Optimizing Biogas Production from Water Hyacinth *(Eichhornia crassipes)* Through Varied Volatile Solids Loading Rates in Batch Anaerobic Digestion

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Abstract- Water hyacinth (WH) is promising biomass for renewable energy production when combined with livestock manure. However, previous studies have only investigated biogas production at low loading rates. In this study, we investigated the effects of various volatile solid (VS) loading rates (0.5%, 1.0%, 1.5%, 2.0%, and 2.5%) on biogas production using WH in batch bioreactors. pH, temperature (ºC), biogas production volume in five days (L), the total volume of biogas production (L), biogas yield ($L/kgVS_{added}$), and methane concentration (%) were measured over 45 days. The results showed that pH and temperature were suitable for methane production. The VS loading rate of 2.5%VS achieved the highest volume of biogas production in five days, while the loading rate of 0.5%VS recorded the lowest volume. Although, the 2.5%VS treatment had the highest cumulative biogas volume, and was significantly different from the other treatments ($p=0.001<0.05$). But, the biogas yield in the 2.5%VS treatment was the lowest yield, and significantly different from the remaining treatments ($p=0.001<0.05$). The study recommends using a 2.0%VS loading rate for optimal biogas when using WH for biogas production. These findings can contribute to the development of sustainable energy sources using WH as a material source for biogas production in the Vietnamese Mekong Delta.

Keywords Batch anaerobic digestion, renewable energy, volatile solid, water hyacinth.

1. Introduction

Anaerobic digestion (AD) is a biological process that decomposes organic matter in absence oxygen, resulting in the production of biogas, a renewable and clean source of energy [1]. It shows effectiveness in improving the rural ecological environment [2]. In additon, methane (CH_4) gas can be utilized as an alternative enerfy source for household activities such as cooking [3-5], provide a large source of electricity in big farming, and contribute to the nutrient contents (N, P, and K) for upland crops and rice paddy field [6-8]. In previously reported that the livestock manures (i.e., cattle manure, goat manure, poultry manure) and biomass (i.e., water hyacinth) used as greatly popular materials in producing biogas [9-10].

Water hyacinth (WH) is a widely utilized biomass source in the VMD due to its ability to easily survive in rivers, lakes, and canals in the region. Optimal conditions for WH growth include a pH range of 6.5-8.8, temperature range of 28-30 ºC, and the presence of nitrogen (20 mg/L), phosphorus (3 mg/L) and potassium (53 mg/L) [11]. Under favorable conditions, WH can grow rapidly, doubling in size every 7 days, and yield 90 to 140 tons of fresh weight per hectare each year [12-13]. Thus, WH represents a significant biomass source in the VMD for CH_4 production in the long term [10].

Previous studies have shown that WH can be used as a supplementary substrate for CH_4 production when combined with pig manure, but these experiments were primarily limited to using 1.0%VS of WH [14, 15]. In this study, we conducted a batch anaerobic digestion experiment to investigate whether increasing the loaded substrate rate of WH would increase biogas production. We used WH with five VS levels ranging from 0.5%VS to 2.5%VS, and measured pH, temperature, daily biogas production volume, cumulative biogas volume, biogas yield, and CH_4 concentration in each reactor over a 45-day period.

2. Materials and Methods

2.1. Material

The WH biomass used in this study was collected from a river and dried under sunlight for approximately three days. To enhance the decomposition process, the WH was chopped into 2 cm pieces before being used in the experiment. To ensure the uniformity of samples, the WH was thoroughly mixed. The characteristics of the WH, including moisture content (92.7%), VS content (75.6%), total organic carbon (TOC; 48.9%), total organic nitrogen (TON; 1.73%), and C/N ratio (28.3), were analyzed to determine its suitability as a substrate for anaerobic digestion.

