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PROPORTIONAL EVALUATION OF WATER HYACINTH AS A RAW MATERIAL FOR BIOGAS, BIOMANURE AND SOIL CONDITIONER GENERATION VIA ANAEROBIC PROCESSES AT LAB AND PILOT SCALE

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ABSTRACT
Water hyacinth is capable of inexplicably concealing an entire water body within two weeks by forming thick mats due to its rapid proliferation intimidating the existence of aquatic organisms. Therefore, this study is aimed to investigate the conversion of WH to biogas, biomanure and soil conditioner (SC) through anaerobic digestion (AD) and composting process. While composting was studied at the lab and pilot scale using leaves and roots, the AD of WH at various food to inoculum (F/I) ratios of 1, 2, and 5 was carried out with leaves, roots, and a mixture of leaves and roots. The study found that leaves can efficiently produce biogas at a 2:1 F/I ratio with 350 mL/g VS reduced methane and 63% CH_4. The qualitative evaluation of biomanure and SC from AD and composting processes revealed that the SC produced from leaves is suitable for agriculture farming (Fenugreek and CoriandrumSativum) while that of roots for horticulture (Hibiscus and Papaya). About 1200 tons of WH was converted to 120 tons of soil conditioner to control the WH growth in Kapra Lake in Hyderabad city.

1. INTRODUCTION

Water hyacinth (WH), also known as Eichhornia Crassipes, is a promising feedstock for anaerobic digestion due to its high proliferation rate and ability to withstand harsh environmental conditions (Patil et al., 2014; Rezania et al., 2015). The rapid reproduction of WH forms a floating cover on water surfaces, obstructing light and oxygen, leading to aquatic fauna death and significant impacts on sustainability, economic growth, biodiversity, and human health. The methods of control used to curb the extensive growth of WH are found either to be not sustainable (chemical), not cost-effective (mechanical), or may require a longer time to degrade and stabilize (biological) (Barua and Kalamdhad, 2017; Bhui et al., 2018; Li et al., 2021; Mathew et al., 2015). However, the use of either of these methods is inevitable because the WH growing in water bodies if left unattended would lead to the extinction of aquatic life.

WH can be utilized efficiently for phytoremediation, compost production, enzyme extraction and bioenergy generation via biological pathways (Barua and Kalamdhad, 2017; Ilo et al., 2020). In India, most of the domestic wastewater is released into the water bodies resulting in the accumulation of micro and macro nutrients along with heavy metals leading to eutrophication. Moreover, heavy metals do not decompose throughout the composting process since they are inert components. Therefore, repeated long-term applications of manures and compost may cause heavy metal components to accumulate in agricultural soil to dangerous levels, which may have an impact on crop yield and health (Rashid et al., 2023). The corresponding detection limits for lead, zinc, cobalt, copper, nickel, and cadmium were 0.4 μg L^{-1}, 0.012, 0.2, 0.17, 0.11, and 0.2 μg L^{-1}(Smirnova et al., 2021). Heavy metals such as phosphorus and nitrogen in water cause algal overgrowth, causing fish and aquatic species to die due to reduced oxygen levels caused by algae death and decomposition.

However, heavy metal accumulation in roots may hinder biological processes (Sanmuga Priya and Senthamil Selvan, 2017). Biological stabilization of waste material is found to be efficient with substrates having high moisture content. WH is an aquatic weed which has more than 90% of moisture which by itself declares the suitable of the substrate for biological valorization (Islam et al., 2021). Further,

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the WH consists of abundant organic matter in the form of volatile organic and chemical oxygen demand which can be efficiently utilized for the generation of bioenergy or its conversion to biofertilizer. Although the WH contains biodegradable organic matter, it contains lignocellulosic material, heavy metals which may substantially hinder the anaerobic digestion process for biogas generation (Koyama et al., 2015; Pellera and Gidarakos, 2018).

Nevertheless, optimizing the critical operational parameters may facilitate efficient biogas production from WH despite its inherent disadvantages. One of the critical parameters in AD is the feed to inoculum (F/I) ratio, that is defined as the ratio between VS of feed and VS of inoculum used. Determination of ideal F/I ratio is necessary to prevent process imbalance due to depletion of food or stressed organic loading resulting in stocked up intermediate products that hinder the activity of microorganisms (Castro and Agblevor, 2020). A study reported by (Nugraha et al., 2018) illustrates that Food to Microorganism (F/M) ratio of 0.2 resulted in the maximum methane of 164.7 g/g COD, while the substrates such as food waste under different operating conditions results in the generation of broad range of cumulative biogas yields between 300 – 600 mL/g VS (Begum et al., 2016; Kuruti et al., 2017). The low amount of biogas produced by WH (53-58% methane) and 45.18% methane can be ascribed to the presence of lignocellulosic complex, heavy metals and bacteria which impede the microorganisms' ability to produce biogas. The available literature on the AD or composting of WH is demonstrated with the complete plant but the novelty of this study lies in the AD of WH with leaves, roots, and a combination of both (Barua and Kalamdhad, 2017; Unpaprom et al., 2021).

Further, in the recent past, application of organic manure has shown a significant interest and impact on farming in comparison to chemical fertilizers and the rising demand of health-conscious food consumers, preferring food which is cultivated with bio manure application rather than chemical fertilizers (Vidya and Girish, 2014). The digestate or the compost derived from AD and composting can be used as biomanure/biofertilizer. An added benefit of biomanure is that it encourages the growth of nitrogen-fixing bacteria, whereas artificial fertilisers inhibit this growth due to their high acid content. Accelerated Anaerobic Composting (AAC) specified as the anaerobic composting process that yields finished compost in less time with the addition of cow dung and Anaerobic mixed microbial consortia (AMMC) in large piles for adequate treatment of WH. Furthermore, it is interesting to note that WH can sequester large amounts of CO_2 from the environment which creates a positive impact in the reduction of GHGs and stabilizing the WH in anaerobic environment would curb the release of GHGs.WH is rich in organic matter, making it a suitable feedstock for anaerobic digestion and generates methane, which can be used as a renewable energy source for electricity generation or as a fuel. The implementation of biogas plant or a composting unit for the valorization of WH depends on the utilization of product at the source of waste treatment facility. Besides, WH plays a crucial role in absorbing carbon dioxide (CO_2) from the atmosphere and storing carbon within its biomass. This weed acts as a carbon sink, aiding in the mitigation of climate change by reducing atmospheric CO_2 levels and consequently decreasing global warming. For instance,in Assam, India, the density of WH was 10.37 plants/m^2 during the pre-monsoon season, which increased to 21.56 plants/m^2 in the monsoon and post-monsoon seasons. However, it decreased to 25.91 plants/m^2 in winter (Lahon et al., 2023). Reports on tropical lakes suggest that the rapid growth of water hyacinth results in a substantial intake of carbon dioxide (CO_2) at rates ranging from 3.4 to 5.4 g C-CO_2 m^{-2} day^{-1}(Junior et al., n.d.).

