Kinetics of Anaerobic Digestion of Water Hyacinth Using Poultry Litter as Inoculum

Jagadish H. Patil, Malourdu Antony Raj, P. L. Muralidhara, S. M. Desai, and G. K. Mahadeva Raju

Abstract—**Water hyacinth (Eichhornia crassipes) is one of the fastest growing aquatic weed known to man. They are free-floating perennial aquatic plants with broad, thick, glossy, ovate leaves with long, spongy and bulbous stalks. The feathery, freely hanging roots are purple-black. In India, water hyacinth is largely found in the Kerala backwaters. But, this natural beauty can be quite deceiving. It chokes waterways, blocks sunlight from reaching native aquatic plants, starves the water of oxygen, killing fish (or turtles) and is a habitat for disease causing vectors. Attempts to curb its fast spread have proved to be futile. However this deleterious weed is a potential source of biomass to produce biogas, which is an eco-friendly biofuel. In this study, poultry litter was used as inoculum at mesophilic conditions. A series of laboratory experiments using 0.25l bio-digesters were performed in batch operation mode and modified Gompertz equation was fitted. The kinetic parameters, biogas yield potential (*P*), the maximum biogas production rate (R_m) and the duration of lag phase (λ) were estimated in each case. The results show that Poultry Litter Inoculum (PLI) improved biogas yield significantly and increased biogas yield nearly two times when compared to water hyacinth substrate without PLI.**

Index Terms—**Anaerobic digestion, water hyacinth, inoculum, modified gompertz equation.**

I. Introduction

Biogas, a clean and renewable form of energy could very well substitute (especially in the rural sector) for conventional sources of energy (fossil fuels, oil, etc.) which are causing ecological-environmental problems and at the same time depleting at a faster rate [1]. Renewable energy plays an important role in reducing the greenhouse gases; particularly energy from biomass could contribute significantly as it is a ‘‘carbon neutral” fuel [2]. Anaerobic digestion (AD) is an environmental friendly biological process in which microorganisms work synergistically to convert organic wastes into biogas and a stable product (soil conditioner) for agricultural practices without any detrimental effects on the environment. The mankind has explored AD of organic materials greatly to generate biogas which comprises of methane (50 to 70%), carbon dioxide (30 to 40%) and traces of other gases which include CO, H_2S, NH_3, H_2, O_2 and water vapor etc [3]. This process involves four major steps: hydrolysis, acidogenesis, acetogenesis and methanogenesis [4]-[5]. A simplified generic chemical equation for the overall processes is given by (1)

$$C_6H_{12}O_6 \rightarrow 3CO_2 + 3CH_4 \quad (1)$$

Water hyacinth and its tendency of fast growth would have a great potential if seen as a raw material for biogas production as it is rich in nitrogen, essential nutrients and has a high content of fermentable matter [6]. Apart from biogas the residual slurry obtained after anaerobic digestion is rich in essential inorganic elements which are good soil conditioners [7]. Numerous studies have been conducted by researchers to improve biomass conversion efficiency and biogas yield. The techniques include using different pre-treatment methods [8]; improving substrate composition by co-digesting with other substrates [9]; optimization of dilution on biomethanation of fresh water hyacinth [10] and effects of particle size, plant nitrogen content and inoculum volume [11]. This study focuses on the use of PLI in AD of water hyacinth. 4g of completely dried and ground water hyacinth were fed to each bio-digester and mixed with PLI and water in different combinations resulting in five different fermentation slurries. The digesters were labelled as PLI-0, PLI-25, PLI-50, PLI-75 and PLI-100 with their corresponding Total Solid (TS) content as 3.8%, 5.5%, 7.2%, 8.9% and 10.6%. Digester PL-100 with PLI was considered as control digester. The best performance for biogas production was from the digester PLI-75 followed by PLI-50 and PLI-100. The data obtained from these experiments were further used to fit and check the fitness of the Modified Gompertz equation (MGE) that describes kinetics of AD process. . These results indicate TS plays an important role and optimum TS range is 7 to 9%.

II. Materials and Methods

A. Sample Collection

Water hyacinth used for the study was obtained from silver lake at HBR layout (Bangalore, Karnataka, India).Poultry litter was collected from J M J Chicken Centre (Bangalore, Karnataka, India).

B. Sample Analysis

1) *pH analysis*: pH was measured by pH meter which consists of a potentiometer, a glass electrode, a reference electrode and a temperature compensating device. Electrodes were connected to the pH meter and were calibrated using buffer solutions before pH analysis.

