

Anaerobic Digestion of Water Hyacinth (*Eichhornia crassipes*) using Cow Dung Inoculum for Enhanced Biogas Production

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ABSTRACT

Background and Objective: Anaerobic digestion is an environmentally sustainable biological process that converts organic biomass into biogas through the activity of microorganisms under oxygen-free conditions. This study investigated the influence of cow dung inoculum on the anaerobic digestion of Water Hyacinth (*Eichhornia crassipes*) leaves for biogas production. **Materials and Methods:** Fresh leaves of Water Hyacinth (*Eichhornia crassipes*) were collected within Sokoto metropolis and subjected to anaerobic digestion, with cow dung serving as the inoculum in the inoculated treatment. Anaerobic digestion was carried out using inoculated and uninoculated substrate of Water Hyacinth (*Eichhornia crassipes*) leaves by water displacement method, while temperature and pH were determined using pH meter and thermometer. Data were subjected using descriptive analyses. **Results:** The inoculated digester produced a higher cumulative biogas yield (620.50 cm³) compared to the uninoculated digester (240.30 cm³). The inoculated system recorded a shorter retention time (18 days) and higher average daily gas production (34.47 mL/day) compared to the uninoculated digester (26 days; 9.24 mL/day). The pH of the inoculated digester ranged from 6.69 before digestion to 8.11 after digestion, while the uninoculated system ranged from 5.78 to 6.57. The operating temperature ranged between 30 and 35°C, which is within the mesophilic range suitable for anaerobic digestion. Percentage degradation was higher in the inoculated substrate (72.40%) than in the uninoculated substrate (61.30%). **Conclusion:** The study concludes that the addition of cow dung inoculum significantly enhances anaerobic digestion efficiency by increasing biogas yield. Future studies should focus on co-digestion with other aquatic weeds to further improve biogas production.

KEYWORDS

Water hyacinth, pH, temperature, water displacement

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INTRODUCTION

Anaerobic digestion of organic materials such as aquatic weeds, animal dung, plant residues, and municipal wastes produces biogas, a renewable, colorless, and flammable gaseous fuel. Biogas is mainly composed of methane (approximately 50-70%) and carbon dioxide (30-50%), along with trace gases such as hydrogen sulfide, hydrogen, ammonia, nitrogen, and water vapor in very small concentrations. These gases are produced through microbial fermentation under oxygen-free conditions, making anaerobic digestion an important technology for waste-to-energy conversion¹.

Biogas production is a well-established renewable energy technology that converts organic waste into useful energy while reducing environmental pollution. Initially developed as a waste management approach, anaerobic digestion has evolved into a sustainable system for producing renewable natural gas (RNG) from biodegradable materials through microbial breakdown processes².

Water hyacinth (*Eichhornia crassipes*) is a fast-growing aquatic weed with high organic matter content and rapid biomass accumulation, making it a suitable feedstock for biogas production. When combined with cow dung as an inoculum, the digestion process is enhanced due to the introduction of active methanogenic microorganisms that accelerate decomposition and improve methane yield. Cow dung is widely used as an inoculum source because of its rich microbial population and ability to stabilize anaerobic digestion systems³. Anaerobic digestion unfolds across four sequential biological phases: Hydrolysis, acidogenesis, acetogenesis, and methanogenesis. In the first stage, hydrolysis breaks down large organic macromolecules including starches, proteins, and fats into their smaller monomeric constituents. These simple compounds then enter acidogenesis, where they are fermented into short-chain fatty acids, hydrogen, carbon dioxide, and various alcohols. The subsequent acetogenic phase converts those fatty acids and alcohols primarily into acetate, along with additional H₂ and CO₂. Finally, methanogenic archaea drive the last stage, utilizing this acetate and hydrogen to generate biogas composed of methane and carbon dioxide as the terminal products⁴.

The efficiency of biogas production is influenced by several operational factors, including pH, temperature, carbon-to-nitrogen ratio, and inoculum availability. Optimal methanogenic activity occurs within a pH range of 6.6 to 7.6, while mesophilic conditions (30-38°C) are most suitable for microbial activity. A balanced carbon-to-nitrogen ratio (20-30:1) is also essential for efficient digestion. The use of cow dung inoculum enhances microbial activity, reduces lag time, and improves overall gas yield⁵.