This study is aimed to valorize WH in two ways via anaerobic processes. In the first method, AD of WH has been investigated to study the impact of substrate composition (roots, leaves and mixture of roots and leaves) and F/I ratio on biogas and biomanure generation potential. The performance of AD is studied in terms of biogas yield, methane (%), VS reduction (%), volatile fatty acids and pH profile. In the second approach, the WH is converted to soil conditioners with leaves and roots separately via AAC process at lab as well as pilot scale. As the digestate obtained from AD process can also be utilized as fertilizer, this study has made a comparative quantitative and qualitative analysis of biomanure, and soil conditioner generated via AD and ACC process respectively for leaves, roots and combination of both. The performance of composting process is assessed based on the response of plants growth (Fenugreek and Coriandrum Sativum). The pilot scale demonstrated for the cleaning of Kapra lake is the first of its kind project in the country where 1200 tons of WH is removed to produce soil conditioner.

2. MATERIALS AND METHODS

2.1 Water hyacinth collection and sample preparation for laboratory scale experiments

About 50 kg of freshwater hyacinth weed was pulled out from Kapra Lake in Hyderabad which was washed to remove mud tangled with WH roots and leaves. The WH was then cut into three parts namely leaves, stalks and roots as shown in Figure 1. The chopped leaves, roots and stalks were coarsely ground into a paste separately with particles of approximately 0.5-1 cm. In addition to both these substrates, a third substrate was prepared with both the pastes mixed in 1:1 ratio. All the three substrates were analysed for total and volatile solids (TS and VS) and stored at 4°C for further experimental studies on anaerobic digestion (AD). The same substrate is used for AAC with a composition of 30% inoculum (AMMC + Cow dung) and 70% of substrate.

2.2 Inoculum

Anaerobic mixed microbial consortia (AMMC) was collected from the inhouse biogas plant facility operated with food and vegetable waste generated in the departmental canteen. The AMMC was used as seed inoculum for AD experiments while a mixture of cow dung and AMMC in the ratio of 1:1 was used for composting experiments. About 50 L of AMMC was collected from an existing biogas plant operated at mesophilic temperature with cooked

WH (Leaves and Stalks) Chopped Leaves and Stalk Chopped Roots

FIGURE 1: The whole WH and the chopped leaves, stalks and roots.

and uncooked (vegetable waste and peels) food waste as feedstocks in Hyderabad while the cow dung was procured from the nearby farm. Both the AMMC and cow dung were analysed for TS and VS to design the experiments.

2.3 Characteristics of substrate and inoculum

The characteristics of substrates, AMMC as well as cow dung in terms of pH, TS and VS is as follows. The pH of Leaves (L), Roots (R), L + R in 1:1 ratio, AMMC and Cow dung are 6.8, 8.4, 6.9, 7.0 and 7.1 respectively. The TS (%) of L, R, L + R, AMMC and Cow dung are 6.6, 8.3, 6.9, 3.8 and 11 respectively. Similarly the VS (%) of L, R, L + R, AMMC and Cow dung are 6.4, 6.9, 6.2, 2.7 and 7 respectively. The VS of substrate and inoculum was used to design the experiments.

2.4 Experimental setup and operational strategy

The potential of WH as an alternative raw material for biogas, biomanure and soil conditioner generation was assessed via two anaerobic processes namely, anaerobic digestion (AD)/biomethanation and composting.

2.4.1 Configuration of anaerobic digestion experiments based on food to inoculum (F/I) ratio

A set of nine anaerobic batch reactors of 500 mL with an effective volume less than 380 mL with required arrangements for feed inlet, gas collection and sampling were operated in parallel along with their duplicates for 30 days. The substrate composition in reactors R1, R2 and R3 was Leaves, Roots and mixture of leaves and roots in 1:1 ratio which remained same in all the reactors. The detailed composition of reactors designed based on the VS content in substrate and AMMC is summarized in Table 1. As can be seen from Table 1, that the reactor R1-L, R2-R and R3-L+R were designed with 30% AMMC and 70% substrate of the effective volume which was found to be operated at an F/I ratio of ~5 while the other reactors were designed to achieve a desired F/I ratio of 1:1 and 2:1 respectively. In addition to these reactors, a set of two blank reactors only with inoculum were set to determine the biogas generation potential of the inoculum. The biogas generated from the inoculum was deducted from the biogas produced from the other reactors to determine the biogas generation potential of the substrate. All the reactors were operated at mesophilic temperature (37 ± 2°C) placed in a water bath shaker at an rpm of 100. The pH of the reactors was not controlled as the substrate as well as the inoculum was at neutral or slightly alkaline pH. Reactor samples were withdrawn on 10th day to check the variation in performance indicators such as VFA and pH and compared to that of initial and final day samples. There is a limit to the ability of bacteria to feed on substrate, or the substrate has entirely broken down and no gas is being produced. Thus, in order to produce gas, more bacteria or substrate must be supplied. The biogas generated was measured every day and analysed for its composition through GC.

2.4.2 Composting of Water hyacinth

Two anaerobic composting reactors (ACR) of 50 L volume were used to perform composting experiments with leaves and stalks (ACR1) and roots (ACR2) as shown in Figure 2. The effective volume of reactors was 35 L which

TABLE 1: Composition of reactor contents based on definite inoculum volume and F/I ratio.

Reactor label	Substrate (g)	AMMC (mL)	Equivalent $gVS_{substrate}$	Equivalent gVS_{AMMC}	F/I
R1 - L	245	105	15.7	2.8	5.60
R2 - R	245	105	16.9	2.8	6.03
R3 - L+R	245	105	15.2	2.8	5.42
R1 (1:1)	110	240	7.04	6.5	1.08
R2 (1:1)	110	260	7.59	7.1	1.07
R3 (1:1)	110	240	6.82	6.5	1.05
R1 (2:1)	155	180	9.92	5.0	2.04
R2 (2:1)	160	200	11.04	5.4	2.04
R3 (2:1)	170	190	10.54	5.1	2.05

were fed with 30% inoculum (AMMC + Cow dung) and 70% of substrate. Each reactor was fed with 24.5 kg of respective substrate, 5.25 L of AMMC and 5.25 kg of cow dung. Further, the reactors were supplemented with micro and macronutrients solutions of 10 mL each to ensure better activity of the microorganisms. The reactors were operated at ambient temperature and ensure proper mixing of inoculum and substrate, then tightly sealed with the lid to ensure perfect anaerobic environment. The possibility of biogas generation in these reactors is less due to ambient temperature conditions. This is because the process of AD for biogas generation occurs efficiently either at mesophilic or thermophilic temperature but not psychrophilic. The product after 40 days of composting has been spread and sun dried and the dried material referred to as soil conditioner was analysed for N, P_2O_5 and K_2O as mentioned earlier.

2.5 Analytical methodology

The three substrates (leaves + stalks, roots and mixture of leaves + stalks + roots) along with AMMC and cow dung were initially characterized individually for pH, TS, VS, moisture content (MC) and chemical oxygen demand (COD) as per the standard chemical procedures (American Public Health Association. Andrew D., American Water Works Association., Water Environment Federation., 2005). Further the substrates were also analyzed for carbon (C), nitrogen (N) and sulphur (S) using CHNS analyser (Vario micro cube elementar, Germany), heavy metals, Phosphates (P_2O_5), potash (K_2O) using inductively coupled plasma - optical emission spectrometry (Thermo scientific, model: iCAP6500DUO). The reactor samples from AD and composting experiments were analyzed for pH and VFA on initial day, 10th and 30th day. VFAs were analyzed using High Performance Liquid Chromatography (HPLC) (LC- 10 AT, VP, Shimadzu, Japan) equipped with ultra-violet detector (C18 chromasil column was used under mobile phase conditions using 0.001 N, 90% H_2SO_4 in aceto nitrile). The flow rate and the temperature of the column were maintained at 0.6 mL/min and 30°C respectively. The volume and composition of biogas was analyzed using wet gas flow meter

FIGURE 2: Composting of WH leaves & stalks and roots in 50 L containers.

