

Investigating the effect of temperature gradient on biogas production from pretreated maize straw and rice husk using multistage anaerobic bioreactor

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The share of biogas in renewable energy sources is increasing as variety of feedstocks are now used for biogas production among which lignocellulosic biomass is emerging feedstock that can be used after proper pretreatment under best suited temperature. Although lot of pretreatments and temperature combinations have been tested but still there is a gap that can be filled by the current study focused on the effect of the temperature gradient (mesophilic and thermophilic) on biogas production potential of maize straw and rice husk using a modified Gompertz equation. Pretreatment was done by using alkali (NaOH and Ca(OH)₂) and acids (HCl and H₂SO₄) each at 2, 4 and 6%. The pretreatment of crop residue with 6% NaOH degraded lignin contents significantly. The pretreated crop residue was further used for biogas production. A multistage anaerobic bioreactor containing three diagonally inline reactors provided with one water bath connected to reactors for better utilization of energy was used for biogas production. The temperature of water bath was adjusted to that the first reactor achieved 37°C and 55 °C for different experiments. The working temperatures found to be 31-37°C and 46-55°C were achieved to maintain the internal temperature of the reactors within mesophilic and thermophilic temperature ranges, respectively. The 36 days incubation time was equally divided for three reactors. The biogas production rate was 297 mL/g-VS_{added} and 244.07 mL/g-VS_{added} from maize straw and rice husk under mesophilic conditions, respectively. The results showed an increased biogas yield for both feedstocks under mesophilic conditions as compared to thermophilic conditions. The central reactor showed better production as compared to other two reactors in all experiments.

Keywords: Biogas, Acid-base pretreatment; crop residue; multistage bioreactor; mesophilic; thermophilic.

INTRODUCTION

Biogas production from organic waste is the most adopted method for centuries. There are number of success stories in the production of biogas from various kinds of feedstock till date and it is adding on daily basis (Weiland, 2010). The first phase of the research on biogas focused on production of biogas whereas in the later step the efficiency of biogas production process was investigated in term of type feedstocks, reactor design and process parameters. In order to improve the production efficiency, variety of feedstocks, many reactor designs were tested but still the opportunity for achieving higher yields is always there. One of the major issues in the biogas production process is to maintain a specific temperature for smooth biogas production which increase the energy input in the process (Xiong, 2012; Rademacher *et al.*, 2012; Sattar *et al.*, 2016). Due to the low energy requirement of mesophilic process, most of the practical designs of reactor work under mesophilic conditions

(Salehiyoun *et al.*, 2019). The other allied issue is to harvest the energy input efficiently. For this purpose, two-stage biogas production was employed in which the two reactors were operated with a single heat source (Van *et al.*, 2020). In such studies, one reactor is under thermophilic conditions whereas other is under mesophilic conditions. Temperature is one of the main factors regulating the digestion process performance and microbial activity (Labatut *et al.*, 2014). Most of the multistage reactors configurations were used to separate the hydrolysis, acetogenesis, methanogenesis phase irrespective of energy input by using multiple heating source that increase the cost of production (Achinas *et al.*, 2017). The other research domain focused on the choice of feedstock by which variety of feedstock like an organic fraction of municipal solid waste (Busch *et al.*, 2009), waste produced from the food industry as well as agricultural waste like crop residues, animal dung, husks etc. were tested (Zhang *et al.*, 2014; Sattar *et al.*, 2016; Lee *et al.*, 2019). In this domain, the utilization of lignin-cellulose waste is gaining attention.

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Maize was observed to be one of the potential and economical feedstocks and showed 235 mL/g-TS biogas yield (Zhong *et al.*, 2011). Maize has the highest latent for methane yield due to high guts of carbohydrates and a high digestibility (Vetter, 2009). Przybyl *et al.* (2017) reported that lignin-cellulose material is the largest source of raw material for the production of biogas in anaerobic digestion. Maize straw digestate can also be used as an organic fertilizer because it is enriched with various nutrient. Maize provides 8,542,000 MT straw residues annually which can be utilized for energy production (Saeed *et al.*, 2015). In the same way, rice husk is also an important feedstock apart from its other uses and also abundant in South-East Asia (Chandra *et al.*, 2012; Sattar *et al.*, 2016). Rice also has high lignin contents which are easily biodegradable. In Pakistan, the total annual production of rice is 6,160,000 MT out of which 1,232,000 MT is rice husk (Saeed *et al.*, 2015). Lignin binding is the only issue in utilizing such feedstocks because cellulose and hemicellulose are not exposed to micro-organisms easily. The issue was addressed by employing physical, chemical and biological pretreatments that exposed the cellulose contents to the microbes (Ngan *et al.*, 2020).