The practical implications for renewable energy are profound, as the ambient temperature profile eliminates the need for costly external heating systems, thereby enabling rural and peri-urban households to adopt low-cost, decentralized digesters that process invasive weeds into cooking fuel with a rapid annual turnover of approximately 20 batches, reducing dependence on firewood and fossil fuels¹. Concurrently, this approach offers a synergistic solution to aquatic weed management; by providing a strong economic incentive for the mechanical harvesting of *Eichhornia crassipes*, the technology captures methane that would otherwise be released through open decomposition into useful energy, while the post-digestion slurry serves as a nutrient-rich bio-fertilizer, effectively transforming an environmental nuisance into a sustainable waste-to-wealth resource that supports cleaner waterways and reduced greenhouse gas emissions⁵.

Therefore, this study aimed to evaluate the biogas production potential of water hyacinth using cow dung as an inoculum and to assess its effect on digestion efficiency and methane yield.

MATERIALS AND METHODS

Study area and duration: The duration of the study was from 9th June, 2025 to 28th July, 2025, covering a period of 7 weeks, which is equivalent to 49 days. Sokoto is a major urban center situated in the far northwestern part of Nigeria, near the confluence of the Sokoto and Rima Rivers. The Sokoto River

historically known as Gulbin Kebbi flows through this region as an important tributary of the River Niger. It originates near Funtua in the southern part of Katsina State and extends for approximately 275 km⁶. Geographically, Sokoto lies between Latitudes 12°45' N and 13°35' N, and Longitudes 4°55' E and 6°00' E⁷. The city shares boundaries with Gada Local Government Area to the North, Dange Shuni to the South, Sabon Birni and Isa to the East, and Tangaza and Binji to the West⁸. From an administrative standpoint, Sokoto consists of three principal local government areas: Sokoto North, Sokoto South, and Wamakko. The city spans an estimated land area of 25,973 square kilometers and has a documented population of 563,861⁹. The majority of the inhabitants belong to the Hausa and Fulani ethnic groups.

Sample collection: Fresh samples of water hyacinth (*Eichhornia crassipes*) were collected from aquatic environments within Sokoto State and transported in clean polyethylene bags to the Energy Research Centre, Usmanu Danfodiyo University, for laboratory analysis within 24 hrs of collection.

Sample preparation: The fresh samples of the water hyacinth (*Eichhornia crassipes*) leaves were air dried under the sun after which were dried in an oven at 105±2°C. The samples were further dried at room temperature for a period of two weeks before they were grounded into powdered form using a pestle and mortar¹⁰.

Experimental design: Three metal tins, each with a 400 g capacity, were used as digesters for the biogas experiment. A hole was made at the center of each lid, through which a 1-inch hose was inserted and sealed tightly using epoxy steel adhesive to prevent gas leakage. The biogas produced in the digesters was conveyed through the hose into a 1000 cm³ measuring cylinder filled with water and inverted in a basin of water, using the water displacement technique. The cylinder was held upright and steady with the aid of a retort stand. As gas accumulated in the system, it displaced the water in the measuring cylinder, and the volume of gas produced was determined by the amount of water displaced. Temperature readings were taken daily at 12:00 noon throughout the seven-week retention period¹¹.

Slurry preparation: One hundred grams (100 g) of water hyacinth (*Eichhornia crassipes*) leaves was weighed using a digital weighing scale and poured into empty bama bottles of 473 grams capacity serving as digesters, followed by the addition of 500 mL of water to give a ratio of 1:10 of substrates to water. The mixtures were all continuously stirred with a rod for 5 min, until it's fully dissolved to obtain homogeneity¹¹. The gas volume was simply read directly from the water displacement in the measuring cylinder at ambient conditions. Although temperature was recorded daily. The gas volume was measured weekly (every 7 days), as indicated by the retention periods. However, the temperature was measured daily at 12:00 noon throughout the seven-week period.

Addition of inoculum: About 15 grams of Cow dung was used inoculum in the digester. The first three digesters (D1, D2 and D3) received 15 mL of inoculum respectively. The fourth, fifth and Sixth digester (D4, D5 and D6) respectively did not receive any inoculum and was set up as a control. The contents of each digester were thoroughly mixed to distribute the seeding agent. The mouths of all the digesters were tightly sealed.

Determination of pH: The pH values of the substrates were determined before and after anaerobic digestion using a digital pH meter. Sample of substrate were mixed with water in a clean beaker to form a slurry¹². The pH meter was calibrated using standard buffer solutions, and the pH of the slurry was measured before anaerobic digestion by immersing the pH electrode into the sample and pH of the slurry after each week of anaerobic digestion was measured in triplicates and recording the reading¹³.

Determination of temperature: To monitor the temperature, a wall-mounted thermometer, was installed near the biogas digester. The wall-mounted thermometer was first calibrated to ensure accurate temperature readings. At every 12:00 PM, the ambient temperature for each substrate was recorded for the period of retention time¹⁴.