(ISI 068; INSCIN private ltd) and Gas Chromatography (GC) respectively which was equipped with TCD detector operated at 250°C at a flowrate of 1 L/min and a plot capillary column at 38°C, injector temperature at 200°C (TCD, GC 2010, shimadzu, Japan).

2.6 Evaluation of Kinetic potential parameters of AD of WH

The modified Gompertz model is an empirical non-linear regression model that is used to analyze the lag phase (λ) in days of AD of WH as shown in equation (1) (Begum et al., 2020). All the data obtained from AD experiments were analyzed using IBM SPSS software version 24.0.

$$Y = M * Exp\ \{-Exp\ [((Rm * 2.718)/M) * (\lambda - t) + 1]\} \qquad (1)$$

where:
Y= Cumulative biogas generation at time't' (L)
λ = Lag phase (day)
M = Highest biogas generation potential (mL)
Rm = Maximum biogas generation rate (mL/day)

3. RESULTS AND DISCUSSION

3.1 Physio-chemical characterization of WH

The extent of biological conversion of any substrate to value-added products is determined based on the presence of quantity of volatile organic matter and the recalcitrant substances. Therefore, the physico-chemical characteristics of WH were analyzed for roots and leaves separately and the average values are presented in Table 2. The BOD/COD, C/N and the V/S ratio of WH leaves illustrate that the leaves and stalk portion are biodegradable and suitable for AD process as the required C/N ratio for AD falls between 20 and 30 (Barua and Kalamdhad, 2019). The pH, C/N and the BOD/COD ratio for roots of WH seem not favourable for AD process which means that roots have to be co-digested with other substrates for better outputs. Further, the concentration of heavy metals like Hg, Pb, As_2O_3, Cr, Cu and Cd are higher in roots that hinders AD compared to leaves. According to World Health Organisation guidelines, the allowable limits of some heavy metal ions in drinking water are as follows: chromium: 5.69 ppm, copper: 11.31 ppm, zinc: 22.8 ppm, etc. Besides, the cumulative generation of biogas and methane volumetric concentration in an AD using olive mill waste as the substrate were 64.6% and 0.426 Nm^3/kg VS, respectively. While the concentration of CH_4 was higher, the biogas generation with the addition of nickel dropped to 0.083 Nm^3/kg VS as compared to no addition at all. As a result of the addition of copper and chromium, the overall production of biogas dropped by 0.233 Nm^3/kgVS (Alrawashdeh, K.A.b.; Gul, E.; Yang, Q.; Yang, H.; Bartocci, P.; Fantozzi, 2020). Therefore, the leaves and stalk portion of WH plant are more amenable for AD or composting than roots. Zinc and Copper were the highest recorded metals in roots at 22 and 12 ppm of dry weight, respectively. In accordance with literature, it was observed that WH can be used as indicator for heavy metal water contamination. Since this plant grows extensively on water bodies, it can be utilized for phytoremediation due to its high metal uptake capacity and later can be converted to biogas com-

TABLE 2: Physico-chemical characteristics of WH leaves and roots.

Parameter	WH leaves	WH roots
C (%)	39.26	38.27
H (%)	5.72	5.46
N (%)	1.5	2.0
S (%)	0.32	1.27
C/N	26	19
TS (%)	6.6	8.3
VS (%)	6.4	6.9
VS/TS	0.96	0.83
COD (mg/L)	56,000	48,000
BOD (mg/L)	32,000	21,200
BOD/COD	0.57	0.43
pH	7.1	8.89
Arsenic (As_2O_3) (mg/kg)	BD*	2.87
Cadmium (Cd) (mg/kg)	BD*	0.19
Chromium (Cr) (mg/kg)	3.69	5.69
Copper (Cu) (mg/kg)	1.64	11.31
Mercury (Hg) (mg/kg)	BD*	0.47
Nickel (Ni) (mg/kg)	0.12	1.54
Lead (Pb) (mg/kg)	0.54	2.94
Zinc (Zn) (mg/kg)	8.93	22.8

BD Below detection limit*

bined heat and power applications and biomanure and soil conditioner for soil nourishment. Therefore, bioconversion of WH via AD and composting process can be regarded as a means of eradication by re-utilization.

3.2 Effect of substrate composition and F/I ratio on the biogas generation profile and yield

The daily biogas generation profile and the cumulative biogas yield obtained from AD of WH with different substrate composition and varied F/I ratio is graphically presented in Figure 3. The gas production of inoculum was measured and subtracted from the gas production of feedstock and the final amount of gas was included in the data. As can be seen from Figure 3 (a) that the daily biogas generation was < 100 mL/day in reactors R2 operated with roots which is relatively lower than R1 and R3 which were operated with leaves and mixture of leaves and roots of WH respectively which could be attributed to the presence of higher concentration of heavy metals like Pb, Cu, Hg, Cr and As_2O_3 in roots. Presence of other metals such as Zn and Ni might have favoured biogas generation but couldn't eliminate the inhibitory effects of other heavy metals that proven to be toxic to micro-organisms (Guo et al., 2019). The critical factors that affect the performance of AD are pH, organic loading rate (OLR), hydraulicretention time (HRT), temperature, solids consistency, food to inoculum and carbon to nitrogenratio, availability of nutrients, microbial community, presence of inhibitors, etc. If the maximum daily biogas production from reactors R1 is compared, the biogas generation sharply increased from 60-140 mL/day from Day 2 to 12 at an F/I ratio of 2:1 while it took longer time in R1-L (day 14-17) which was operated with 30% inoculum and 70% substrate i.e., F/I ratio of 5 to produce maximum biogas of 150 mL/day. This indicates that the balance of food and micro-organism number is essential to prevent organic loading shock and facilitate biogas production from even hardly biodegradable substrates. As the biogas generation process is a biological process dependant on the activity of biomass, the maintenance of optimal F/I ratio in the reactor is important to ensure availability of the active biomass to the amount of food, balance in their nutrient availability, maintenance of pH etc., are some of the critical performance indicators resulting in efficient biogas production. In the present study, at an F/I ratio of 2:1, the biogas generation increased from day 2 to 12 indicating the optimal F/I and C/N ratio. In contrast to that, the reactor R1-L with only leaves exhibited lagphase as the reactor did not had adequate nitrogen to maintain optimal C/N ratio. Therefore, the study concludes that the optimal F/I ratio varies depending on the specific characteristics of the feedstock and the microbial community. Generally, a balanced F/I ratio is crucial for efficient biogas production. Too high of a substrate concentration (high F/I ratio) may lead to process inhibition, while too low of a substrate concentration may result in underutilization of the microbial capacity.