2.2. Experimental Design

To conduct the batch anaerobic digestion experiment, 1.5 liters (L) plastic bottles were used. In which, 1.2 L was allocated for liquid and the remaining 0.3 L for the gases produced, such as CH_4, CO_2, and others (Fig. 1). The experiment lasted for 45 days, during which five treatments with varying VS ratios were tested: 0.5%VS, 1.0%VS, 1.5%VS, 2.0%VS, and 2.5%VS. Each treatment had three replicates, and the reactors were randomly assigned. In each reactor, 0.2 L of stability biogas digester was added as a microbial source, and tap water was used to adjust the water volume to 1.2 L. The properties of the inoculum were analyzed, and it was found to have a pH of 6.78, alkalinity of 213.5 mg $CaCO_3$/L, ammonium concentration of 65.5 mg/L, phosphate concentration of 104.6 mg/L, total solids of 40.2 mg/L, and VS of 26.5 mg/L. The loaded total solid (TS), VS, and weight for each treatment are summarized in Table 1.

2.3. Analytical Methods

The properties of WH were analyzed in accordance with Standard Methods [16], which allowed for the determination of moisture content, TS, VS, TOC, TON, and C/N ratio. The temperature (°C) and pH values in each reactor were manually measured using a portable meter (DKK TOA, Japan) for the first 10 days, and then every five days until the end of the 45 days experiment. The volume of biogas produced was collected daily in aluminum bags and measured using a gas meter (Ritter TG 05, Germany). CH_4 concentration was analyzed at the 10 days of the experiment and then every five days using a biogas meter (Gauge meter GA 5000, Geotech Inc., England). These measurements allowed for a comprehensive analysis of the biogas production process and the quality of the produced biogas. The biogas yield was calculated by the below formula:

Biogas yield (L/kgVS) = Volume of biogas produced (L)/VS loaded (kg)

Where, volume of biogas produced (L) is the total volume of biogas produced in the reactor during the experiment (45 days); VS loaded (kg) is the VS content loaded into the reactor at the beginning (VS_0) - the VS content in batch bioreactors after 45 days of the experiment (VS_{45}).

Table 1. Weight of TS, VS and material loaded in each bioreactor

Reactors	Added TS (g)	Added VS (g)	Wet weight (g)
0.5%VS	6	4.53	83.7
1.0%VS	12	9.07	165.39
1.5%VS	18	13.60	248.09
2.0%VS	24	18.14	330.78
2.5%VS	30	22.67	413.48

Fig. 1. Model of bioreactor producing biogas production (left) and raw material (right)

2.4. Statistical Analysis

At the end of experiment, data was analyzed to compare the difference between all treatments by derermining the total biogas production and biogas yield. To determine significant differences between the five treatments, a one-way factor analysis of variance (ANOVA) was performed. The Duncan's test was used to identify the significant differences means at a 5% level of significance ($p<0.05$). All figures were prepared in R Studio software. The statistical analyses were conducted using SPSS (version 26.0, SPSS Inc., Chicago, USA).

3. Results

3.1. Variation of Temperature within the Reactors during the Experimentation

The temperature variation in the incubation bottles was presented in Fig. 2. During the 45 days experiment, the water temperature ranged from 27.0 ºC to 34.6 ºC. In the first 15 days, the temperature fluctuated from 27.0 to 31.8 ºC. Subsequently, the water temperature continued to rise steadily until the end of the experiment, with a range of 30.0 - 34.6 ºC. These findings indicate that the temperature inside the incubation bottles was suitable for the anaerobic digestions process, which could have contributed to the biogas production.