Various studies highlighted the effect of feedstock pretreatment on increasing the digestion and biogas production rate. Pretreatment reduced the cellulose crystallinity and improves the biodegradability of lignocellulosic feedstock (Liu and Cheng, 2009). Saccharification with dilute alkali and acid, ammonia solution, ethanol fermentation, pretreatment with hot water and recycled liquid from anaerobic digestion are some common pretreatment strategies (Adl *et al.*, 2012). Pretreatment with dilute acid is an efficient and cost-effective method (Keikhosro *et al.*, 2006). However, alkaline pretreatment is effective in lignocellulose delignification (Pavlostathis *et al.*, 1985). Under anaerobic digestion, maize straw pre-treatment with NaOH and CaO can reduce the digestion time of biogas production up to 80% (You *et al.*, 2019) and produced 87.5% more biogas and 111.6% higher methane than untreated feedstocks (Chandra *et al.*, 2012). Sulfuric acid pretreated feedstock also improved biogas yield with short lag time (Syaichurrozi *et al.*, 2019). Keeping in view the facts, this study was designed to observe the biogas production potential of pretreated alkaline and acidic rice and maize straw from three-stage reactors (multisector) operated within mesophilic range in comparison with a thermophilic range from the single heating source to harvest maximum input energy.

MATERIALS AND METHODS

Feedstock preparation: The study was conducted at Environmental Engineering Laboratory, University of Agriculture, Faisalabad, Pakistan. The maize straw and rice husk samples were collected from field research area of

University of Agriculture, Faisalabad, Pakistan. Samples were air-dried, chopped and grinded finely to form a homogenous mixture (Fig. 1). Grinded samples were sieved with 2 mm mesh size after which they were pretreated with 2, 4 and 6% solutions of NaOH, Ca(OH)₂, H₂SO₄ and HCl. The samples were soaked in above mentioned reagents, covered with aluminium foil and kept for 24 has described (Liu and Wyman, 2003). Samples were washed properly to remove residues and were oven-dried at 105°C and then placed in a desiccator for moisture removal as described by (Chen *et al.*, 2007). The effect of pretreatment was determined on the basis of cellulose, hemicellulose and lignin contents measured by the method adopted by Ververis *et al.* (2007). The slurry collected from the wastewater treatment plant was used as an inoculum in biogas production. Volatile solids, pH and total solids of slurry were 15.3%, 6.4 and 18.3%, respectively.



Figure 1. Grinded Feedstock for Pretreatment (Left) and washing of treated straw (Right)

Experimental setup: A multistage bioreactor system was developed by constructing three similar double jacketed anaerobic bioreactors which were aligned vertically so that residual material from reactor A would be easily passed to reactor B and then from reactor B to reactor C.

An anaerobic reactor was designed which was prepared from stainless steel and comprised of two tanks i.e., inner tank and outer tank. In the inner tank, having the capacity of 11.398 L with 30×11 cm dimension, decomposition of crop residues took place. Whereas the outer tank maintained the temperature of the inner tank by circulating the hot water around it. Each reactor had an inlet valve, a sample collection port and an outlet valve. The reactor was equipped with a temperature sensor (HANNA Instruments ML1010) and biogas analyzer (Gasboard 3200 L).

A gas collection port was also installed at the upper cover of the reactor which was associated with exit regulator. For thoroughly mixing of inner tank material, an agitator was fixed which was operated with a DC motor (Fig. 2). The water bath was installed for attaining and maintaining mesophilic (37°C) and thermophilic (55°C) temperature of the first reactor only.

Analytical and assay method: The total solids (TS), volatile solids (VS), volatile fatty acids (VFAs) and alkalinity were measured by standard methods (APHA, 2005). Lignin, cellulose and hemicellulose contents of maize straw and rice husk was analyzed by procedure opted by Ververis *et al.* (2007). The reactor A was connected to water bath and the temperature of water bath was adjusted to achieve 37°C temperature in reactor for mesophilic temperature experiment. The water then moved to reactor B and C and the temperature of all three reactors was found in 31-37 °C range. Similarly, for thermophilic set experiments, the temperature was 46-55°C.



Figure 2. Multistage Bioreactor used for biogas production.

