made in the past and in recent times to improve on the economy of biogas plant through enhancing methane rich biogas production, reducing the start-up time and optimizing biogas yield by biostimulation of the microbial activity using various biological and chemical/inorgamic additives under different operating conditions (Ogunleye *et al*., 2016). One of the attempts made to improve biogas yield through improving biomass conversion efficiency was to improve the substrate composition by co-digesting with other biological substrate (additives) (Kennedy *et al*., 2015; Latinwo and Agarry, 2015; Oliveira *et al*., 2015; Aworanti *et al*., 2017).

Thus, co-digestion of animal biomass or waste with other biodegradable organic biomass or waste as co-substrate has been reported in the literature, such as co-digestion of cattle dung with food waste and sludge (Quiroga *et al*., 2014), cattle dung with plantain peels (Latinwo and Agarry, 2015), cattle manure with organic kitchen waste (Aragaw *et al*., 2013), cow dung with fruit waste, food waste and vegetable waste (Otun *et al*., 2015), cow dung and poultry waste with water hyacinth (Imam *et al*., 2013), pig/swine manure with grass silage and grass clippings (Matheri *et al*., 2016) swine and poultry manure with municipal sewage sludge (Borowski *et al*., 2014), chicken droppings/manure with *Cymbopogon citratus*, water hyacinth (Owamah *et al*., 2013), cow dung, chicken manure, pig manure with sewage sludge (Sebola *et al*., 2015), and cattle manure, pig manure, poultry manure with pineapple fruit waste (Aworanti *et al*., 2017). Co-digestion enhances the anaerobic biodegradability of two or more complementary substrates due to synergetic effects (Oliveira *et al*., 2015). The efficiency of any co-digestion process depends on different factors, improving the balance of the mixture of substrate and co-substrates, including the carbon: nitrogen ratio, dry matter, micro and macronutrients, and toxic or inhibitory compounds (Oliveira *et al*., 2015). Agitation is significant in maintaining intimate contact between the microbes and the substrates so as to ensure more active metabolism as well as useful in setting gases that are trapped in the substrates free, and exposing fresh microorganisms to fresh substrates (Santosh *et al*., 2004; Aworanti *et al*., 2017). The major factors that affect agitation/stirring are the agitation strategy, agitation intensity and duration as well as the location of the agitator or stirrer (El-Bakhshwan *et al.,* 2015). Literature reports on the effect of agitation or stirring on biogas/biomethane yield and anaerobic bio-digesters performance have been either positive when agitation is used in minimal or intermittent form (Karim *et al*., 2005; Kaparaju *et al.* 2008; El-Bakhshwan *et al.,* 2015; Aworanti *et al*., 2017) or negative and/or of no significant or considerable effect when agitation is used in the continuous form (Hoffmann *et al.,* 2008). Nevertheless, because of these conflicting reports on the effect of agitation/stirring on biogas yield in the literature, there is the need for further extensive studies on the effect of agitation on bio-digesters performance and biogas yield.

Total solid content (TSC) is an important parameter that provides useful information about biogas yield and has been reported to affect parameters such as: fluid dynamics, clogging, viscosity and rheology of the digester contents, and sedimentation of solid that can directly have effect on the overall rates of mass transfer within the digester (Karthikeyan and Visvanathan, 2013). Because of the importance of TSC, three forms of anaerobic digestion processes based on TSC have been developed: conventional wet or liquid (0.5-10% TSC), semi-dry (10–20% TSC) and modern dry or high-solids (20-40% TSC) anaerobic digestions (Shi *et al*., 2013). Thus, the role of TSC on the performance/efficiency of biomethanization or anaerobic digestion in order to determine its conditions for optimum biogas production has been investigated and reported in the literature (Abbassi-Guendouz *et al*., 2012; Yi *et al*., 2014; Chen *et al*., 2014; Maamri and Amrani, 2014; Deeparanj *et al*., 2015). Abbassi-Guendouz *et al*. (2012) reported that in the batch anaerobic digestion of cardboard under mesophilic conditions, the total methane production decreased as TSC increased from 10 to 25%. The results obtained by Maamri and Amrani (2014) revealed that biogas yield increased with TSC increasing from 12 to 35% in the thermophilic anaerobic digestion of waste activated sludge in the presence of cattle dung as inoculums. Also, Deeparanj *et al*. (2015) in their study of the mesophilic anaerobic digestion of food waste reported that the cumulative volume of biogas produced increased with increasing TSC from 5 to 7.5% and then decreased when TSC was above 7.5%. This result is in contrast with the results obtained by Yi *et al*. (2014) who have reported increased biogas and methane production with increasing TSC from 5 to 20% in their study of the mesophilic anaerobic digestion of food waste. While Forster-Carneiro *et al*. (2008) in their results for dry batch anaerobic digestion of food waste revealed that the biogas and methane yield decreased with the TSC increasing from 20% to 30%.