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2) Total solids (TS) and total Volatile Solids (VS): TS were determined at 104 ^{0}C to constant weight (Standard method part 2540 B) and VS were measured by the loss on ignition of the dried sample at 550 ^{0}C (Standard method part 2540 E) [12].

3) *Biogas analysis*: Gas chromatograph (Chemito 1000) equipped with a thermal conductivity detector was used to analyze the biogas sample. Hydrogen was used as carrier gas (25 ml/min) with Porapak Q column. Standard calibration gas mixture was used for calibration. Biogas samples were collected in rubber bladders; the sample and standard were injected using a gas tight syringe into the gas chromatograph. The parameters were set at oven temperature of 40^{0}C, detection temperature of 80^{0}C and the detector current of 180 mA. The concentrations of different components were calculated using (2):

$$\%\text{ of X} = \left(\frac{\text{Area of X in sample}}{\text{Area of X in standard}}\right)\times\%\text{ of X in standard} \tag{2}$$

C. Biomethanation Unit

Biomethanation unit consists of a temperature controlled thermo bath which is maintained at the mesophilic temperature range of 30°C to 35°C. It has a battery of bio-digesters. Each bio-digester is connected to a graduated gas collector by means of a connecting tube. Each of the gas collectors are in turn immersed in a trough of water to ensure complete sealing. A stand holds all the gas collectors. Biogas evolved is collected by the downward displacement of water.

D. Inoculum Preparation

In a 2.5 L glass bottle, 660 gm of poultry litter was mixed with 1340 gm of water to obtain a slurry of 7% TS. The bottle was maintained at 35^{0}C and was fitted with a rubber cork having one hole. A glass tube was inserted in the hole which remained above the layer of the slurry. The other end was connected with Teflon tubing, the outlet of which was dipped in a container filled with water. The gas produced during the incubation period could bubble through the water but no air would enter the slurry thus, maintaining the anaerobic condition. After an incubation period of 50 days, the biodegradable volatile organic matter contained in the slurry almost gets completely degraded and can be used as inoculums [13].

E. Fermentation Slurry Preparation

Fresh water hyacinth (leaves, stem and root) on collection was chopped to small sizes of about 2cm and allowed to dry under the sun for a period of 7 days. They were then dried in an oven at 60^{0}C for 6hours. This oven-dried water hyacinth was ground to fine particles using a grinding mill. The influence of PLI to biogas production was studied by varying primary sludge inoculums and total solids content in the bio-digesters. A series of laboratory experiments using 0.25l bio-digesters were performed in batch operation mode. Each bio-digester was fed with 3.21g of volatile solid* (VS) by adding 4g of finely dried and ground water hyacinth. This was mixed with various combinations of PLI and water, resulting in five different fermentation slurries PLI-0, PLI-25, PLI-50, PLI-75 and PLI-100 with total solids content of 3.8, 5.5, 7.2, 8.9 and 10.6% respectively. Digester PL-100 fed with PLI without water hyacinth was considered as control digester. All digesters were given 0.3ml of 10% by volume of acetic acid. Table I presents detailed content of digesters. AD of these digesters was carried out in duplication with a retention period of 60 days in the mesophilic range. Cumulative biogas production, slurry temperatures were monitored throughout the period of the study.

F. Modified Gompertz Equation

The kinetic data obtained from all digesters were checked for the fitness of modified Gompertz equation [14]. The modified Gompertz equation, that gives cumulative biogas production from batch digesters assuming that biogas production, is a function of bacterial growth. The modified Gompertz equation is given by (3)

$$M = P\times\exp\left\{-\exp\left[\frac{R_m\times e}{P}(\lambda - t)+1\right]\right\} \tag{3}$$

where

M Cumulative biogas production, l/(g VS) at any time t
P Biogas yield potential, l/(g VS)
R_m Maximum biogas production rate, l/(g VS d)
λ Duration of lag phase, d (days)
t Time at which cumulative biogas production M is calculated, d

The parameters P, R_m and λ were estimated for each of the digesters using POLYMATH software. These parameters were determined for the best fit.

TABLE I: CONTENTS OF DIGESTERS

Digester	Water hyacinth	Water	PLI	Acetic acid (ml)
PLI-0	4 g.	100 g.	-	0.3
PLI-25	4 g.	75 g.	25 g.	0.3
PLI-50	4 g.	50 g.	50 g.	0.3
PLI-75	4 g.	25 g.	75 g.	0.3
PLI-00	4 g.	-	100 g.	0.3
PL-100	-	-	100 g.	0.3

*Biodegradable VS from PSI were negligible and were not accounted for VS added to each digester.