Reactor A was fed with 2 kg of slurry and 2 kg of pretreated feedstock. The volume of the reactor was maintained up to 8 L with distilled water for maintaining 10% TS content. Digestion was performed for 36 days (Zhang *et al.*, 2015), and the material was kept in reactor A for 12 days and then transferred to reactor B and then after 12 days, residual material from reactor B was transferred into reactor C where the material was further digested for another 12 days to get maximum biogas production from crop residues. Production of biogas and methane (daily and cumulative) at mesophilic and thermophilic temperature ranges and pH in each reactor was analyzed regularly at 24 h intervals along with VFAs. Once the biogas production was ceased, TS and VS of residual material were determined. For kinetic parameters determination, production of methane gas was modelled by Gompertz Equation (Ramos *et al.*, 2012).

$$H = P \exp \left\{ - \exp \left[\frac{Rm}{P} (\lambda - t) + 1 \right] \right\}$$

Where, H represent cumulative methane production (ml), P is methane production potential, t is the time of incubation, h is

duration of lag phase, Rm is maximum methane production rate (ml/h) and e is 2.71828. Values of H, P, t and Rm were identified by using curve fitting tool in MATLAB.

Batch experiment: Equal proportion of slurry and pretreated crop residue (maize straw/ rice husk) was added in the reactor and further water was added in the mixture for keeping the TS level 10%. Initial pH of the 6% NaOH pretreated mixture was maintained at 7 (Guo *et al.*, 2010; Arslan *et al.*, 2010). The pH was maintained by adding 5M NaOH and/or 5M HCl as per requirement. After addition of materials, the lid of the first reactor was closed tightly to develop anaerobic conditions and materials were digested in the first reactor for 12 days and then transferred to reactor B and then to reactor C after 12 days. Circulating hot water in the outer jacket of the reactor maintained the temperature of digestion chambers. The volume of produced biogas, methane, pH and VFAs of the digestion material was measured after the interval of 24 h. On completion of digestion (after 36 days), TS and VS were analyzed (Kim *et al.*, 2013) and the residual material was discarded.

RESULTS AND DISCUSSION

Effect of pretreatment on the degradation of feeding material:

In the direction of upgrading the degradation of maize straw and rice husk and improving biogas yield in a multistage anaerobic setup, maize straw and rice husk were pretreated with alkali and acids at various percentages. The pretreatment of maize straw and rice husk was accomplished with 2, 4 and 6% solution of NaOH, Ca(OH)₂, H₂SO₄ and HCl. The consequences of concentration and treatments on the maize straw and rice husk degradation were presented in Fig. 3. The comparison of processed and unprocessed maize straw revealed that concentration and treatments owned the dominant effects on lignocellulose contents of crop residue. The results also suggested that the degradation rate of lignocellulose contents comparatively higher with alkali while lower with acid pretreatment. In the consideration of acid pretreatment, the relatively higher percentages of lignin and cellulose contents were perceived from the pretreatment experiment. In response to this higher percentage the aggregate of hemicellulose considerably decreased (Wang *et al.*, 2015). It can be seen that hemicellulose and lignin were highly degraded by alkaline treatments in comparison with cellulose. That was due to the cellulose crystallinity which opposed the cellulose degradation

The delignification process is considered mandatory to enhance the lignocellulose degradation because lignin is reported as a physical barrier and restricts enzyme activity (Yoon *et al.*, 2014). The NaOH pretreatment exhibited a positive substantial influence on lignocellulose degradation in comparison with Ca(OH)₂ (Salehian *et al.*, 2013). The consequences of NaOH concentrations on the maize straw and rice husk degradation were presented in Fig. 3. When the

concentration of NaOH escalated from 2% to 6% lignin percentages decreased from 6.9 % to 5.4 % and 7.0 % to 6.4 % for maize straw and rice husk, respectively. The outcome of pretreatment indicated that with the increment of NaOH concentration the hydrolysis of cellulose gradually increased because the hemicellulose and the lignin elimination authorized the cellulose hydrolysis development in a rapid way (Teater *et al.*, 2011). It can be observed from the consequences that 6% concentration of NaOH significantly decreased the lignin content, and lignin was considered unfavorable ingredient during the fermentation process. Therefore, pretreatment with 6% NaOH was recommended for this research by considering the degradation of lignin, comprehensive gas production rate and total gas production.