In a study carried out by Brown *et al*. (2012) to evaluate several lignocellulosic feedstocks (switchgrass, corn stover, wheat straw, yard waste, leaves, and maple) for biogas production under conventional wet or liquid anaerobic digestion (5% TSC) and semi-dry anaerobic digestion (18–19% TSC) observed that there were no significant differences in methane yield between conventional wet and semi-dry anaerobic digestions. In most of these studies, the effect of TSC on biogas or methane yield has been on the use of mono-substrate or feedstock and there has been conflicting variation in the effect of TSC which may be due to the type of substrates involved in the anaerobic digestion. Information on the effect of TSC on biogas/biomethane yield using main substrate with co-substrate and sometimes with inoculum is very limited (Ogunleye *et al*., 2016; Aworanti *et al*., 2017). Our previous study (Ogunleye *et al*., 2016) have revealed that increasing TSC from 8 to 40% resulted in the decrease of biogas/biomethane yield during the biomethanation of animal waste co-digested with fruit waste using chicken rumen as inoculum. This result indicated that TSC of 8% gave rise to the highest biogas/biomethane yield with the use of main substrate co-digested with another substrate (i.e. co-substrate) and inoculum. Thus, there is the need to know how conventional wet or liquid anaerobic digestion (i.e. TSC < 8%) would affect the cumulative biomethane/biogas yield in the anaerobic digestion of major substrates and co-substrates in the presence of an inoculum, of which information on it in the literature is still very limited (Aworanti *et al*., 2017). This is because it is important to know the threshold of the TSC below which biogas or biomethane production from conventional wet or liquid anaerobic digestion is higher or comparable to the output of semi-dry or high-solids anaerobic digestion. It was on this basis that Aworanti *et al*. (2017) investigated the effect of TSC ranging from 2 -10% on biogas/biomethane yield in the biomethanization of mixed animal waste co-digested with pineapple fruit waste in the presence of chicken-gizzard contents as inoculum. The results they obtained showed that biogas/biomethane yield increased with TSC increasing from 2 to 10%. However, in this study, we want to ascertain how this range of TSC would influence cumulative biogas/biomethane yield using the same major substrates but co-digested with mixed fruit waste.

Different research workers have reported in the literature the substantial relevance of inoculums in the process kinetics of biogas/biomethane generation (Sunarso *et al*., 2010). The duration of start-up phase and yield of biogas/biomethane depends on the type of inoculum and feedstock (Aworanti *et al*., 2017). Some workers have used animal manure as inoculum (Maamri and Amrani, 2014) while others have used it as a major feedstock (Latinwo and Agarry, 2015) in anaerobic digestion. Few studies have reported the influence of co-substrate in conjunction with the usage of inoculums on the yield of biogas/biomethane production using animal manure as the main substrate (Aragaw *et al*., 2013; Ogunleye *et al*., 2016; Aworanti *et al*., 2017). The duration of start-up phase and biogas yield depends on the type of feedstock and inoculums. The use of rumen fluid and chicken rumen or chicken-gizzard have been observed to be good for the enhancement of biogas/biomethane production from animal manure (Aragaw *et al*., 2013; Awaranti *et al*., 2017). Higher percentage of inoculums gave the higher production of biogas (Forster-Carneiro *et al*., 2008). However, in most of the anaerobic digestion studied, the inoculums used were dominated by the digested feedstock or substrate in the anaerobic digester. In addition, report or information on the influence of the ratio of feed (i.e. mixed substrate and mixed co-substrate) to inoculums on cumulative biomethane/biogas yield is seldom scarce or limited.

Information on the kinetics of anaerobic digestions of agricultural wastes biomass is very important and a significant factor for understanding anaerobic digestion process, the design, development and performance of an anaerobic biodigester that is efficient for a renewable energy recovery from biomass conversion. The reaction kinetics of biogas generation and the kinetic models developed for the anaerobic digestion process have been reported (Wanasolo *et al*., 2013; Ghatak and Mahanta, 2014; Latinwo and Agarry, 2015; Aworanti *et al*., 2017). However, there is a paucity of information on the kinetics of bioenergy recovery (i.e. biogas/biomethane production) from the anaerobic digestion of mixed animal wastes co-digested with mixed fruit wastes. Therefore, in continuation of our study, the effects of agitation speed, total solid content (2 to 10%) and feed-inoculum ratio on bioenergy recovery (cumulative biogas/biomethane yield) from mixed animal wastes as main substrates and mixed fruit wastes as co-substrates in the presence of chicken-gizzard contents as inoculum were evaluated. Furthermore, to predict the bioenergy recovery (or cumulative biomethane/biogas production) with time, modelling the kinetics of bioenergy recovery was carried out by the application of three known kinetic models (Logistic, Exponential Rise to Maximum and Modified Gompertz) to the mixed substrate-mixed co-substrate-inoculum biomethanization data.

2. MATERIALS AND METHODS

2.1. Sample Collection and Feedstock Preparation

The main substrates consist of cattle dung, pig dung, and poultry droppings and were obtained from a commercial farm in Ogbomoso, Nigeria. The co-substrates which consists of fruit wastes (orange, mango and pineapple) were obtained from a fruit processing industry located in Ibadan, South-West of Nigeria and the inoculums made up of contents of chicken-gizzard was obtained from Ladoke Akintola University of Technology (LAUTECH) poultry farm. The substrates and co-substrates were dried in the sun for twenty (20) days and then dried in the oven at 110 ^{o}C for 10 h, after which they were mechanically crushed with the use of a mortar and pestle to ensure homogeneity. The mixed animal wastes were obtained by mixing the cattle dung, pig dung and poultry droppings in ratio 1: 1: 1 while the mixed fruit wastes were obtained by mixing orange, mango and pineapple fruit wastes in ratio 1: 1: 1. The physical and chemical properties of the biomass wastes (animal and fruit wastes) have been presented elsewhere (Aworanti *et al*., 2017). The micro-organisms identified in each of the waste; orange waste, mango waste, pineapple waste, cattle dung, pig dung, poultry droppings and contents of chicken-gizzard were predominantly bacteria and they are E*scherichia coli, Staphylococcus* species*, Salmonella* species*, Bacillus species, Lactobacillus* species*, Flavobacterium* species, *Methanobacterium* species and *Pseudomonas* species (Aworanti *et al*., 2017)..