Similarly, from Figure 3 (b) it can be observed that the maximum cumulative biogas yield that can be achieved from roots of WH (R2) is between 120 to 160 mL/g VSreduced, while it is between 220 – 352 mL/ g VS reduced from leaves (R1) and 189 to 301 mL/ g VSreduced from mixture of leaves and roots (R3). The maintenance of F/I ratio played a role in biogas generation which is evident from the maximum yield obtained at F/I ratio of 2:1 than 1:1 or 5:1. A study reported by (Raposo et al., 2009) demonstrated the increase in F/I ratio may lead to the accumulation of VFA during the process. Accumulation of VFA in higher F/I ratio reveals the kinetic uncoupling between acid producing and acid consuming microorganisms. It was reported that the F/I ratio of 1:2 was found to be ideal for substrates like food waste to prevent accumulation of acids, but in the present study, the higher F/I ratio of 5 is favoured but at 2, the biogas generation was better compared to 1 and 5 (Begum et al., 2021). The maintenance of F/I ratio and proportional mixing of roots and leaves have favoured the process because the maximum biogas generation was observed in reactors R1 and R3. This indicates that the biomethanation of roots alone is not a feasible option but could be made suitable for biogas generation when the carbon rich stalk and leaves are co-digested.

3.3 Performance evaluation of anaerobic digestion of water hyacinth: Impact of F/I ratio and substrate composition on methane yield, methane concentration and volatile solids reduction

The biogas generation potential from WH investigated at different F/I ratios and substrate composition is illustrated in terms of VS reduction (%), methane yield and concentration as shown in Figure 4. As can be seen from Fig-

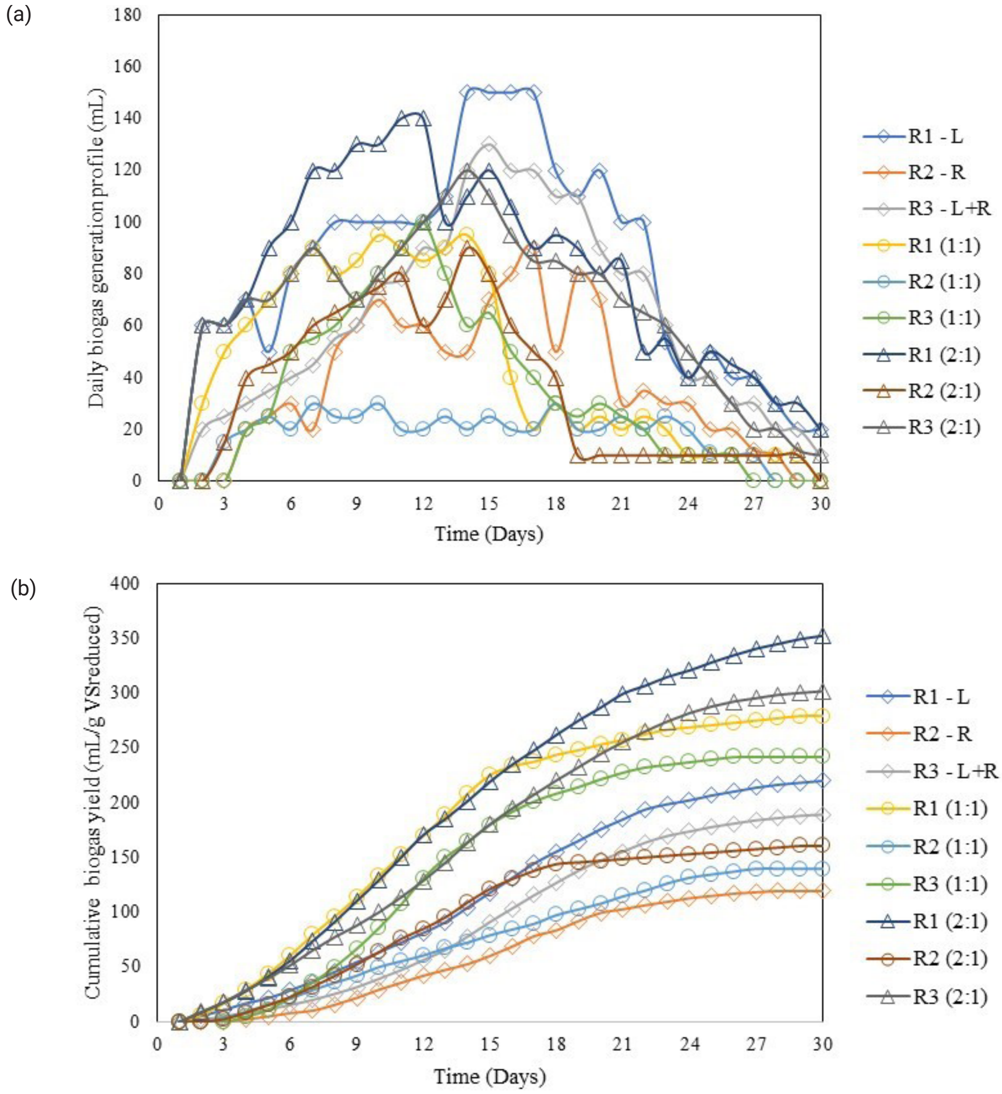

FIGURE 3: Influence of substrate composition and F/I ratio on the biogas generation (a) Daily biogas generation profile, (b) Cumulative biogas yield.

ure 4 that the maximum biogas yield that can be achieved is 350 mL/g VSreduced from leaves of WH at an F/I ratio of 2:1 followed by a combination of roots and leaves i.e., 300 mL/g VSreduced in R3 with a corresponding CH_4 concentration of 63% and 64% and VS reduction of 69% and 63% respectively. According to a study, the methane concentration is 60% for 40 days despite the macrophytes Brachiaria, Oxycariumcubense, Eichhornia crassipes, Pistiastratiotes, and Salvinia being processed at 120°C, with Brachiaria having the greatest lignin content (Paulo et al., 2021). A study reported by (Barua and Kalamdhad, 2017) stated that highest biogas yield that can be achieved from untreated WH at an F/I ratio of 2 is 250±14 mL/g VS (57% CH_4) on the 32nd day whereas in the present study, the maximum biogas yield of 350 mL/g VS with 63% CH_4 was obtained at the same F/I ratio of 2:1 which is 40% higher, however the study is in agreement with the optimized F/I ratio of 2:1. Different studies yielded different amount of biogas and methane in certain peiod of time with varying F/I ratios. In a study reported by Simbayi, the biogas yield was 185 ml/g Vs with an F/I ratio of 2:1, generating 65% methane in 45 days (Simbayi et al., 2023). At the same F/I ratio, Wembe reported biogas yield of 175 ml/g Vs with 51% methane in 42 days (Wembe et al., 2023). At an F/I ratio of 1:1, three studies by Ali, Bote and Pachaiyappan yielded 226, 170 and 200-300 ml/g Vs of biogas with 50, 45 and 24% methane during 35, 32 and 30 days residence time respectively (Ali and Sun, 2019), (Bote et al., 2020) and (Pachaiyappan et al., 2014). It is interesting to note that the present study demonstrated superior biogas yields amongst all the reported literature at an F/I ratio of 2:1 with an yield of 350 mL/g $VS_{reduced}$.The methane content in biogas is also notable with 63% CH_4.