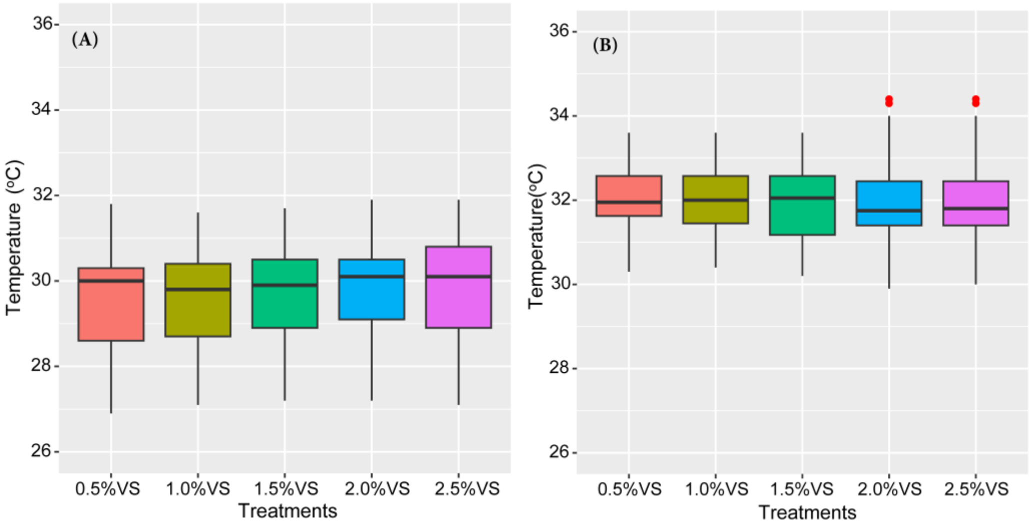

Fig. 2. Temperature inside of batch bioreactors from 0-15 days (A) and 16-45 days (B)

3.2. Variation of pH Value within the Reactors during the Experimentation

Fig. 3A illustrates the pH values of all bottles during the first 15 days of fermentation. The results showed that the pH values decreased rapidly below 5.5 during this period, except 0.5%VS experiment. On the first day of the experiment, the pH values in the 0.5%VS and 1.0%VS treatments were higher compared to the 1.5%VS, 2.0%VS, and 2.5%VS treatments. However, the pH values in the 0.5%VS and 1.0%VS treatments quickly dropped from 6.3 to 4.9 within the first 15 days of the experiment, similar to the other treatments. Meanwhile, the pH values in the 1.5%VS, 2.0%VS, and 2.5%VS treatments slightly decreased from 5.3 to 4.8 during the same period. The findings indicated that the rates of volatile solid had an impact on the pH values, where increasing the VS rates led to a reduction in the pH values.

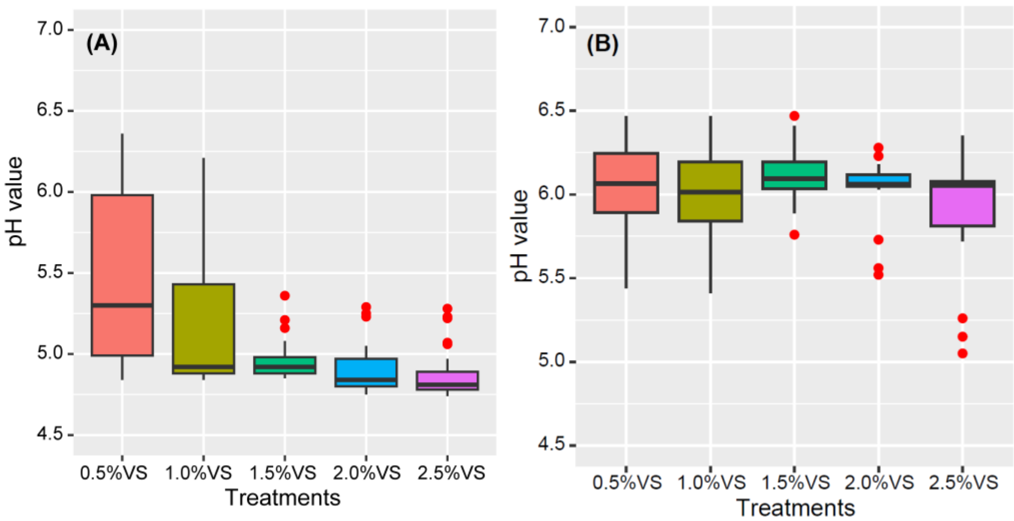

Fig. 3. pH value inside of batch bioreactors from 0-15 days (A) and 16-45 days (B).

3.3. The Daily Biogas Production

The study monitored the daily biogas production in batch bioreactors, as shown in Fig. 4, with biogas volume recorded every 5 days. During the initial 10 days of the experiment, the volume of biogas produced varied across the treatments, with 0.07 L, 0.09 L, 0.24 L, 0.27 L, and 0.22 L for the 0.5%VS, 1.0%VS, 1.5%VS, 2.0%VS, and 2.5%VS treatments, respectively. Subsequently, the volume of biogas produced was recorded every 5 days for the remaining duration of the experiment (45 days). The 2.5%VS treatment recorded the highest daily biogas production volume of 1.7 L compared to the other treatments, with 0.62 L, 0.90 L, 1.15 L, and 1.26 L for 0.5%VS, 1.0%VS, 1.5%VS, and 2.0%VS treatments, respectively. These findings suggest that the level of VS significantly influenced the production of biogas.