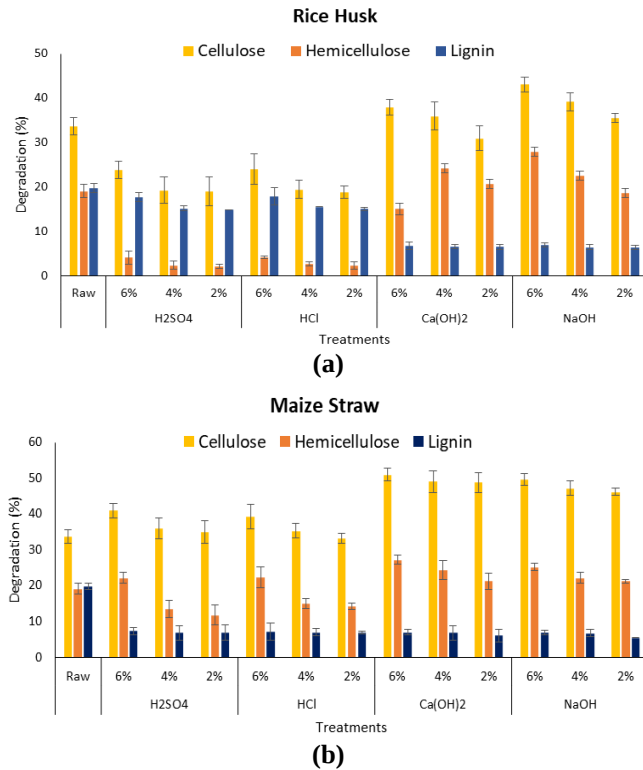


Figure 3. The lignocellulose degradation of (a) rice husk and (b) maize straw under different pretreatments.

Cumulative Gas production:

Mesophilic temperature: Anaerobic digestion of pretreated maize straw and rice husk was initiated at $37 \pm 0.5^\circ\text{C}$ (mesophilic temperature) perpetuated for reactor A. The residual material was moved to reactor B after completing 12 days in reactor A where the temperature was reduced to $33 \pm 1.3^\circ\text{C}$ and then to reactor C after completing the period of 12 days in reactor B where the temperature was further dropped to $31 \pm 1.3^\circ\text{C}$ due to continuous transfer of heat. Cumulative biogas and methane production from maize straw and rice husk in different reactors under different mesophilic

conditions is presented in Fig. 4 and Fig. 5. The cumulative biogas and methane production of maize straw and rice husk almost exhibited equivalent trends in the corresponding reactors under the mesophilic temperature conditions.

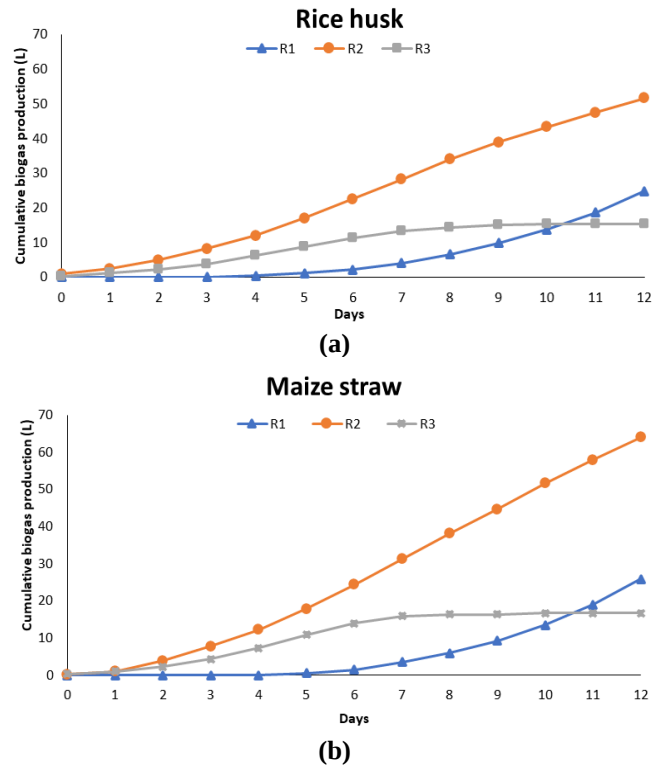


Figure 4. Cumulative biogas production from (a) rice husk and (b) maize straw under mesophilic conditions in A (R1), B (R2) and C (R3) reactors.