From roots of WH, the maximum biogas yield that can be obtained is 160 mL/g VSreduced at an F/I ratio of 2:1 at a corresponding VS reduction and CH_4 % of 60% and 62% respectively. Another interesting observation that can be made from Figure 4 is that concentration of CH_4in biogas can be achieved above 60% if the F/I ratio is maintained at 1:1 or 2:1 while it drops down to 56% if the F/I ratio is

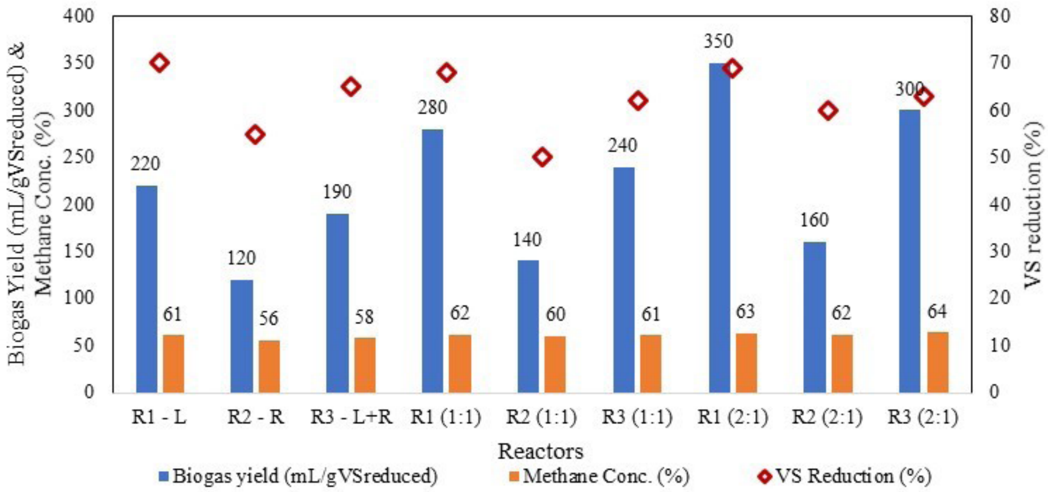

FIGURE 4: Performance evaluation of AD of WH at different F/I ratios and substrate composition in terms of VS reduction (%), methane yield and concentration.

increased to 5 which is evident from the numerical shown in R2-R and R3-L+R. In summary, the results presented in Figure 4 illustrate the AD of WH results in minimum yield of 220 mL/g VSreduced and maximum reaching up to 350 mL/g VSreduced upon maintaining suitable F/I ratio and substrate composition. The results obtained in this study agree with the study carried out by O'Sullivan et al., (O'Sullivan et al., 2010) who has demonstrated the biomethane potential of WH to be 292 ± 43 g/g VS respectively. The substrates such as food waste under different operating conditions results in the generation of broad range of cumulative biogas yields between 300-600 mL/g VS.

3.4 Effect of major performance indicators on the biogas generation potential of water hyacinth

The concentration of VFA in the reactors and its impact on pH was regarded as the major performance indicators in AD process. To assess these performance indicators, samples were withdrawn on 10th day and analysed for VFA and pH. The VFA, pH and biogas profile on 0th, 10th and 30th day along with% increase in VFA and biogas is presented in Table 3. It can be interpreted from Table 3, that the concentration of VFA increased substantially from 0 to 10th day in all the reactors with % increase in VFA ranging between 80 to 208%, which is indictive of dominance of hydrolysis in the reactors by the conversion of complex organic material to bio accessible organic acids than the methanogenesis, because the biogas generation from the reactors was less than 50% from day 0 to 10 while it increased substantially from day 11 to 30 (i.e., 52 to 208%), because the consumption of acids had taken place during that period of operation except for reactor R1(1:1) (Table 3) (Ganesh et al., 2005). The reactors R1, R2 and R3 operated with L, R and L+R and reactors at an F/I ratio of 2:1 showed relatively higher increase in VFAs compared to reactors at F/I ratio of 1:1 which could be attributed to the abundant availability

TABLE 3: Effect of performance indicators such as VFA and pH profile on biogas generation potential of WH.

Reactor	VFA (mg/L)					pH			Biogas generation, (mL/g VS)			
	0th Day	10th Day	30th Day	% Increase in VFA	% Utilization of VFA	0th Day	10th Day	30th Day	0th Day	10th Day	30th Day	% Increase in cumulative biogas
R1 - L	1.2	3.7	1.8	208	51	6.9	6.4	7.2	0	710	1754	147
R2- R	0.7	1.6	1.1	129	31	7.1	6.8	8.1	0	275	847	208
R3- L+R	0.8	2.4	1.2	200	50	7	6.6	7.6	0	385	1468	281
R1 (1:1)	1.1	2.9	1.4	164	52	6.8	6.6	7.1	0	640	700	9.4
R2 (1:1)	0.9	1.7	1.1	89	35	6.9	6.5	7.8	0	190	341	79
R3 (1:1)	1	1.8	1.2	80	33	6.9	6.7	7.5	0	360	655	82
R1 (2:1)	0.9	2.3	0.6	156	74	7	6.5	7.3	0	880	1516	72
R2 (2:1)	0.8	1.8	1.1	125	39	7.1	6.7	7.6	0	420	640	52
R3 (2:1)	0.8	2.1	0.7	163	67	7	6.6	7.2	0	660	1332	102

0th, 10th, and 30th day VFA is measured in 'mg/L'
0th, 10th, and 30th day Biogas generation is measured in 'L'

of food to micro-organisms for conversion while opposite is the case in reactors at an F/I ratio of 1:1. The increase in VFAs from day 0 to 10 resulted in the reduction of pH considerably to less than 6.8; therefore, the anaerobic chamber is purged with an inert gas, generally nitrogen, to remove most oxygen. However, the consumption of VFAs began after day 10 which in turn improved the biogas production due to the predominant activity of acetoclastic and hydrogenotrophic methanogens during acidogenic and methanogenic process of AD. This observation agrees with the observation made by (Barua and Kalamdhad, 2019) who had demonstrated the AD of pre-treated, untreated, and co-digested WH. These observations indicate that, to anaerobically digest the WH, a longer residence is needed which should be more than 30 days or a pre-treatment step is required to improve the hydrolysis rate and % increase in VFAs within 5 days of operation. On 30th day, the pH levels in the reactors varied from 7.2 to 8.1, indicating that the reactors were operating in anaerobic conditions with low levels of methane production.

3.5 Kinetic potential evaluation of biogas generation from water hyacinth

The kinetic parameters of AD of WH were evaluated using modified Gompertz model equation is presented in Table 4. It can be observed from Table 4 that the substrate composition and F/I ratio had its effect on the lag phase of the process. This is due to certain cells that exhibit growth and metabolic lag, particularly those with more stringent dietary needs. This time frame might align with the duration required for the concentration on essential intermediaries to build up (Bertranda, 2019). In particular, the lag phase in the reactors operated with roots alone and mixture of roots and leaves of WH showed longer lag phases ranging between 3.5 and 6.6 days while the reactors operated with leaves alone showed shorter lag phases ranging between 3.5 to 4.7 days. Furthermore, the rate of biogas generation from the reactors R2-R, R2 (1:1) and R3 (2:1) was 73, 27 and 83 mL/day respectively which is much lower compared to that of R1-L, R1 (1:1) and R1 (2:1).

The experimental data was found to be a good fit to the modified Gompertz model equation which can be ascertained from the R^2 values as shown in Table 4. The experimental and predicted biogas generation obtained from WH is plotted and shown in Figure 5. The difference in the experimental and predicted biogas is not substantial which again affirms the good fit of the experimental data to the Gompertz model equation.