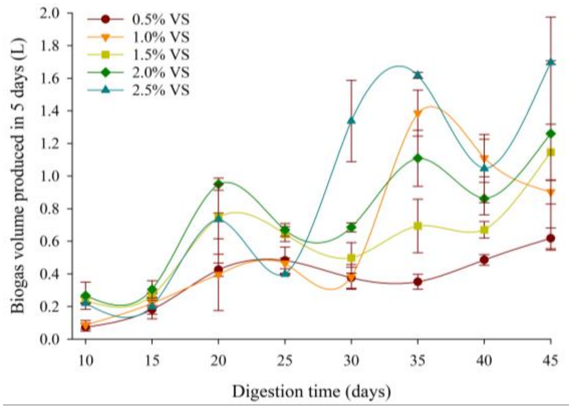

Fig. 4. The biogas volume produced in batch bioreactors with different VS. Value is presented as mean±SD (n=3)

3.4. Total volume of Biogas Production and Biogas Yield

The cumulative biogas volumes in the anaerobic digestion reactors were measured over 45 days, and significant differences were observed among the treatments (*p<0.05*; Fig. 5A). The results revealed that the cumulative biogas volumes could be classified into four groups based on their mean values: the 2.5%VS treatment had the highest value (7.3±0.2 L) and was significantly different from all other treatments (*p<0.05*). In contrast, the 0.5%VS treatment had the lowest cumulative biogas volumes (3.0±0.3 L). The other treatments (1.0%VS, 1.5%VS, and 2.0%VS) were grouped in the second and third positions, with the cumulative biogas volumes values of 4.95±0.4 L, 4.95±0.01 L, and 6.2±0.9 L, respectively. These results indicated that the cumulative biogas volumes were strongly influenced by the levels of VS.

The biogas yield data presented in Fig. 5B. The yield for each treatment was calculated by subtracing the volatile solids (VS_{added}) lost during the digestion process from the total biogas production. As shown in Fig. 5B, the biogas yield of the 0.5%VS, 1.0%VS, 1.5%VS, 2.0%VS, and 2.5%VS treatments after 45 days of digestion were 146.2±4.0, 152.4±4.3, 150.7±3.8, 154.5±1.6 and 135.5±1.5 $L/kgVS_{added}$, respectively. While the cumulative biogas volumes of the 2.5%VS treatment were the highest and sifnificantly different from the other treatments (*p<0.05*; Fig. 5B), the biogas yield of the 2.5%VS treatment was the lowest and significantly different from the other treatments (*p<0.05*; Fig. 5B). Interestingly, the treatment of 2.0%VS had the highest biogas yield among all treatments.