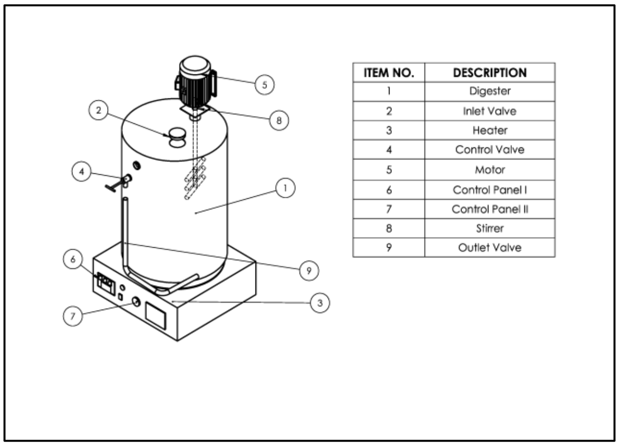

Figure 1: A Pilot Batch Bio-digester

2.2. Description of a Pilot Batch Laboratory Bio-digester

A pilot batch bio-digester (Figure 1) was fabricated according to the design of Fantozzi and Buratti (2009). The description of the bio-digester has been reported (Aworanti *et al*., 2017). It is a cylindrical vessel of 20 kg capacity with 30 cm inner diameter and height of 40 cm. It is made up of stainless steel, fiber glass and galvanized steel equipped with an airtight lid. The lid has inlet and outlet valve on it with two rubber hoses connected to them; one for injection of feed substrates and the other connected to a gas bag for collection of the produced biogas. The bio-digester has an agitator mechanism (Model: RW 16 Basic I KA) about 5 cm below the liquid surface level, operating at a speed range of 10 to 100 rpm to mix the feedstock. Efficient agitation is achieved by propeller with flat stirring paddles and four vertical baffles. Flexible silicon rubber heaters with a maximum operating temperature of 100 ºC was fitted to the bottom of the digester to provide the heat necessary to maintain the feedstock at the required temperature. The digester is equipped with a control panel which permits the regulation of temperature and agitation.

2.3. Biogas/Biomethane Design of Experiment

The hose for feeding and collecting sludge samples was clamped with a bag clamp in order to keep air out and gas in while the other hose connected to the gas bag was made airtight and has a three-way valve that can be opened to collect gas samples. Nitrogen gas was purged through the bio-digester to expel oxygen from the digester and make it air tight in order to ensure anaerobic conditions in the headspace of the anaerobic bio-digester. Twelve (12) bio-digesters each labelled D1 to D5 were charged or seeded with each of the prepared fermentation slurry presented in Table 1. The fermentation slurry was prepared at different operating variables of agitation, total solid content and feed-inoculum ratio according to the method of Ituen *et al.* (2007).

Table 1: Composition of biomass waste in different digester at different operating conditions

		Substrate (kg)							
Biodigester	Parameter	Cattle dung	Pig dung	Poultry droppings	Inoculum	Orange waste	Mango waste	Pineapple waste	Water
D1	No Agitation	0.34	0.34	0.34	2.05	0.34	0.34	0.34	7.92
D2	With AG (30 rpm)	0.34	0.34	0.34	2.05	0.34	0.34	0.34	7.92
	TSC (%)								
D1	2	0.08	0.08	0.08	0.47	0.08	0.08	0.08	11.07
D2	4	0.16	0.16	0.16	0.96	0.16	0.16	0.16	10.09
D3	6	0.27	0.27	0.27	1.64	0.27	0.27	0.27	8.72
D4	8	0.34	0.34	0.34	2.05	0.34	0.34	0.34	7.92
D5	10	o.44	0.44	0.44	2.65	0.44	0.44	0.44	6.71

	F/I Ratio								
D1	1:1	0.37	0.37	0.37	2.21	0.37	0.37	0.37	7.92
D2	1:2	0.25	0.25	0.25	2.94	0.25	0.25	0.25	7.92
D3	2:1	0.49	0.49	0.49	1.47	0.49	0.49	0.49	7.92
D4	1:3	0.18	0.18	0.18	3.31	0.18	0.18	0.18	7.92
D5	3:1	0.55	0.55	0.55	1.10	0.55	0.55	0.55	7.92

D = Biodigester; AG = Agitation; TSC = Total solid content; F/I Ratio = Feed/Inoculum Ratio

The bio-digester was operated at selected total solid contents and feed-inoculum ratios, incubated at ambient temperature (28 ± 2°C) and agitated at a constant speed of 30 rpm. The fermentation or anaerobic digestion was carried out by varying the level of one factor at a time (OFAT) over a certain range while holding other variables constant. As biogas production started in the bio-digester, it was delivered into a gas bag and the produced biogas was measured by means of a weighing scale (Umar *et al*., 2013). The mass of biogas produced was calculated from the difference between the initial mass of the biogas bag and the final mass of biogas bag plus biogas. The volume of biogas produced was estimated using Eq. (1) (Aworanti *et al*., 2017):

$$Volume\ of Biogas\ (dm^3) = \frac{Mass\ of\ Biogas(g)}{Density\ of\ Biogas(g/dm^3)} \quad (1)$$

The yield of biogas (Y) was determined using Eq. (2) (Adeniran *et al*., 2014):

$$Y(dm^3/g) = \frac{Volume\ of\ biogas\ generated\ (dm^3)}{Mass\ of\ feedstock\ in\ biodigester\ (g)} \quad (2)$$

The biogas produced was analysed by means of gas chromatography using the Thermal Conductivity Detector (TCD). The model of the gas chromatograph is Hp6890 with HP ChemStation and Rev. A09.01 (1206) software. The carrier gas is helium at 20 ml/min; it has an inlet temperature of 145 ºC with inlet flow of helium at 26 ml/min. The column dimension is 30 m ×1/8 mm × 0.85 µm and column type Haysep DB 100/120; Deerfield, Illinois. The oven temperature of the gas chromatograph was programmed at 140 ºC in 6 minutes and ramped at 50 ºC/minutes.

2.4 Energy Content (EC) Present in the Generated Biogas

The energy content of biogas in Btu/ft3 can be obtained from the lower heating value (LHV) of biomethane, since it is the combustion of biomethane that produces the heat energy (Prasad, 2012);

$$LHV_{CH_4}(biogas) = LHV_{CH_4} \times F_{CH_4} \quad (3)$$

where LHV_{CH_4} is 33942.86 mJ/m^3 at standard conditions 15 °C and 1 atm, F_{CH_4} is the fraction of biomethane in biogas.