3.6 Quantitative and qualitative evaluation of nutrients in biomanure and soil conditioner generated from water hyacinth in lab scale experiments

The sun dried anaerobic digestate i.e., biomanure and soil conditioner generated at the end of the AD and composting experiments respectively were further analysed for macronutrients, namely nitrogen (N), phosphorous (P_2O_5), and potash (K_2O). The quantity of biomanure and soil conditioner produced along with their nutrient concentration is tabulated and shown in Table 5. The quantity of biomanure produced after AD and composting was estimated by mass balance in terms of TS, VS and fixed solids (FS) as shown in Table 7. The nutrient values obtained from bioma-

TABLE 4: Kinetic potential parameters evaluated using modified Gompertz model.

Reactor Label	M (mL)	Rm (mL/day)	λ (Days)	R^2
R1 -L	2464	133	4.7	0.998
R2 -R	1122	73	6.6	0.998
R3 -L+R	1853	112	6.7	0.998
R1 (1:1)	1340	99	3.3	0.999
R2 (1:1)	531	27	3.6	0.996
R3 (1:1)	1015	87	5.6	1.0
R1 (2:1)	2396	133	3.5	1.0
R1 (2:1)	1060	83	4.5	0.998
R1 (2:1)	1992	105	3.5	0.999

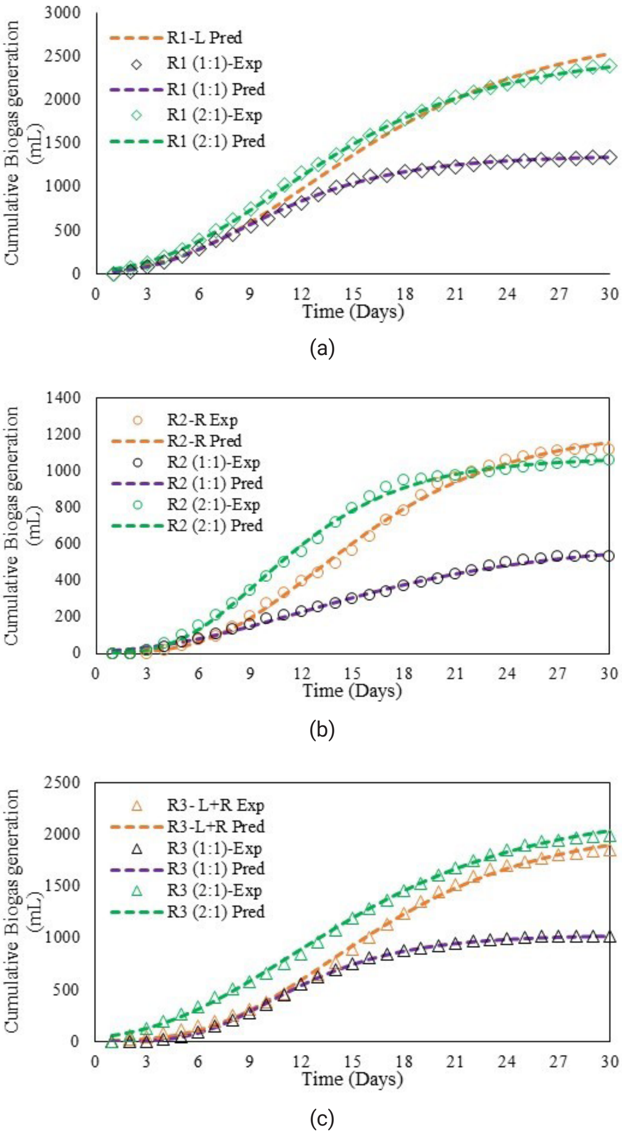

FIGURE 5: The experimental and predicted cumulative biogas generation profiles calculated at different F/I ratio using modified Gompertz model (a) R1; (b) R2 and (c) R3.

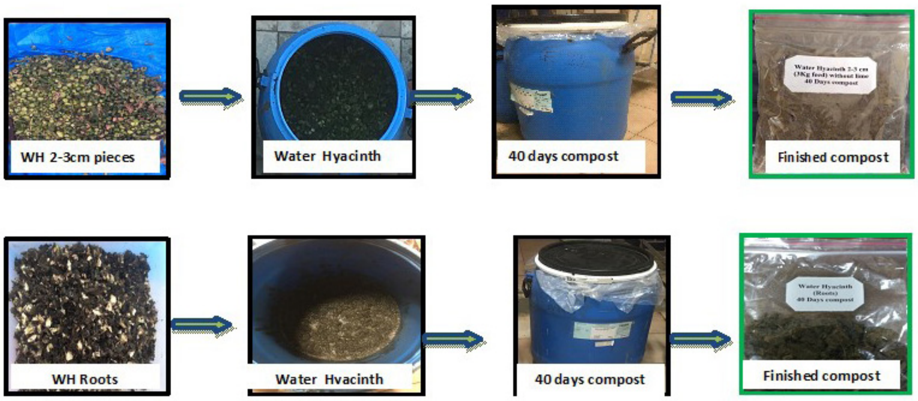

FIGURE 6: The whole WH and the chopped leaves, stalks and roots.

nure, and soil conditioner were compared with the standards given by the FCO (Fertilizer Control Order) for organic manure (FCO, 1985). The end product obtained from roots and leaves of WH is shown in Figure 6. From Table 5, a substantial difference in the quantity of biomanure and soil conditioner generation is observed i.e., 0.14 and 0.13 kg of soil conditioner is produced from 1 kg of substrate via composting process while in AD it was found to be 0.44, 0.62 and 0.53 g for leaves, roots and leaves + roots per g of total solids. For leaves and roots, WH determined that a C/N ratio of 25-32 was suitable to reduce or prevent ammonia inhibition at thermophilic AD (Krakat et al., 2017).

Further, the nutrient value in terms of N, P_2O_5 and K_2O for organic manure as per the standards of FCO is 0.5 each while in our study, the nutrients in leaves, roots and mixture of leaves and roots varied greatly in both AD and composting processes. The N concentration was relatively higher manure and conditioner produced from roots as feed either directly or in mixture with leaves. This could be due to natural assimilation of N from soil by roots compared to plant leaves and stem. Nevertheless, the low N in anaerobic digestate compared to anaerobic compost could be fact that, most of the N content would have been consumed by the methanogens for the generation of biogas as they require an optimum C/N ratio between 25 and 30. In the same way, P_2O_5 and K_2O concentrations found to be low in biomanure generated from both the rectors operated with leaves as feed than other two reactors. The results suggested that it is feasible to produce good quality fertilizer with desired macro nutrients from WH plant waste that is superior to the standards prescribed FCO with less cost.

3.7 Demonstration of accelerated anaerobic composting of WH to clean the KapraLake in Hyderabad city: A pilot scale societal project

The experimental results from the bench scale experiments of WH indicated that composting could be one of the best options to be implemented at pilot or full scale to create wealth out of waste weed. Composting the leaves and roots of WH is done separately because the concentration of heavy metals is high in roots with more absorption capacity. One of the lakes in Hyderabad city called Kapra lake or Oora Cheruvu which spreads around 1254 m near Sainikpuri in the north-east part of Greater Hyderabad has become hazardous to the aquatic fauna ("Kapra Lake," n.d.). Therefore, in association with the local policy makers, an initiative has been taken to install the pilot plant at Kapra lake, as it is filled with WH. The composting process proven successful in the laboratory experiments and has been implemented at the pilot facility where, 1200 tons of WH is pulled out using JCB and boats followed by washing and chopping into roots, stalks and leaves separately as shown in Figure 7. The leaves and roots were chopped into small pieces using a chaffer machine. About 30 compost pits have been constructed for composting, 15 each for roots and leaves of WH and about 4 tons of chopped WH was fed into each pit. The compost pits were initially added with equal quantity of cattle dung and AMMC developed in the laboratory to maintain the methanogenic population and subsequently the WH was thoroughly mixed with the inoculum in each pit maintaining appropriate moisture. Daily samples were collected through sampling ports were made to ensure

TABLE 5: Comparison of quantity and quality of biomanure and soil conditioner to that of organic manure by FCO.