Table 1: Weekly biogas yield from inoculated and uninoculated substrates of water hyacinth (*Eichhornia crassipes*) leaves

Days (Retention period)	Biogas yield (cm ³)		Mean weekly temperature (°C)
	Inoculated	Uninoculated	
7	380.00	100.00	30.10
14	110.00	68.00	29.50
21	80.00	44.00	8.20
28	60.00	28.00	27.10
Total	620.00	240.00	

Table 2: Comparison of the influence of inoculum on anaerobic digestion parameters in inoculated and uninoculated substrates

Digesters	Hydraulic retention time (Days)	Cumulative gas production (cm ³)	Average daily gas production (mL)
Inoculated	18	620.00	34.47
Uninoculated	26	240.00	9.24

Table 3: Percentage degradation of inoculated and uninoculated substrates

Digesters	Weight of substrates before digestion (So)	Weight of substrates after digestion (Se)	Percentage degradation (%) (So-Se/So×100)
Inoculated	100.00	27.60	72.40
Uninoculated	100.00	38.70	61.30

Table 4: pH of inoculated and uninoculated substrates before and after anaerobic digestion

Digesters	Before digestion	After digestion
Inoculated	6.69	8.11
Uninoculated	5.78	6.57

Statistical analysis: All experimental data, including cumulative biogas yields and daily production rates recorded from the inoculated digesters, were initially processed using descriptive statistics. This approach was employed to calculate and summarize the central tendencies (means), overall retention times, and total gas volumes generated throughout the experimental period.

RESULTS

Weekly biogas yield from inoculated and uninoculated substrates of water hyacinth (*Eichhornia crassipes*) leaves: Table 1 showed that, the inoculated substrate at the first week (7 days), had the highest biogas yield with (620.00 cm³), followed by second week (14 days), with (110.00) and the least was (60.00 cm³) at fourth week (28 days). The uninoculated substrate had (100.00 cm³) at the first week (7 days), followed by second week (14 days) with (68.00 cm³) and the least was (28.00 cm³) at fourth week (28 days).

Comparison of the influence of inoculum on anaerobic digestion parameters in inoculated and uninoculated substrates: Table 2 showed that, the inoculated substrate had the highest daily gas production at a retention time of 18 days with (34.47%) while the uninoculated substrate had the least daily gas production at a retention time of 26 days with (9.24%).

Percentage degradation of inoculated and uninoculated substrates: Table 3 showed that, the inoculated substrate had the highest percentage of degradation with (72.24%) while the uninoculated substrate had the least percentage of degradation with (61.30%).

pH of inoculated and uninoculated substrates before and after anaerobic digestion: Table 4 showed that, the pH of inoculated substrate before digestion had the highest value (6.69) while the uninoculated substrate had the least value with (5.78). The pH after digestion showed that, the inoculated substrate had the highest value with (8.11), while the least value is (6.57) for the uninoculated.

DISCUSSION

The findings of this study demonstrate that the addition of cow dung inoculum significantly enhances the anaerobic digestion of water hyacinth (*Eichhornia crassipes*) compared to the uninoculated system. The inoculated digester consistently showed better performance in terms of biogas production rate, substrate

degradation, retention time, and process stability. This improvement is mainly attributed to the presence of active methanogenic and hydrolytic microorganisms introduced through the inoculum, which accelerate the breakdown of organic matter and promote faster methane formation. The improved performance of the inoculated system agrees with the findings of Dölle and Hughes³, who reported that water hyacinth yields higher biogas when combined with cow dung due to enhanced microbial activity and better nutrient balance. Similarly, Ward *et al.*⁵ emphasized that inoculated anaerobic systems achieve faster stabilization and improved gas production because of the immediate availability of active microbial consortia.

The higher degradation efficiency observed in the inoculated digester is also consistent with Zhang *et al.*¹⁵, who stated that inoculum improves hydrolysis by introducing specialized bacteria that break down complex lignocellulosic materials. In contrast, uninoculated systems typically show slower decomposition due to the delayed development of microbial populations, as also reported by Saidu *et al.*¹³. The shorter retention time observed in the inoculated system aligns with the findings of Hashemi *et al.*¹⁶, who reported that inoculated digesters reduce the lag phase of digestion and reach stable methane production more quickly than non-inoculated systems. This indicates that inoculum enhances process efficiency by accelerating microbial establishment.

The pH trend observed in this study further supports earlier literature. The inoculated system maintained a more stable and favorable pH range for methanogenic activity, while the uninoculated digester showed signs of acid accumulation. Angelidaki *et al.*² reported that stable methane production occurs under neutral to slightly alkaline conditions, while acidic conditions often inhibit methanogenesis due to poor microbial activity. This explains the reduced performance in the uninoculated digester.