2.5 Modelling the Kinetics of Biogas Generation

The description and evaluation of anaerobic digestion was carried out by fitting the biogas production experimental data to some kinetic models.Cumulative biogas production rates of mixed animal wastes co-digested with mixed fruit wastes (mango, orange and pineapple fruit wastes) was thus simulated using the logistic, exponential rise to maximum and modified Gompertz kinetic models. Logistic kinetic model equation is given in Eq. (4):

$$C = \frac{a}{1 + b\exp(-kt)} \quad (4)$$

where, C, cumulative biogas production (dm^3/g); k, kinetic rate constant (day^{-1}); t = hydraulic retention time (days); a, b are the constants.

Exponential rise to maximum kinetic model equation is given in Eq. (5) (De-Gioannis *et al*., 2009; Lo *et al*., 2010; Latinwo and Agarry, 2015):

$$C = A(1 - \exp(-kt)) \quad (5)$$

Modified Gompertz kinetic model equation is presented in Eq. (6) (Yusuf *et al*., 2011; Latinwo and Agarry, 2015). The model is based on the assumption that cumulative biogas production is a function of hydraulic retention time.

$$P = A\exp\{-\exp[\frac{r_m e}{A}(\lambda - t) + 1]\} \quad (6)$$

Where, P is the cumulative of the specific biogas production (dm^3/gm), A is the biogas production potential (dm^3/g), r_m is the maximum biogas production rate (dm^3/g/day), λ is the lag phase period or the minimum time required to produce biogas (day). The non-linear regression tool of MATLAB 7.0 software package was used to fit the models in Eq. (4) to Eq. (6) to the biogas/biomethane production experimental data.

3. RESULTS AND DISCUSSION

3.1. Effect of Operating Conditions on Cumulative Biogas Yield and Biomethane

3.1.1 Effect of feed-inoculum ratio on biogas/biomethane yield

Figure 2 shows the effect of feed-inoculum ratio on the cumulative biogas/biomethane yield obtained from the anaerobic digestion of mixed animal wastes, co-digested with mixed fruit wastes and contents of chicken-gizzard as inoculums.

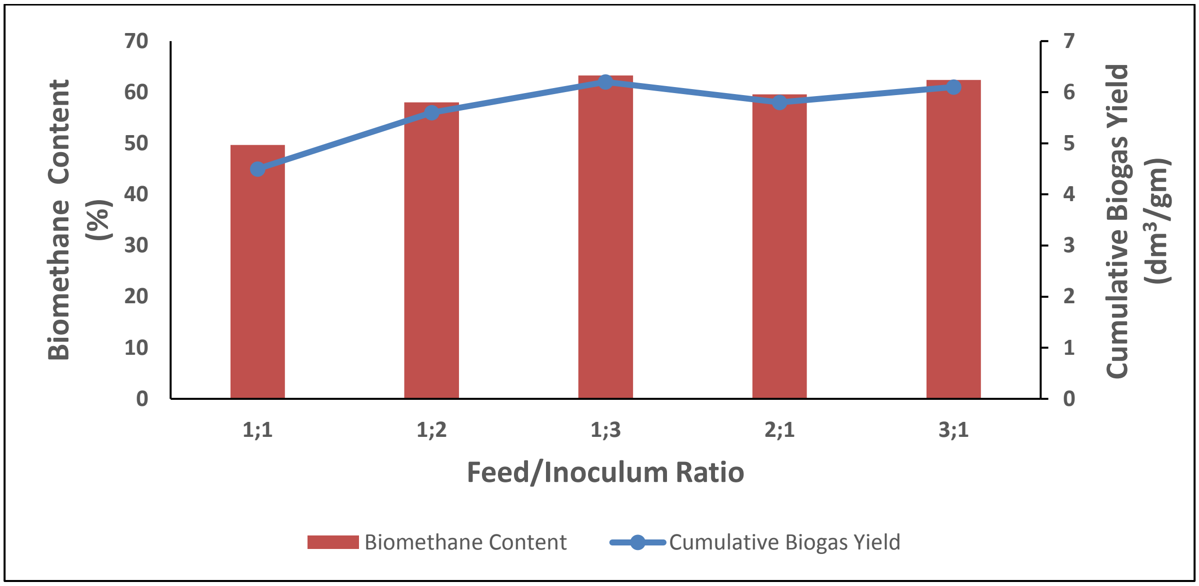

Figure 2: Effect of feed/inoculum ratio on anaerobic digestion of mixed animal wastes co-digested with mixed fruit wastes and contents of chicken-gizzard as inoculum

As shown in figure 2, the cumulative biogas yield and its biomethane content relatively increased with increase in the feed-inoculum ratio (F/I) where the inoculum fraction of the ratio was increased (1:1, 1:2, 1:3) as well as where the feed (substrate) fraction of the ratio was increased (1:1, 2:1, 3:1), respectively. A similar observation has been reported (Sunarso *et al*., 2010). The reason for this observation may be due to higher carbon/nitrogen ratio as well as increase in the community or population of microorganisms present as both the feed (which contain microorganism) and the inoculum were increased which resulted in increased biodegradation of the substrates in the bio-digester. Minimum cumulative biogas yield was achieved with an F/I ratio of 1:1 and the maximum with an F/I ratio of ratio 1:3 where the inoculum fraction of the ratio was increased as well as with an F/I ratio of 3:1 where the feed fraction of the ratio was increased. A higher cumulative biogas yield of 6.2 dm^3/g with a biomethane content of 63.3% having an energy content of 22.6 MJ/m^3 was obtained with an F/I ratio of 1:3. Also, a higher cumulative biogas yield of 6.1 dm^3/g with a biomethane content of 62..4% having an energy content of 22.3 MJ/m^3 was attained with an F/I of 3:1. This observation is in agreement with our previous study where we used the same main substrate (cattle dung, pig dung, poultry droppings), feed-inoculum ratio (1:1, 1:2, 1:3, 2:1 and 3:1) but one type of fruit waste (pineapple fruit waste) (Aworanti *et al*., 2017). The maximum cumulative biogas yield of 6.2 dm^3/g with a biomethane content of 63.3% obtained with mixed fruit waste as co-substrate is relatively lower than the value of 6.4 dm^3/g of cumulative biogas yield with biomethane content of 65.1% that was achieved in our earlier study with pineapple fruit waste as the only co-substrate used. The difference may be as a result of low nitrogen (0.204%), acetic acid (0.481%) and lignin (13.3%) contents of pineapple fruit waste as compared to the high total nitrogen (1.19%), acetic acid (1.61%) and lignin (36.8%) contents of the mixed fruit wastes which tends to lower digestibility (Oliveira *et al*., 2015).