Parameter/ Label	Biomanure (kg/kg substrate)			Soil Conditioner (kg/kg substrate)		FCO standard (min)	Standard deviation (s)
	R1-L	R2-R	R3-L+R	ACR1	ACR2	Organic manure	
Kg product/ kg substrate	0.44	0.62	0.53	0.14	0.13	-	0.225
N (%)	0.32	0.85	0.64	0.46	10.09	0.5	4.263
P_2O_5 (%)	0.16	0.42	0.38	0.48	0.46	0.5	0.151
K_2O (%)	0.18	0.34	0.36	0.5	0.57	0.5	0.151

TABLE 6: Detailed compositional analysis of soil conditioner produced from leaves, roots and organic manure by FCO.

Parameters	Water hyacinth roots	Water hyacinth leaves	According to FCO, Schedule-iv, specification of organic matter Standard values - Organic manure	Standard deviation (S)
MC by weight (%)	32.33	12.18	25	14.248
Colour	Brown	Green	-	-
Odor	Ammonia smell	Wet grass smell	-	-
Bulk Density (g/cm^3)	0.21	0.31	<1.0	0.070
Total Organic Carbon, wt. (%), minimum	16.23	0.83	14	10.889
Total Nitrogen (as N), wt. (%), minimum	10.09	0.46	0.5	6.809
Total Phosphates (as P_2O_5), wt. (%), minimum	0.46	0.48	0.5	0.014
Total Potash (as K_2O), wt. (%), minimum	0.57	0.50	0.5	0.049
C:N ratio	19:1	39:1	<20	-
pH	8.72	9.10	6.5-7.5	0.268
Conductivity (as dsm-1), Not more than	2.16	1.21	Not more than-4.0	0.671
Arsenic (as As_2O_3) (mg/kg)	2.87	Below detection	Maximum- 10	-
Cadmium (as Cd) (mg/kg)	0.19	Below detection	Maximum-5	-
Chromium (as Cr) (mg/kg)	5.69	3.69	Maximum-50	1.414
Copper (as Cu) (mg/kg)	11.31	1.64	Maximum-300	6.837
Mercury (as Hg) (mg/kg)	0.47	Below detection	Maximum-0.15	-
Nickel (as Ni) (mg/kg)	1.54	0.12	Maximum-50	1.004
Lead (as Pb) (mg/kg)	2.94	0.59	-	1.661
Zinc (as Zn) (mg/kg)	22.80	8.93	Maximum 1000	9.807

TABLE 7: Mass balance to estimate the generation of biomanure from AD of WH.

Anaerobic digestion (AD) of WH											
	$TS_{Substrate}$ (g)	TS_{AMMC} (g)	$VS_{Substrate}$ (g)	VS_{AMMC} (g)	TS_{Total} (g)	VS_{Total} (g)	FS (g)	VS reduction (g)	VS remaining (g)	VS+FS (g)	g manure/g of TS or kg/kg of TS
R1- L	16	4	16	3	20	19	1	11	8	9	0.4
R2- R	20	4	17	3	24	20	4	9	11	15	0.6
R3-L+R	17	4	15	3	21	18	3	10	8	11	0.5
Accelerated Anaerobic Composting (AAC) of WH											
	$TS_{Substrate}$ (kg)	TS_{CD} (kg)	TS_{AMMC} (kg)	$VS_{Substrate}$ (kg)	VS_{CD} (kg)	VS_{AMMC} (kg)	FS (kg)	VS increment (kg)	VS remaining (kg)	VS+FS (kg)	kg soil conditioner/kg of substrate
ACR1	1.6	0.6	0.2	1.6	2	0.1	1	0.1	3.7	5	0.1
ACR2	2.1	0.6	0.2	1.7	2	0.1	1	0.2	3.9	5	0.1

that the proper environmental and operational conditions were being maintained.

Temperature, pH, alkalinity and toxicity, mixing, nutrient requirements, sludge retention time (SRT), and microbial capacity are some of the factors that influence the AAC at different stages. The final compost after 40 days was removed, and sun dried to get the solid compost. The final product obtained from roots and leaves was evaluated for detailed compositional analysis as shown in Table 6. Although the nutrients N P and K value of soil conditioner agreed with the organic manure by FCO but the concentration of heavy metals in roots was relatively higher which could indirectly impact the human health. Therefore, the application of soil conditioner produced from leaves was found to be suitable for agriculture field application while that of roots was found suitable for horticulture. In light of this, both the soil conditioners from leaves and roots were tested in real field for the growth of different plants and differences were observed and explained in subsequent section. The dried product named as "Soil Conditioner" was

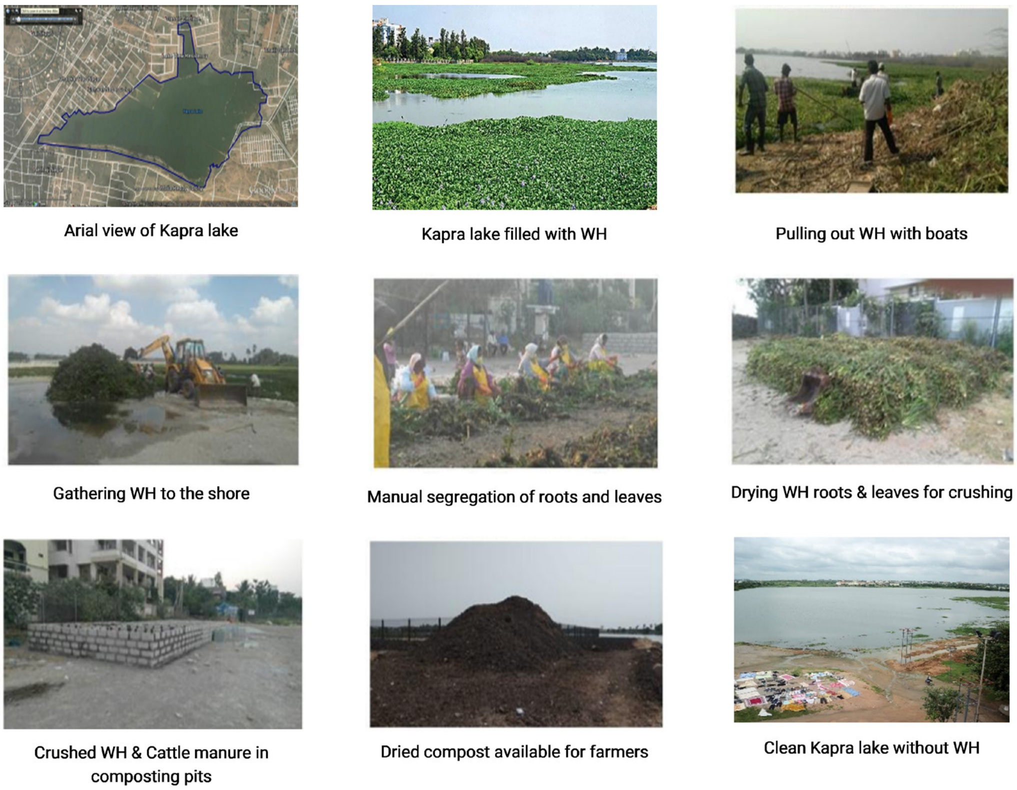

FIGURE 7: Composting process implemented for the removal and treatment of WH from Kapra Lake, Hyderabad, India.

packed and sealed for distributing into the outlets for sale. As anaerobic inoculum and cow dung were added which was kept under anaerobic conditions, biogas including methane will be produced. The biogas thus produced was in turn used as carbon source of energy for the compost microbes in the production of microbial cells (Hamer et al., 1967). The product obtained was stable and the odors were also resistible with wet grass smell in its vicinity.