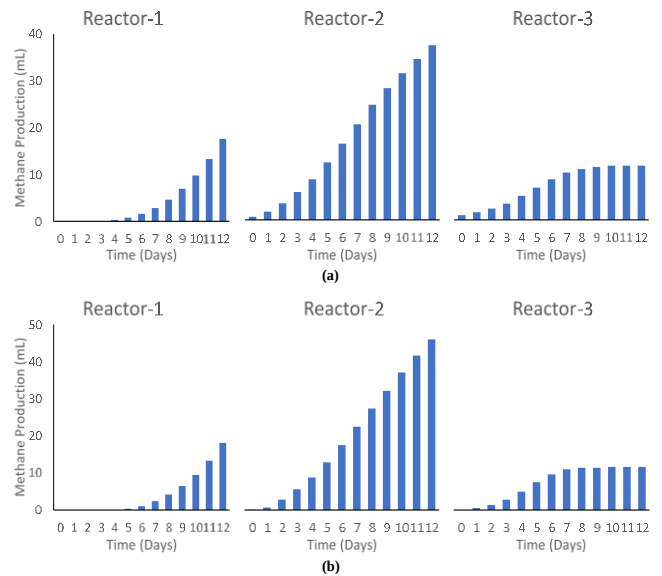


Figure 5. Accumulated methane production from (a) rice husk and (b) maize straw under mesophilic conditions.

It was observed from the consequences of the experiment that on-set of production of methane in reactor A for both the crop residues (maize straw and rice husk) was started on 4th day of incubation. Maize straw owned a higher percentage of lignocellulose contents in comparison with sugars and starch percentage (Zareei and Khodaei, 2017). Therefore, the lower concentration of lactic acid and the higher concentration of volatile fatty acid caused the lower methane production at the early days of incubation in reactor A (Wang *et al.*, 2020). Methanogens were considerably sensitive in the perception of temperature variations because maturation and metabolic action of methanogens were remarkably influenced by temperature (Yang *et al.*, 2018). During the transportation of remnant material of maize straw and rice husk from reactor A to B, the temperature of the substrate decreased from $37\pm0.5^{\circ}\text{C}$ to $33\pm1.3^{\circ}\text{C}$ accordingly. Therefore, initially the methane production in reactor B significantly decreased as compared with the final stage of methane production in reactor A. When the merits of temperature and other conditions are optimized then the more stable the microbial metabolic system is established to produce sustainable methane gas (Gu *et al.*, 2020). The one cause behind this substantial addition in the accumulated methane production in reactor B was NaOH pretreatment. The lignocellulose contents of pretreated maize straw and rice husk was being degraded easily during the digestion. Therefore, the acid producing bacteria and methanogenic bacteria in the incubation process can effectively utilize the substrate to grow and reproduce (Feng *et al.*, 2018).

In reactor C the accumulated methane production initially increased to a certain level and then maintained a constant level because the daily methane production declined and eventually methane production was ceased in the digestion process. This downturn in methane production occurred because of complete consumption of organic components of crop residues (Chen *et al.*, 2020). The growth and reproduction of bacteria decelerated rapidly and as a result of this methane production declined gradually. As the contents of maize straw and rice husk were continuously decomposed and utilized, the quantity of residual functional substrate in reactor C was becoming less and less. Therefore, methane production was gradually reduced until the gas production has stopped.

Thermophilic temperature: Anaerobic digestion of pretreated maize straw and rice husk was conducted for 36 days and ($55\pm0.5^{\circ}\text{C}$) initial mesophilic temperature was perpetuated for reactor A. Same conditions and procedures were adopted in thermophilic digestion which was adopted in mesophilic digestion. The temperature of remnant material of maize straw and rice husk measured in Reactor B and C was ($51\pm1.5^{\circ}\text{C}$) and ($46\pm2.01^{\circ}\text{C}$), respectively. Cumulative biogas and methane production of maize straw and rice husk in different reactors under different thermophilic conditions is shown in Fig. 6 and 7. The maximal yields of biogas in these

three digesters for rice husk under thermophilic and mesophilic temperature were observed 244.07 mL/g and 199.92 mL/g, respectively.