3.1.1. Effect of total solid content on biogas/biomethane yield

Figure 3 shows the effect of total solid content on the cumulative biogas/biomethane yield obtained from the anaerobic digestion of mixed animal wastes, co-digested with mixed fruit wastes (orange, mango and pineapple) and contents of chicken-gizzard as inoculums.

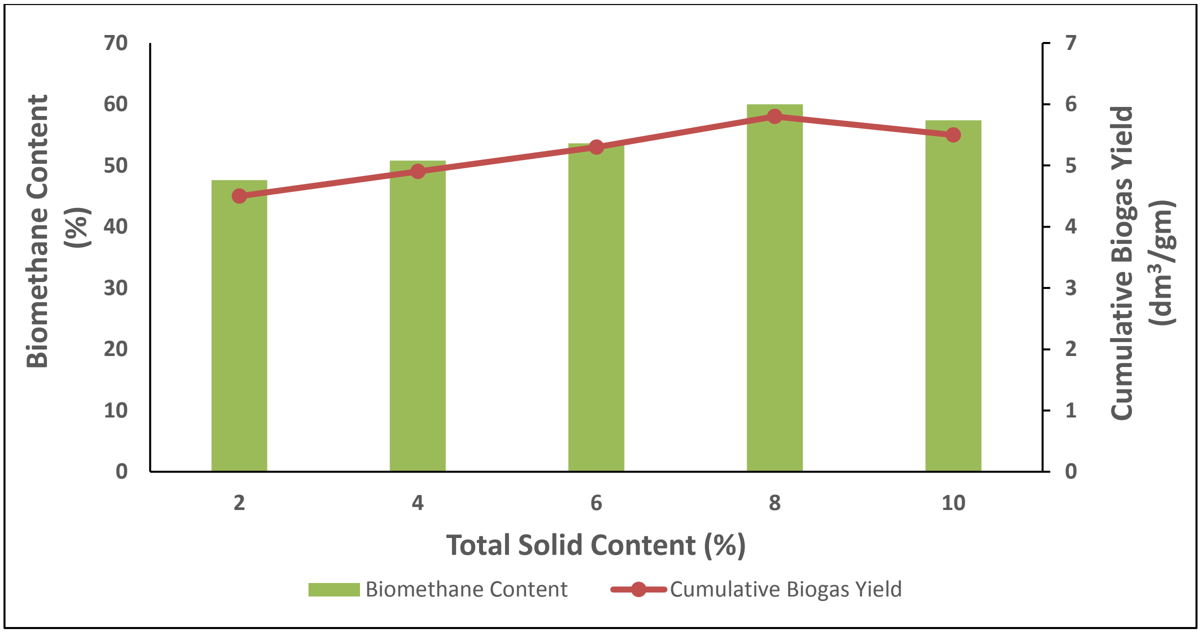

Figure 3: Effect of total solid content on anaerobic digestion of mixed animal waste co-digested with mixed fruit waste and contents of chicken-gizzard as inoculum

From figure 3, it could be seen that the cumulative/biomethane yield generally increased with increase in TSC from 2% to 10%. This observation may be due to the increase in the readily biodegradable substrate available for the microorganisms as the TSC increases thereby resulting in the exponential growth of microorganisms involved in the digestibility (Yi *et al*., 2014; Deepanraj *et al*., 2015). Deepanraj *et al*. (2015) in their study of the anaerobic digestion of food waste have reported an increased biogas yield with increasing TSC from 5 to 7.5% and above this value, there is a decrease in the biogas production. However, the result obtained in this study is in contrast with our earlier investigation, where TSC range of 8 – 40% were used and biogas/biomethane yield decreased with increasing TSC from 8 to 40% (Ogunleye *et al*., 2016). This might be due to poor microbial/enzymatic substrate contact despite the increased readily available biodegradable substrate (Deepanraj *et al*., 2015). .The poor microbial/enzymatic substrate contact may be as a result of decreased water content. Water content is one of the important factors that affects the anaerobic digestion of solid wastes in such a way that it makes possible the movement and growth of microorganism by facilitating the dissolution and transport of nutrients. It also reduced the mass transfer limitation of non-homogeneous substrate (Deepanraj *et al*., 2015). The result obtained in this study is in agreement with the result we obtained for the biomethanization of cattle manure, pig manure and poultry manure mixture co-digested with pineapple fruit waste where a TSC of 2 – 10% were used. A maximum cumulative biogas yield of 5.8 dm^3/g with a biomethane content of 60% having an energy content of 21.4 MJ/m^3 was obtained with TSC of 8%. A similar observation has been reported by Budiyono *et al*. (2010) that biogas yield is significantly affected by total solid content. In their observation, biogas yield decreased as total solid content increased from 9.2 to 18.4% in the production of biogas from cattle manure. These observations implies that maximum cumulative biogas/biomethane yield could be obtained with the use of TSC that ranges between 8 and 10%.