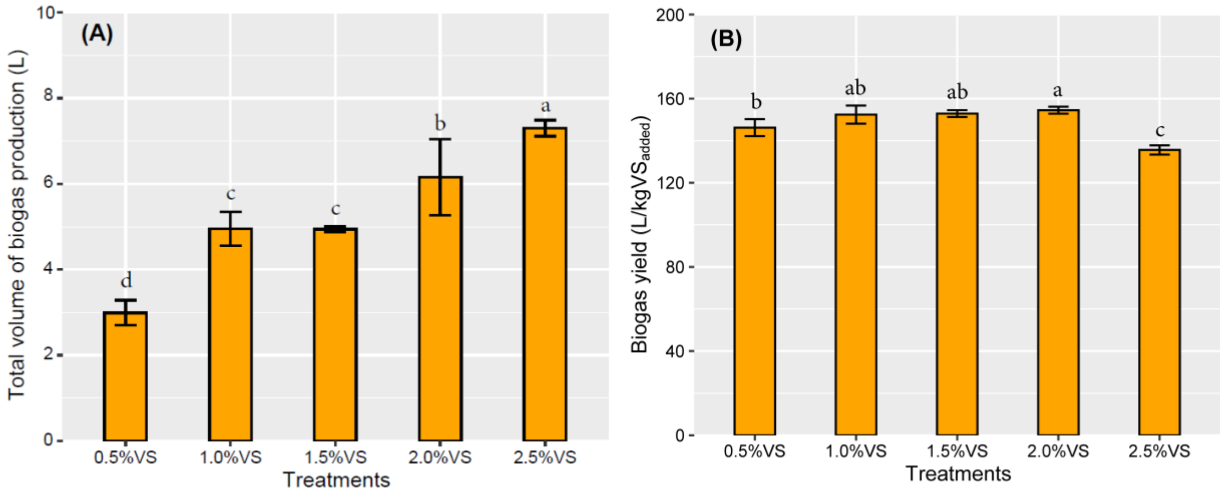

Fig. 5. Total volume of biogas production (A) and biogas yield (B) in batch bioreactors. Value is presented as mean±SD (n=3).

3.5. Methane Concentration (%)

The CH_4 content during the experiment is presented in Fig. 6, which shows the average CH_4 content recorded every 5 days over the 45 days experiment period for all treatments. The experiment time was divided into three phases based on the measured CH_4 content. In the first 10 days, the lowest CH_4 content was recorded, ranging from 2.5 to 5.6%. From 15 days of the experiment, the CH_4 content increased and reached its highest value from 25-30 days. Finally, from 30-45 days, the CH_4 concentration decreased but still remained above 40%.

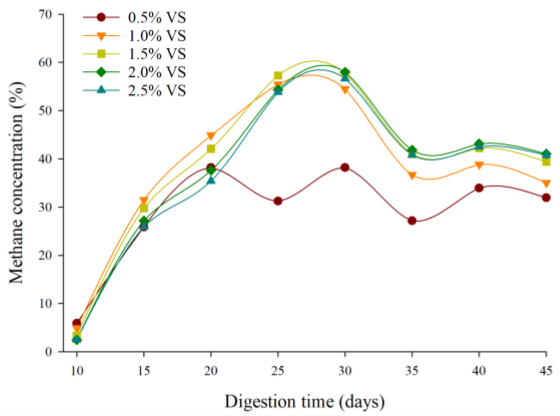

Fig. 6. Methane concentration in batch bioreactors.

4. Discussion

4.1. Effect of Different Volatile Solids Levels on the pH Value and Temperature

The water temperature and pH value are important factors in anaerobic digestion, affecting the growth and activity of methanogenic bacteria [17-20]. In this study, we recored the water temperature within the reactors over a period of 45 days, as shown in Fig. 2. There are different temperature ranges at which anaerobic digestion can occur, including psychrophilic (<30°C), mesophilic (30-40 °C), and thermophilic (50-60 °C) [17]. Previous studies have reported that the water temperature inside anaerobic digesters can range from 22.6 - 29.7 ºC [21] and from 24.2 - 31.1 ºC [10]. Our results showed that the temperature range inside all bottles was suitable for the growth of methane-producing bacteria. These findings are consistent with previous study in the Mekong Delta that used water hyacinth and water lettuce for biogas production under batch/semi-continuous digestion conditions [21].

In addition to water temperature, pH is also a critical factor in the growth of microbes during anaerobic digestion. The pH value should be maintained within a range of 6.8 - 7.2 [21], as it affects the efficiency of CH_4 production. However, Jain and Mattiasson [22] found that a pH value higher than 5.0 results in CH_4 production efficiency of more than 75%. Anaerobic digestion is a four-stage process, as described in [23], consisting of hydrolysis, acidogenesis, acetogenesis, and methane production. pH values generally decrease during the acidogenesis and acidification stages. Additionally, the results in [24] indicated that pH values can decrease during the hydrolysis and acidogenesis phases. Although pH values decreased during the first stage in this study, it sharply increased from day 15 until 45 days and was maintained within suitable conditions for methane-producing bacteria (Fig. 3). Previous studies have reported pH values ranging from 6.6 to 7.6 [21]. In general, the pH values in this study were consistent with previous studies and suitable for biogas production