III. RESULTS AND DISCUSSION

A. Solids and pH Analysis

Total solids (TS) are the sum of suspended solids and dissolved solids. Total solids analysis and pH are important for assessing anaerobic digester efficiencies. TS analysis is done using standard methods while pH is measured using pH meter (Systronics). The TS are composed of two components, volatile solids and fixed solids. The VS are organic portion of TS that biodegrade anaerobically. Table II gives the solid analysis and pH data of poultry litter and water hyacinth.TS and VS are calculated using (4) and (5).

$$\text{TS, \%} = \frac{(A-B)}{(D-B)}\times 100 \tag{4}$$

$$\text{VS, \%} = \frac{(A-C)}{(A-B)} \times 100 \tag{5}$$

where
A is weight of dish + dried sample at 104^{0}C (g)
B is weight of dish (g)
C is weight of dish + sample after ignition at 550^{0}C (g)
D is weight of dish + wet sample (g)

B. The Influence of PLI to Cumulative Biogas Production

The cumulative biogas production with time for all the digesters is shown in Fig.1. It can be observed from Fig.1 biogas production rate tends to obey sigmoid function (S curve) as it generally occurs in batch growth curve. Fermentation slurries PLI-25, PLI-50, PLI-75 and PLI-100, exhibit higher biogas production than substrate that contains water hyacinth and water (PLI-0). In other words, specific biogas production per gram volatile solid added to digesters PLI-25, PLI-50, PLI-75 and PLI-100 are higher than PLI-0. This suggests that high concentration of anaerobic bacteria content in PLI works effectively to degrade organic substrate from water hyacinth. Biogas production is slow at the beginning and end of observation; this indicates that the biogas produced in batch condition corresponds to specific growth rate of methanogenic bacteria [15]. During the first 8 days, biogas production is low due to the lag phase of microbial growth. After 10 days, biogas production significantly increased due to exponential growth of microorganisms. After 35 days, biogas production decreased due to stationary phase of microbial growth. Finally, the most important finding from this research is that the PLI seeded to bio-digester has significant effect on cumulative biogas production.

C. Effect of Total Solids content on Biogas Production

The effect of TS content on biogas production was studied by varying TS from 3.8% to 10.6 %. Fig. 1 shows cumulative biogas production of PLI-25, PLI-50, PLI-75 and PLI-100 as 0.36, 0.44, 0.48 and 0.41 l/gVS respectively, while sample PLI-0 with 0 % PLI gave cumulative biogas production of 0.23 l/gVS. The best performance of biogas production is given by PLI-75 (TS of 8.9%) followed by PLI-50 (TS of 7.2%) and PLI-100 (TS of 10.6%). These results suggest that, TS content affects the biogas yield. This is similar to the findings of Balsam [16] and Zennaki et al., [17] that the optimum solid content is in the range 7-9 % for highest biogas production. Furthermore, Baserja [18] reported that the process was unstable below a total solids level of 7% (of manure) while a level of 10% caused an overloading of the fermenter.

These results are expected due to the function of water in biodigester since the TS content will directly correspond to water content. According to Sadaka and Engler [19] water content is one of very important parameter affecting anaerobic digestion of solid wastes. There are two main reasons (a). Water makes possible the movement and growth of bacteria facilitating the dissolution and transport of nutrient; and (b) water reduces the limitation of mass transfer of non homogenous or particulate substrate.

From Fig. 1 it can be observed that PL-100 yield less quantity of biogas in comparison with PLI-25, PLI-50, PLI-75 and PLI-100. Hence, in all digesters biogas produced was originated only from water hyacinth.

TABLE II: SOLID ANALYSIS AND PH DATA

Material	% TS	% VS	pH
Poultry litter	21.21	83.47	7.1
Water hyacinth	16.89	82.85	6.4