3.1.2. Effect of agitation on biogas/biomethane yield

Figure 4 shows the effect of agitation on the cumulative biogas/biomethane yield obtained from the anaerobic digestion of mixed animal wastes, co-digested with mixed fruit wastes and inoculums (contents of chicken-gizzard).

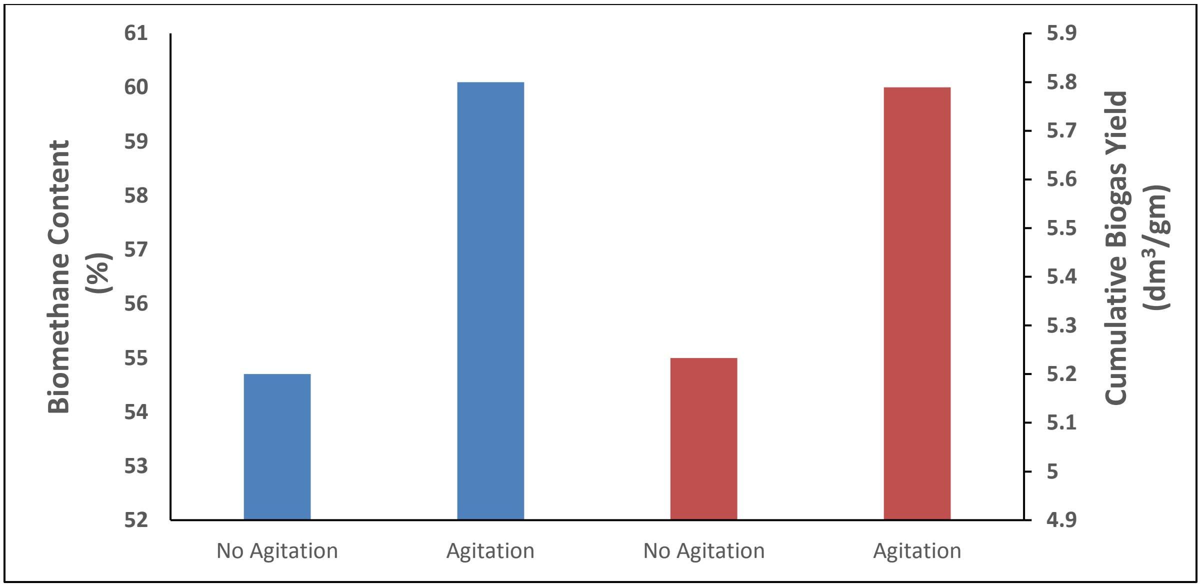

Figure 4: Effect of agitation on biogas/biomethane yield from anaerobic digestion of mixed animal wastes co-digested with mixed fruit wastes and contents of chicken-gizzard as inoculum

As shown in Figure 4, it could be seen that the cumulative biogas/biomethane yield was higher when anaerobic digestion was carried out with agitation as compared to anaerobic digestion without agitation. This higher yield may be due to increased substrate conversion as a result of the agitation or mixing that assisted to uniformly distribute the substrates and the microorganisms in the bio-digester. A cumulative biogas yield of 5.8 dm^3/g with biomethane content of 60% possessing an energy content of 21.4 MJ/m^3 was recovered or obtained with an agitation process of 30 rpm as compared to cumulative biogas yield of 5.2 dm^3/g and biomethane content of 55% having an energy content of 19.6 MJ/m^3 that was achieved without an agitation process or mixing. This corresponded to 11.5% and 9.1% increase in biogas and biomethane production, respectively. This observation is in agreement with the report of Karima *et al*. (2005) and Muthanna *et al.* (2006) whom in their study have respectively observed an increase of 10 to 30% and 15% in biogas production with agitation as compared to an unmixed one. The 11.5% and 9.1% increase in cumulative biogas and biomethane production obtained in this study are lower than what was obtained in our earlier study, where 31.3% and 26.1% increase in biogas production and biomethane was respectively attained with agitation speed of 30 rpm when pineapple fruit wastes were utilized as the only co-substrate in the biomethanization of mixed animal wastes with contents of chicken-gizzard as inoculum (Aworanti *et al*., 2017). This lower increase may probably be due to the presence of high amount of acetic acid, citric acid (contribution from the orange fruit waste), high total nitrogen and lignin contents of the mixed fruit wastes which tends to lower the substrates digestion (Oliveira *et al*., 2015).Therefore, a well agitated or mixed digester is required for optimal biogas production.

3.2. Kinetic Modelling of Animal Wastes Anaerobic Digestion Process

In order to evaluate the performance of the bio-digester, kinetics of anaerobic digestion process has to be evaluated. The biogas/biomethane production experimental data were fitted to the models in Eq. (4) to Eq. (6) (logistic, exponential rise to maximum and modified Gompertz kinetic models) (plots not shown) and the estimated kinetic parameters and coefficient of determination (R^2) obtained from the plots are presented in Tables 2 and 3, respectively. Table 2 shows the values of the model constants and R^2 obtained from the kinetic models fitted to the cumulative biogas production data obtained at different total solid contents.