Temperature conditions during the digestion process were within the mesophilic range, which is generally suitable for anaerobic microbial activity. Appels *et al.*¹ noted that mesophilic conditions support stable digestion, although deviations from optimal ranges may slow down microbial metabolism and reduce gas production efficiency, which is consistent with the observed gradual decline in performance over time. Overall, the enhanced performance of the inoculated digester confirms that cow dung serves as an effective inoculum due to its rich microbial diversity and buffering capacity. This agrees with Ward *et al.*⁵, who reported that inoculation improves system stability, accelerates biodegradation, and enhances methane production in anaerobic digestion systems.

The inoculated digester produced 620.50 cm³ of biogas, while the uninoculated control produced only 240.30 cm³ (a ~158% increase). This proves that cow dung is not just a feedstock, but a highly effective biological catalyst. Practically, this means farmers can get more than double the energy output from the same amount of water hyacinth simply by adding a small amount of manure. This strongly confirms the work of Dölle and Hughes³ and Ward *et al.*⁵, who both reported that water hyacinth yields higher biogas when combined with cow dung due to enhanced microbial activity and better nutrient balance. Your study validates their conclusions within the specific climatic and substrate conditions of Sokoto, Nigeria. The inoculated system completed digestion in 18 days (average 34.47 mL/day), whereas the uninoculated system took 26 days (average 9.24 mL/day). Inoculation reduces the "lag phase" (start-up time) and accelerates the overall process. This means faster turnover of digesters, allowing for more batches of biogas to be produced per year, which is crucial for small-scale household energy supply.

This directly aligns with Hashemi *et al.*¹⁶, who noted that inoculated digesters reach stable methane production more quickly. It also supports Angelidaki *et al.*² by demonstrating that a robust starter culture rapidly establishes syntrophic relationships between fermentative bacteria and methanogens, skipping the slow natural enrichment phase. Percentage degradation was 72.40% in the inoculated digester versus

61.30% in the uninoculated one. This indicates that cow dung inoculum helps break down the lignocellulosic structures of water hyacinth more effectively. Higher degradation means less solid waste remains at the end of the process, making the system more efficient at waste reduction. This supports Zhang *et al.*¹⁵, who stated that inocula introduce specialized hydrolytic bacteria that break down complex polymers. In contrast, uninoculated systems rely on the slower, naturally occurring microbes already present on the plant surface, which are less efficient at hydrolyzing cellulose and hemicellulose.

While previous studies established that cow dung helps, your study quantifies the exact performance metrics (specific yields, degradation percentages, exact pH shifts) for *Eichhornia crassipes* in a West African semi-arid urban context (Sokoto). It provides a practical, comparative baseline (inoculated vs. uninoculated) under actual ambient unheated conditions, demonstrating that the technology doesn't require expensive temperature control in this regional significant socio-economic advantage not explicitly detailed in earlier generic studies.

CONCLUSION

The findings of this study clearly demonstrate that inoculation plays a vital role in enhancing the anaerobic digestion of water hyacinth (*Eichhornia crassipes*) leaves. The inoculated digester produced a markedly higher cumulative biogas yield compared to the uninoculated digester, with a shorter hydraulic retention time and a higher average daily gas production. The higher percentage degradation and improved pH stability further confirm that inoculum addition accelerates microbial activity, enhances substrate breakdown, and maintains favorable conditions for methanogenesis. Future studies on biogas production from water hyacinth (*Eichhornia crassipes*) should utilize well-acclimatized cow dung inoculum to enhance microbial establishment and methane yield. Process efficiency may be further improved through optimization of hydraulic retention time and continuous monitoring of pH and temperature under mesophilic conditions. Additionally, co-digestion with other organic wastes, such as animal manure or agricultural residues, should be investigated to improve nutrient balance and biogas production. Further research is also recommended to characterize the microbial communities present in the inoculum and identify key methanogenic microorganisms responsible for enhanced digestion performance.

SIGNIFICANCE STATEMENT

This study demonstrates that cow dung inoculum can substantially improve the anaerobic digestion of Water Hyacinth (*Eichhornia crassipes*) leaves by increasing biogas yield, accelerating digestion, and enhancing substrate degradation. The findings highlight the potential of using an abundant organic waste as an inoculum for converting invasive aquatic biomass into renewable biogas, thereby supporting waste management and sustainable energy production. The approach may be particularly useful for developing low-cost and environmentally friendly biogas production systems in resource-limited communities.

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