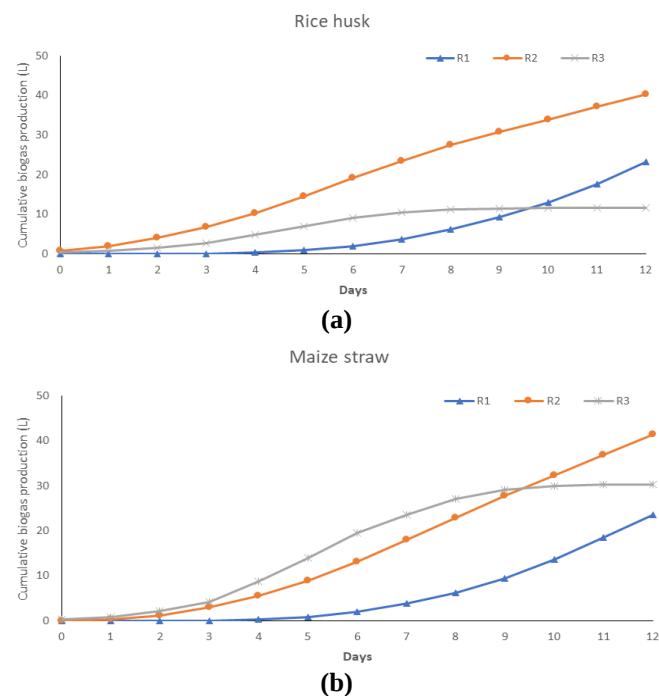


Figure 6. Cumulative biogas production from (a) rice husk and (b) maize straw under thermophilic conditions in A (R1), B (R2) and C (R3) reactors.

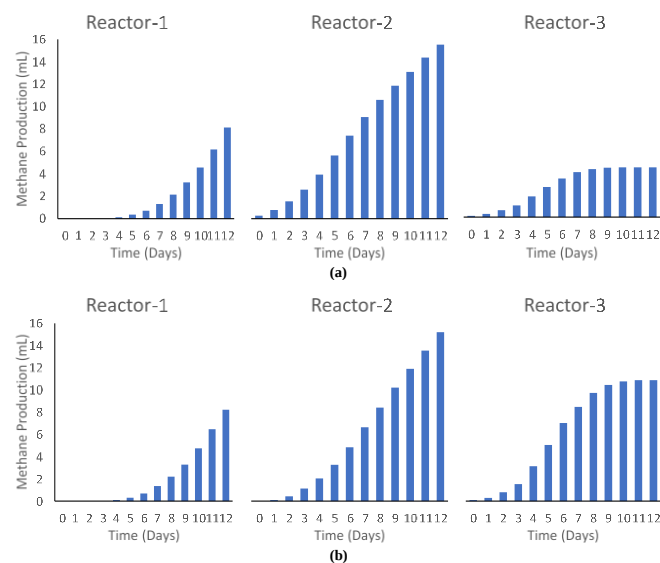


Figure 7. Accumulated methane production from (a) rice husk and (b) maize straw under thermophilic conditions.

While maximal yields of biogas in these three reactors for maize straw under thermophilic and mesophilic temperature

were observed 286.42 mL/g and 255.86 mL/g, respectively. Methane yield was maximized in reactor 2 in both maize straw and rice husk digestion as shown in Figure 7. Under thermophilic digestion temperature, 27.35L and 33.01L cumulative methane was produced by rice husk and maize straw, respectively. Cumulative methane production from rice husk was 8.13, 14.91 and 4.30L while from maize straw was 8.22, 14.4 and 10.91 L produced in reactor 1, 2 and 3, respectively.

The outcomes of the maize straw and rice husk digestion indicated that biogas production dynamics significantly assorted with mesophilic and thermophilic conditions. The comparative analysis of reactor A under mesophilic temperature indicated that cumulative methane production was relatively high while under thermophilic temperature cumulative methane production was low (Sun *et al.*, 2019). Based on examination of sequences obtained from mesophilic and thermophilic in terms of biogas production yield, normally it can be recapitulated that mesophilic temperature was more appropriate for the sustainable digestion of pretreated maize straw and rice husk. The acetic acid was considered an imperative source of methane production during anaerobic digestion. While the thermophilic fermentation improved the concentration of butyric acid which resulted from the degradation of sugar and acetic acid (Capson-Tojo *et al.*, 2017).

Kinetic parameters: In this research, the modified Gompertz model was applied to evaluate the biogas production potential (R_m) L/d, cumulative production (P) L and lag phase (λ) in days. Kinetic parameters and goodness fit derived from the estimated model were shown in Table 1, and a, b, I represented the cumulative biogas production, biogas production potential and lag phase, respectively. This method was applied to represent the effects of thermal conditions and multi-stage biogas reactor. Maximum values of cumulative biogas production (100.7 mL/g VS for rice husk and 118 mL/g VS for maize straw) were acquired in the Modified Gompertz equation under mesophilic conditions. The biogas production potential for maize straw was relatively higher than rice husk and noted values were 5.5 and 4.54 L, respectively. Modified Gompertz model predicted the rice husk owned the comparatively shorter lag phase of approximately 8 days while the value of lag phase (f) was higher for maize straw i.e., 10 days. Generally, the estimated values of the goodness of fit indicated that there was an inclusive consensus among the evaluated model and the experimental data. The comparison of the performance of the evaluated model revealed that the best fit was derived for both

rice husk and maize straw and obtained the dominant regression values for R-square 0.9963 and 0.9942, respectively.