Table 2: Value of Model Constants and Coefficient of Determination (R^2) Obtained from Kinetic Models Fitted to Cumulative Biogas Production Data of Anaerobic Digestion of Mixed Animal Wastes Co-digested with Mixed Fruit Wastes and Chicken-Gizzard Contents at Different Total Solid Content (TSC)

Models	Constant Variables	Total Solid Content (%)				
		2	4	6	8	10
Logistic	A	1.794	2.016	2.297	3.837	2.858
	B	-1.794	-2.016	-2.297	-3.837	-2.858
	k (day^{-1})	6.2	6.897	7.762	9.158	9.758
	R^2	0.4388	0.4146	0.4122	0.4704	0.4045
Exponential Rise to Maximum	A (dm^3/g)	2.470	2.888	3.299	5.065	4.106
	k (day^{-1})	0.0393	0.0395	0.0438	0.4913	0.0394
	R^2	0.9693	0.9703	0.9679	0.9714	0.9622
Modified Gompertz	A (dm^3/g)	2.283	2.537	3.001	4.576	3.721
	μ_m (dm^3/g/day)	0.0022	0.0029	0.0541	0.0597	0.0363
	λ (day)	0.9312	0.9261	0.9108	0.9031	0.9472
	R^2	0.9925	0.9889	0.9908	0.9853	0.9884

The coefficient of determination (R^2) is comparatively higher for both modified Gompertz kinetic model (0.9853-0.9925) and exponential rise to maximum (0.9622- 0.9714) than that of the logistic kinetic model (0.4122- 0.4704) at different TSC, respectively. The higher value of R^2 above 0.90 for both modified Gompertz and exponential rise to maximum kinetic models indicates that both models fitted very well to the experimental anaerobic digestion data. Thus the two models can respectively be used to simulate bioenergy recovery or biogas production from anaerobic digestion of animal wastes co-digested with mixed fruit wastes and contents of chicken-gizzard used as inoculum at different condition of TSC. From Table 2, it could be seen that the model constants (i.e. the first-order kinetic constant (k) and the cumulative biogas production (A)) of exponential rise to maximum generally increased with increase in TSC, however, with a higher value at TSC of 8%. Similarly, for modified Gompertz equation, biogas production potential (A) and biogas production rate (μ_m) generally increased with increase in TSC, however, with a higher value at TSC of 8 %. TSC of 2% demonstrated the lowest biogas production rate (μ_m) of 0.0022 dm^3/g/day while TSC of 8% exhibited the highest μ_m of 0.0597 dm^3/g/day. Therefore, the quantity of biogas/biomethane produced at the end of bio-digestion period was highest for TSC of 8%. This might be because the mixed animal wastes together with the mixed fruit wastes are rich in nutrients and contains adequate carbon/nitrogen ratio, phosphorous, potassium and some trace elements which are very essential for the anaerobic bacteria growth (Maamri and Amrani, 2014). The lag phase (λ) generally decreased with increase in TSC up to 8% and later increased at TSC of 10%. The lag phases exhibited by TSC of 2 - 10% were relatively short which indicated a good microbial acclimation in the bio-digester. However, the shortest lag phase (0.90 days) was attained by TSC of 8% while the longest lag phase (0.94 days) was exhibited by TSC of 10%. This longest lag phase might be due to low activity of methanogenic bacteria and/or the number of methanogens in the digester that could result in volatile fatty acid (VFA) accumulation produced during the acidogenic step. High concentrations of VFA could lead to methanogenesis inhibition (Maamri and amrani, 2014). Nevertheless, when TSC is higher the quantity and species of anaerobic bacteria required to biodegrade or digest more of the readily available substrate content in the mixed animal wastes and mixed fruit wastes is higher and the bioenergy recovery or biogas/biomethane yield is faster.

Table 3 shows the values of the model constants and coefficient of determination (R^2) obtained from the kinetic models fitted to the cumulative biogas production data obtained at different feed-inoculum ratios.

Table 3: Values of Model Constants and Coefficient of Determination (R^2) Obtained from Kinetic Models Fitted to Cumulative Biogas Production Data of Anaerobic Digestion of Mixed Animal Wastes Co-digested with Mixed Fruit Wastes and Chicken- Gizzard Contents at Different Feed/inoculum (F/I) ratio

Models	Constant Variables	Feed/Inoculum Ratio				
		1:1	1:2	1:3	2:1	3:1
Logistic	A	3.837	4.04	4.285	4.056	4.163
	B	-3.837	-4.04	-4.285	-4.056	-4.163
	k (day^{-1})	9.158	9.264	9.191	9.13	9.203
	R^2	0.4704	0.3852	0.3842	0.4584	0.4058
Exponential Rise to Maximum	A (dm^3/g)	5.065	5.972	6.293	5.446	5.925
	k (day^{-1})	0.0369	0.0375	0.0491	0.0404	0.0468
	R^2	0.9714	0.9596	0.9518	0.9745	0.9607
Modified Gompartz	A (dm^3/g)	4.576	4.701	4.868	4.58	4.688
	μ_m (dm^3/g/day)	0.0022	0.0073	0.0128	0.0134	0.0350
	λ (day)	0.9108	0.8986	0.8206	0.9009	0.8922
	R^2	0.9853	0.9439	0.9606	0.9228	0.9327