4.2. Effect of Different Volatile Solids Levels on Biogas Production

The daily biogas production in this study was monitored every 5 days (Fig. 4). The findings were consistent with the research [15], where the biogas production ability of WH was dependent on the VS concentration. It was observed that the organic loading rate of WH ranging from 2.0 to 2.5 gVS/L (equivalent to 2.0 - 2.5%VS) resulted in the highest biogas production. Furthermore, the total biogas production showed a trend similar to the daily biogas production, with the CBV of the 1.5%VS treatment and 2.0%VS treatment being not significantly different ($p>0.05$) but significantly different from the other treatments (i.e., VS from 0.5-2.0%; $p<0.05$). This observation is consistent with previous study in the Mekong Delta, which reported that WH is a promising biomass source for biogas production, especially when mixed with livestock manure at a loading rate of 1.0%VS and 100%WH used [21]. This study highlights the efficacy of increasing the loading rate in the biogas production process, with a recommended VS content of 2.0% in the batch digestion experiment. Additionally, we found a positive linear relationship between cumulative biogas production and VS rates in the experiment, with a coefficient of determination (R^2 = 0.94; data not shown). In general, the biogas volume would increase with the increase in VS in reactors.

The biogas yield measures the amount of gas produced per unit of degraded substrate, calculated after 45 days of the experiment (Fig. 5B). It was observed that biogas yield decreased when the reactors were applied with more than 2.0% VS. This could be attributed to the degradation of WH during the first phase, where various bacteria ferment VS into volatile fatty acids and eventually into acetic acid through acetogenesis [25]. In addition, there is a positive relationship between pH and volatile fatty acids; as the pH decreases, volatile fatty acid accumulation in the anaerobic digester increases [26]. In this study, the pH value in the 2.5% VS treatment reached its lowest value in the first 25 days of the experiment, potentially limiting biogas production volume and inhibiting the activities of methanogenesis bacteria [27]. However, when compared to previous studies that used WH with a loading rate of 1% VS for biogas production, the biogas yield ranged from 95.7 – 140.2 $L/kgVS_{added}$ at the 30 days [28], and from 286 – 292 $L/kgVS_{added}$ [29]. These findings suggest that while cumulative biogas production improves with volatile solids, biogas yield is most efficient with VS rates ranging from 1.0 to 2.0% VS. Overall, the results of this study are consistent with those of previous studies.

4.3. Feasibility of Using Methane for Cooking and Slurry for Agricultural Cultivation

Methane is a crucial component of biogas for energy generation, and its concentration is vital for determination the effectiveness of the anaerobic digestion process. The initial phase of the anaerobic process is the hydrolysis and acidogenesis phase, during which microorganisms primarily convert organic matter into volatile fatty acids, leading to low methane levels. In contrast, the second phase is dominated by methanogenic microbes, which convert the volatile fatty acids to biogas, leading to a rise in methane levels [17, 30]. In our study, the CH_4 concentration was consistent with previous reports and consistently higher than 40%, the recommended minimum level for household activities [31-33]. The results in [28] also observed a similar trend, with CH_4 concentration ranging from 17 to 37% during the first phase, then stabilizing at 51 - 63% by day 60. Similarly, Khanh [21] also recorded methane concentration ranging from 31.7 - 60.7% and 32-47.5% [34]. Overall, the methane concentrations in all reactors were appropriate for household activities such as cooking and lighting, as reported in previous studies [28-29; 35].

In addition to replacing dried fireworks for cooking, the AD also produces bio-slurry, which is a nutrient-rich by product. The high N, P, and K contents in bio-slurry make it an excellent fertilizer for agricultural cultivation [36]. Moreover, biogas effluent from cattle and pig manures has also been showed to be an effective N fertilizer for rice and upland crops due to its high concentrations of N, P, K [6-8; 37]. Therefore, further research should investigate the potential of using the bio-slurry from the WH biogas digester as an N fertilizer to replace synthetic fertilizer under large-scale household conditions.

5. Conclusion

This study provides valuable insights into the potential of using water hyacinth as a material source for biogas production. The experiment demonstrated that the VS loading rate was found to be 2.0% VS, which resulted in the highest biogas yield. The findings of this study could be useful in designing and operating biogas digesters for household or community-scale biogas production, particularly in the Vietnamese Mekong Delta region, where water hyacinth is readily available. Furthermore, the study highlights the potential of using water hyacinth as a sustainable source of renewable energy, which can contribute to reducing environmental pollution.

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