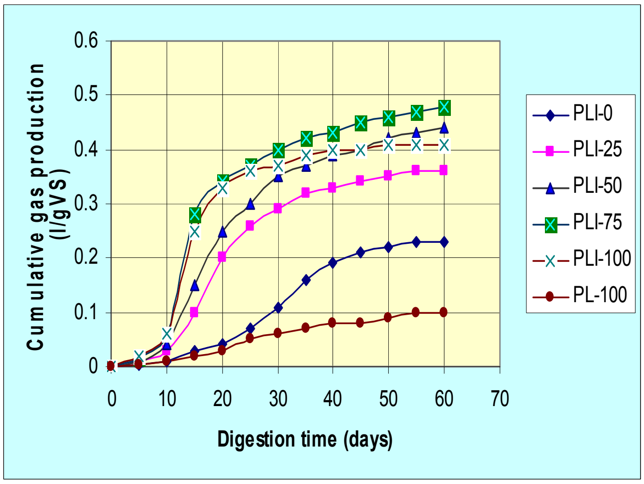

Fig. 1. Daily biogas production

D. Kinetics of Biogas Production

With an assumption that biogas produced is a function of bacterial growth in batch digesters, modified Gompertz equation relates cumulative biogas production and the time of digestion through biogas yield potential (*P*), the maximum biogas production rate (R_m) and the duration of lag phase (λ) [20]. To analytically quantify parameters of batch growth curve, a modified Gompertz equation was fitted to the cumulative biogas production data. Values of parameters obtained are listed in Table III. The best fit to Gompertz equation is compared with experimental data in Fig. 2, 3, 4, 5, 6 and 7. From Table III the following observations were made.

- Shortest lag phase (λ) was exhibited by PLI-75, 6.625 days. While the largest lag phase (λ) was exhibited by PLI-0, 15.28 days. This suggests that PLI-0 does not have the essential microbes to produce biogas early. However when PLI is added the yield is faster.
- The biogas production rate (R_m) for PL-100 is the lowest of 0.0027 l/(g VS d) and the highest is shown by PLI-100 with a value of 0.0328 l/(g VS d). This indicates that anaerobic digestion with PLI plays a vital role in enhancing the biogas production rate.
- The biogas yield potential (*P*) is maximum for PLI-75 of 0.4494 l/(g VS) and is minimum for PL-100 of 0.1065 l/(g VS).
- PLI-75 produced the maximum amount of biogas of 0.48 l/(g VS), followed by PLI-50 with a value of 0.44 l/(g VS). The least amount of biogas was produced by PL-100 of 0.1065 l/(g VS).
- From Fig. 2, 3, 4, 5, 6 and 7 it is clear that Modified Gompertz equation fits well to all the experimental

data. In particular the best fit is for PLI-25 which is described by higher R^2 and lower Rmsd values.

TABLE III: SUMMARY OF KINETIC DATA

Digester	Biogas Yield l/(gVS)	Modified Gompertz parameters (model)			R^2	Rmsd
		P, l/(gVS)	R_m, l/(gVS d)	λ, (d)		
PLI-0	0.23	0.2501	0.0081	15.28	0.9970	0.0018
PLI-25	0.36	0.3540	0.0170	8.756	0.9975	0.0018
PLI-50	0.44	0.4246	0.0185	7.313	0.9951	0.0031
PLI-75	0.48	0.4494	0.0279	6.625	0.9809	0.0065
PLI-100	0.41	0.3988	0.0328	7.826	0.9932	0.0035
PL-100	0.10	0.1065	0.0027	8.037	0.9945	0.0007