Table 3 shows that the R^2 is relatively higher for both modified Gompertz kinetic model (0.9228-0.9853) and exponential rise to maximum (0.9518- 0.9745) as compared to that of the logistic kinetic model (0.3842- 0.4704) at different feed-inoculum ratio. The higher value of R^2 above 0.90 for both modified Gompertz and exponential rise to maximum kinetic models indicates that both models fitted very well to the experimental anaerobic digestion data. Therefore, both models can be used to simulate bioenergy recovery or biogas production from anaerobic digestion of animal wastes co-digested with mixed fruit wastes and contents of chicken-gizzard used as inoculum at different condition of feed-inoculum ratio. As presented in Table 3, it could be seen that the model constants (i.e. the first-order kinetic constant (k) and the cumulative biogas production (A)) of exponential rise to maximum generally increased with increase in the F/I ratio where the inoculum fraction of the ratio was increased (1:1, 1:2, 1:3). F/I ratio of 1:3 demonstrated the highest k value of 0.0491 day^{-1} and correspondingly exhibited the highest cumulative biogas production (A) of 6.293 dm^3/g. This might be due to the increase in the community or population of microorganisms present in the bio-digester resulting in faster substrate digestibility or degradation. F/I ratio of 1:1 resulted in the lowest k value of 0.0369 day^{-1} with the corresponding lowest cumulative biogas production of 5.065 dm^3/g. While k also increased with increase in the feed-inoculum ratio where the feed (substrate) fraction of the ratio was increased (1:1, 2:1, 3:1). Thus, F/I ratio of 3:1 exhibited the highest k value of 0.0468 day^{-1} with a corresponding highest cumulative biogas production (A) of 5.925 dm^3/g. This could be due to increase in the readily available biodegradable substance present in the main and co-substrates in the bio-digester. Similarly, for modified Gompertz equation, biogas production potential (A) and biogas production rate (μ_m) generally increased with increase in the F/I ratio where the inoculum fraction of the ratio was increased (1:1, 1:2, 1:3). F/I ratio of 1:1 demonstrated the lowest biogas production rate (μ_m) of 0.0022 dm^3/g/day while F/I ratio of 1:3 exhibited the highest μ_m of 0.0128 dm^3/g/day. Hence, the amount of biogas/biomethane produced at the end of bio-digestion period was highest for F/I of 1:3. This might be because of increase in the population of microbial species due to increasing amount of the inoculum which the mixed animal wastes in conjunction with the mixed fruit wastes have contact with it. Also, the biogas production potential (A) and biogas production rate (μ_m) generally increased with increase in the F/I ratio where the feed fraction of the ratio was increased (1:1, 2:1, 3:1). Therefore, F/I ratio of 3:1 resulted in the highest μ_m value of 0.0350 dm^3/g/day with a corresponding highest cumulative biogas production (A) of 5.925 dm^3/g. This could be due to increase in the readily available biodegradable substance present in the main and co-substrates in the bio-digester. The lag phase (λ) generally decreased with increase in the F/I ratio where the inoculum fraction of the ratio was increased (1:1, 1:2, 1:3). The lag phases exhibited by F/I ratios of 1:1, 1:2 and 1:3 were comparatively short which showed a good microbial adaptation in the bio-digester. Nevertheless, the shortest lag phase (0.8206 days) was achieved by F/I ratio of 1:3 while the longest lag phase (0.9108 days) was exhibited by F/I ratio of 1:1. This longest lag phase might be due to the activity of the methanogens being low and/or the number of methanogenic bacteria in the bio-digester that could result in volatile fatty acid (VFA) accumulation which is produced during the acidogenesis. Furthermore, the lag phase (λ) generally decreased with increase in the F/I ratio where the feed fraction of the ratio was increased (1:1, 2:1, 3:1). The shortest lag phase (0.8922 days) was achieved by F/I ratio of 3:1 while the longest lag phase (0.9108 days) was exhibited by F/I ratio of 1:1. Similar observation has been reported in our earlier study using pineapple fruit waste as the only co-substrate with contents of chicken-gizzard as inoculum. Table 4 shows the comparison of the values obtained for both modified Gompertz and exponential rise to maximum models' constants with that obtained for the biomethanization of mixed animal wastes co-digested with pineapple fruit wastes and contents of chicken-gizzard as inoculum (Aworanti *et al*., 2017).

Table 4. Comparison of the values of kinetic models constants obtained for anaerobic digestion of mixed animal wastes co-digested with mixed fruit wastes using an inoculum with the values obtained for the biomethanization of mixed animal waste co-digested with pineapple fruit waste using an inoculums at feed-inoculum ratios of 1:1 to 1:3

Kinetic Model	Model Constants	Mixed Animal Waste + Mixed Fruit +Inoculum	Mixed Animal Waste + Pineapple Fruit Waste + Inoculum (Aworanti *et al*., 2017)
Exponential Rise To Maximum	A (dm^3/g	5.07 – 5.93	5.16 – 7.39
	k (day^{-1})	0.0369 – 0.0491	0.0422 – 0.0473
Modified Gompertz	A (dm^3/g	4.58 - 4.87	4.79 – 6.23
	μ_m (dm^3/g/day)	0.0022 – 0.0350	0.0268 – 27.5
	λ (day)	0.911 – 0.821	1.49 – 0.492

From Table 4, it is seen that for the use of pineapple fruit wastes as co-substrate; the values of A, μ_m and k are higher and the λ values are lower than the corresponding values obtained for mixed fruit wastes (pineapple, mango and orange) used as co-substrate in this study. This observation indicates that the use of pineapple fruit wastes as co-substrate in the anaerobic digestion of mixed animal wastes has a higher potential of biogas generation with higher rate of biogas production and lower lag phase (i.e. faster start-up time) than its mixed form with other fruit wastes such as orange and mango. This is due to the presence of low amount of acetic acid and lignin contents as compared to the total high amount of acetic acid, lignin content and citric acid (contribution from the orange waste) present in the mixed fruit wastes. High amount or concentration of these substances tends to decrease anaerobic substrates digestion (Oliveira *et al*., 2015).

4. CONCLUSION

The following conclusions can be deduced from this study of the anaerobic digestion of the mixed animal wastes (cattle dung, pig dung and poultry droppings) co-digested with mixed fruit wastes (orange, mango and pineapple waste) using the contents of chicken-gizzard as inoculum:

1. the bioenergy recovery or cumulative biogas yield with its biomethane content generally increases with increasing feed-inoculum ratio (where the inoculum fraction is increased as follows 1:1, 1:2 and 1:3 and the feed fraction is also increased as 1:1, 2:1 and 3:1, respectively) as well as with increase in total solid content that ranges from 2 to 10%;
2. higher cumulative biogas yield with high biomethane content having higher energy content can be obtained at the feed-inoculum ratio of 1:3 and 3:1, respectively, while higher cumulative biogas yield with higher biomethane content possessing higher energy content can be obtained at an optimum total solid content of 8 to 9%;
3. higher cumulative biogas yield with high biomethane content and energy content can also be attained with minimum agitation of the biodigester than without agitation;

4. the rate of biogas/biomethane production can adequately be simulated and predicted with both modified Gompertz and Exponential Rise to Maximum kinetic equations as both respectively had higher correlation with the biogas/biomethane production data than the Logistic kinetic model equation.

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