Fig. 8 represented the evaluated model fit and the experimental data for the anaerobic digestion of both rice husk and maize straw under mesophilic and thermophilic temperature. The comparison of consequences of accumulated biogas yield for the experimental and evaluated model indicated that the value of accumulated biogas yield at thermophilic temperature was higher than the evaluated model. While on other hand, the value of accumulated biogas yield at mesophilic temperature was lower than the evaluated model. The same trend was observed in the consideration of rice husk and maize straw. Initially, the production of biogas owned the slower rate and this slower rate extended to 8 to 10 days for rice husk and maize straw, respectively. It happened due to the existence of the lag phase during the microbial growth. The observation of outcomes also showed that the growth of biogas production increased during the range of 10 to 25 days due to the remarkable growth of microorganisms. As the time duration passed the 25 days that rate of biogas production decreased due to persistent growth of microorganisms. It also can be concluded that the rate of biogas production was slightly higher for maize straw in comparison with rice husk.

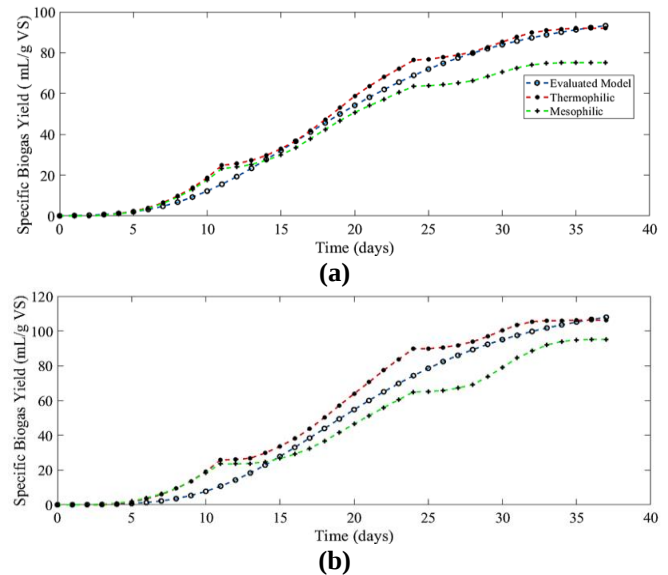


Figure 8. Experimental and evaluated model production of accumulated biogas from (a) rice husk and (b) maize straw under mesophilic and thermophilic conditions.

Table 1. Kinetic parameters and goodness fit derived from the estimated model.

Feeding material	a (mL/g VS added)	b (mL/g VS)	I (Days)	R-square	Adj. R-square
Rice husk	100.7	4.543	7.943	0.9963	0.9961
Maize straw	118.0	5.500	10.02	0.9942	0.9939

pH variation during fermentation: The pH value is considered an important indicator in the reaction process of biogas fermentation and it is mandatory that pH must be between 4.0 and 8.5 for biogas microbial fermentation but the optimum value is 7.0 (Dai *et al.*, 2016). The variations in pH value during anaerobic fermentation of maize straw and rice husk in different reactors under mesophilic and thermophilic conditions are shown in Fig. 9. The pH value of 7.5 at the initial phase of fermentation in every reactor was inaugurated for both mesophilic and thermophilic conditions. The consequences indicated that at the initial phase of fermentation in reactor A the pH value substantially decreased until a certain period of time for both maize straw and rice husk. The identical decreasing trend in pH value was observed for both mesophilic and thermophilic temperature. It happened because at the beginning of the digestion process, the contents of the rice husk were tightly bound and the concentration of available substrate was relatively low (Ravi *et al.*, 2018). Therefore, the activity of methanogens was low and acid producing bacteria was active. In response to this the concentration of organic acid was increasing while the value of pH was decreasing. It was also observed that after 4 to 5 days of digestion pH value started to gradually increase and perceived its value between 7.0 to 7.5 for the rest of the digestion process considered in reactor A. As the digestion process proceeded, the organic acid in the fermentation process was continuously consumed by methanogens and the concentration of methane, carbon dioxide and other gases started to increase and pH value also increased in response to this acid degradation (Wang *et al.*, 2018).