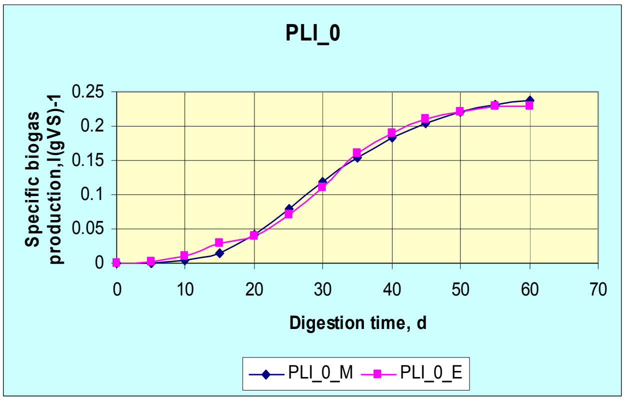

Fig. 2. Modified Gompertz equation fit for PSI-0

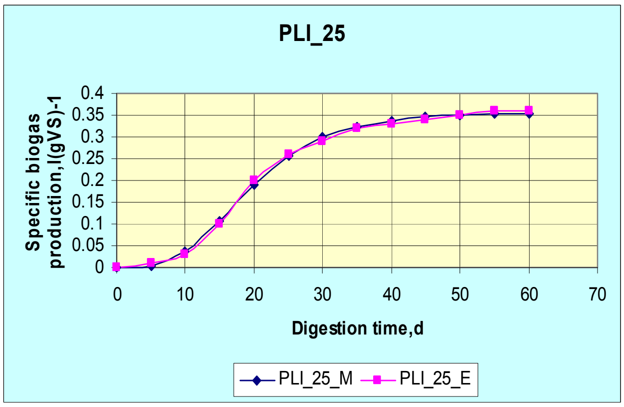

Fig. 3. Modified Gompertz equation fit for PSI-25

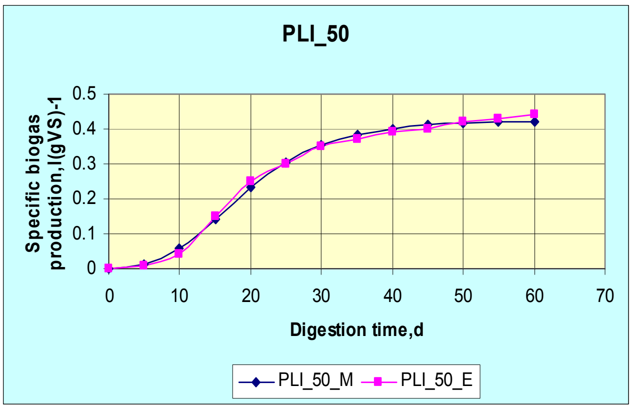

Fig. 4. Modified Gompertz equation fit for PSI-50

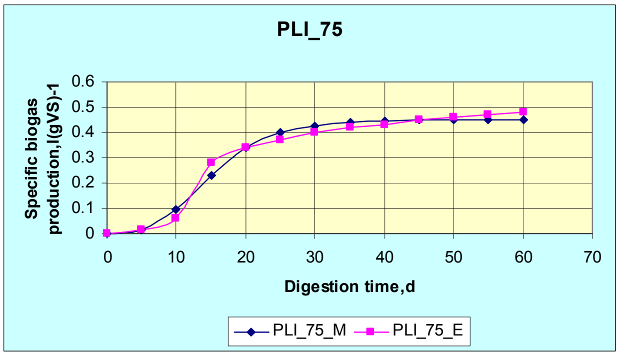

Fig. 5. Modified Gompertz equation fit for PSI-75

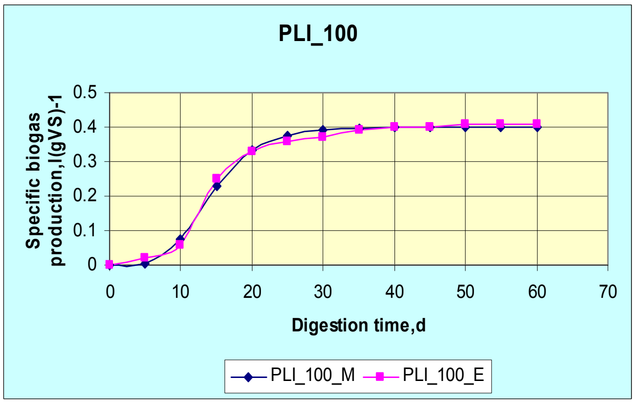

Fig. 6. Modified Gompertz equation fit for PSI-100

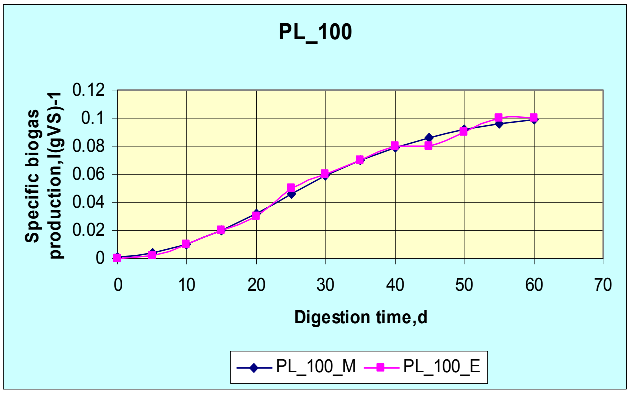

Fig. 7. Modified Gompertz equation fit for PS-100

IV. CONCLUSION

The conclusions drawn in context to the use of PLI were as follows:

- In today's energy demanding lifestyle, water hyacinth proves to be a promising renewable source of energy in the form of biogas.
- Anaerobic digestion of water hyacinth with PLI increased the cumulative biogas yield almost two folds when compared to feed without PLI.
- The best performance of biogas generation was observed in digesters PLI-75 and PLI-50.
- The graphs have verified that Modified Gompertz equation best describes cumulative gas produced as a function of retention time.

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