The variations in the pH value of reactor B were almost sustained and adopted the range of 7.2 to 7.6. While the variations in pH value of reactor C were fluctuated and adopted the range of 7.2 to 8.0. (Fig. 9). These fluctuations are closely related to the activity of microorganisms in the fermentation process. During the initial phase of fermentation in reactor B, the pH value was regulated, and 7.5 pH was achieved. At the inauguration of digestion in reactor B, the first pH value gradually decreased to small-scale and then started to increase. Due to the pretreatment of feeding material and progress time the rice husk components were decomposed into small molecules. Therefore, the available substrate concentration of methanogens at this stage of digestion was considered high. The growth and reproduction of methanogenic bacteria increased, and rapid rehabilitation of pH value was observed in reactor B. The pretreatment of feeding material with a high concentration of NaOH was also appraised in the direction of perceiving optimum range of pH during the digestion under mesophilic and thermophilic conditions. The supplication of higher concentration of NaOH engendered the greater decomposition of organic contents of rice husk and maize straw. In response to these more available substrates and activated growth and reproduction of microorganisms synchronized the pH value accordingly (Kang *et al.*, 2018). It was also observed that after NaOH pretreatment the pH value can fluctuate within an appropriate range which is beneficial to the activities of microorganisms (Wang *et al.*, 2017). It was concluded from the consequences that the optimal variation range of pH was observed in reactor B which was capable of producing sustainable biogas production and accumulated methane production. It was also concluded that there was a little effect of mesophilic and thermophilic temperature on the variation trend of pH value on the same reactor.

The TAN concentrations were considered censorious constituents during the aerobic fermentation process because the fatty acids owned the capability of entering in the cell which caused the disruption in the stability of sodium and potassium metabolism (Thushari *et al.*, 2018). It can be observed from the consequences that the value of TAN is increasing in all three reactors under both mesophilic and thermophilic conditions in the consideration of rice husk. Almost corresponding tendency was observed in the examination of maize straw under mesophilic and thermophilic conditions in all three reactors. The consideration of results indicated that under thermophilic conditions the liberation of fatty acids was inflated into the environment and was considered consistent under thermodynamics. As the concentration of TAN crossed the optimal limit in reactor C the daily production of methane yield started to decrease and ultimately stop the working of the aerobic system (Wu *et al.*, 2016). It can be concluded that the total ammonia nitrogen and fatty acids are considered vital

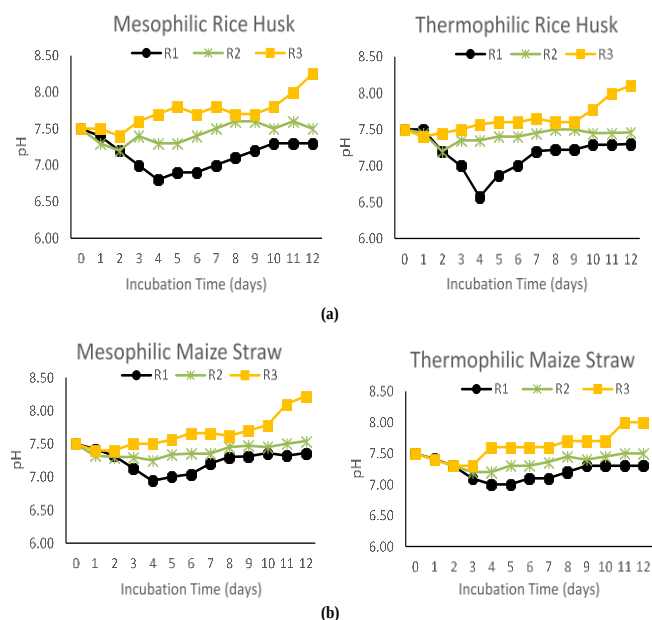


Figure 9. The pH variations of rice husk (a) and maize straw (b)

inhabitants in the direction of sustainable methane production.

Conclusion: The present study focused on biogas production potential of pretreated maize straw and rice husk under mesophilic (31-37°C) and thermophilic (46-55°C) temperature range. The acid-base pretreatment showed considerable lignin degradation with maximum degradation by using 6% NaOH which was recommended for further biogas production experiment by using multistage bioreactor for inline batch production of biogas. By keeping the initial pH 7.5 of each reactor, pH variations showed that pH substantially decreased initially upto 6.5 with for 4-5 days and increased up 8.25 for the rest of the experiment. Mesophilic biogas production was 11% higher for maize and 18% higher for rice husk as compared to thermophilic production. The average methane contents were 49% for maize and 57% for rice husk. Keeping in view all these facts it is concluded that maize straw pretreated with 6% NaOH is a suitable feed stalk for biogas production under mesophilic temperature conditions.

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