3.8 Field application of biomanure and soil conditioner: Response of Fenugreek and Coriandrum sativum plants

The soil conditioner produced from leaves was tested in real field for the growth of Fenugreek and Coriandrum Sativum plants and the periodic growth of the plants from seeding to complete plant i.e., 0 to 30 days is shown in Figure 8 and 9, while the soil conditioner from roots was applied for horticulture and the impact of it on the growth and freshness of plants and flowers is shown in Figure 10 respectively. To compare the growth of Fenugreek and Coriandrum upon application of soil conditioner from WH leaves, commercially available general compost was also administered and the control plants were grown without the application of any fertilizer for proportional evaluation. The response of the plant growth was measured on every 5th day from seeding and the height of the plants was measured in 'cm', as plant height is the key parameter to determine the impact of any fertilizer. One of the interesting observations that can be made from Figure 8 and 9 is that the germination of Coriandrum seeds was delayed by 5 days compared to Fenugreek plants which grew about 2 cm by the end of 5th day in all the cases. Another intriguing and common observation from Figure 8 and 9 is that the height of Fenugreek plants is relatively higher than Coriandrum and the plant growth with soil conditioner and general compost administered plants was relatively higher than control which illustrates that the application of soil conditioner and compost has promoted the growth of plants rapidly. Therefore, the soil conditioner produced from leaves of WH was found suitable for application as a soil conditioner in the agriculture fields.

Similarly, the soil conditioner developed from roots of WH was administered to Hibiscus rosa-sinensis and Carica papaya plants to check the growth of the plants. As can be seen from Figure 10 that the soil conditioner administered hibiscus flower and papaya plant was bright and fresh compared to control. This is indictive of positive impact of soil conditioner on the growth horticulture plants.

4. CONCLUSIONS

This study investigated the feasible ways to valorize WH by two methods namely, anaerobic digestion for the generation of biogas and biomanure and secondly, accelerated anaerobic composting for the conversion of WH to soil conditioner at bench and pilot scale. The highest biogas yield of 350 mL/g VSreduced with 63% CH_4 can be achieved from leaves of WH at an F/I ratio of 2:1 followed by roots in combination with leaves. While no substantial biogas generation was observed from roots which could be attributed to high concentration of heavy metals. Furthermore, the results illustrate the positive impact on agricul-

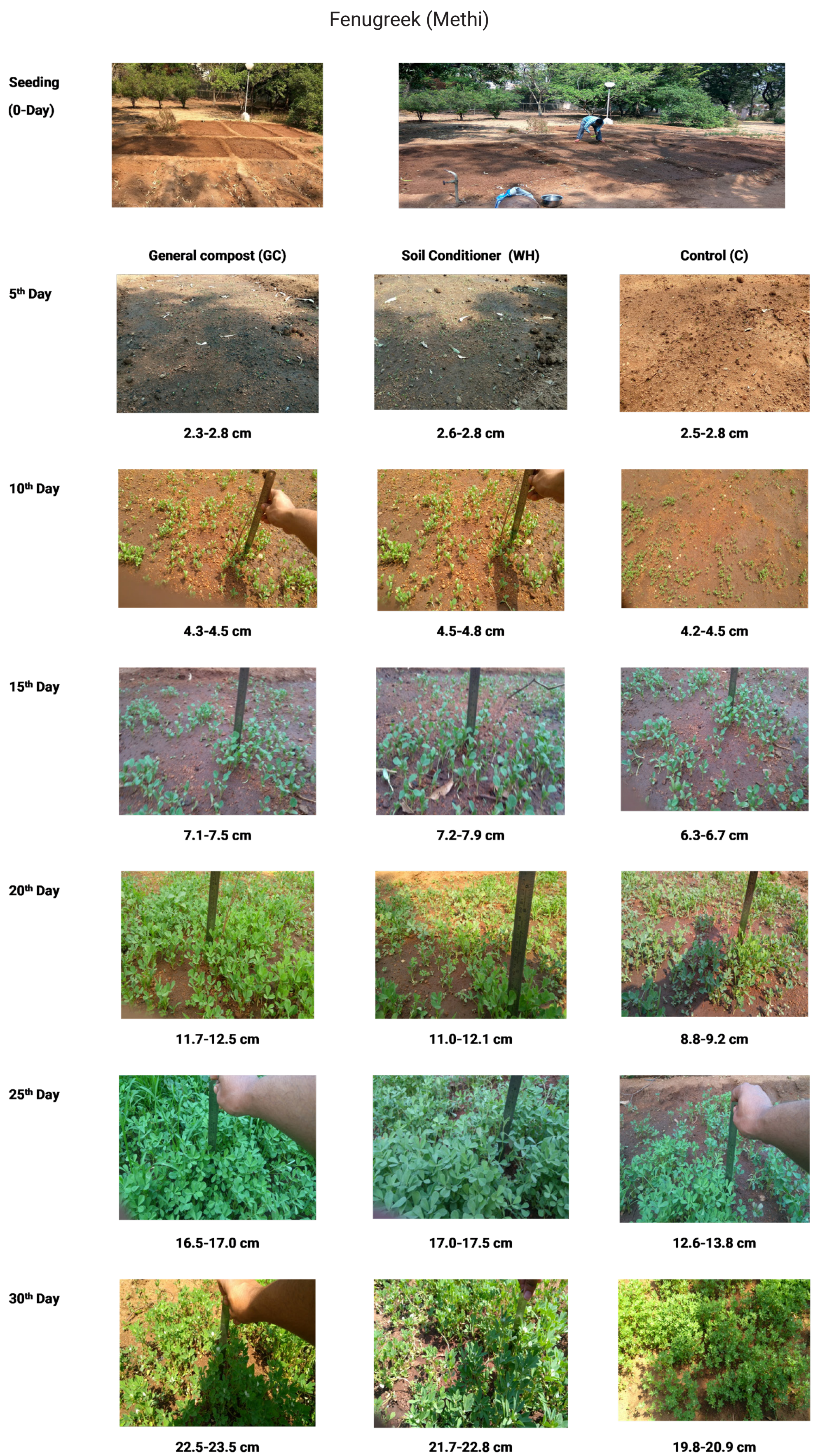

FIGURE 8: Comparison of Fenugreek plant growth estimation from 0 to 30th day upon application of general compost and soil conditioner over the control.

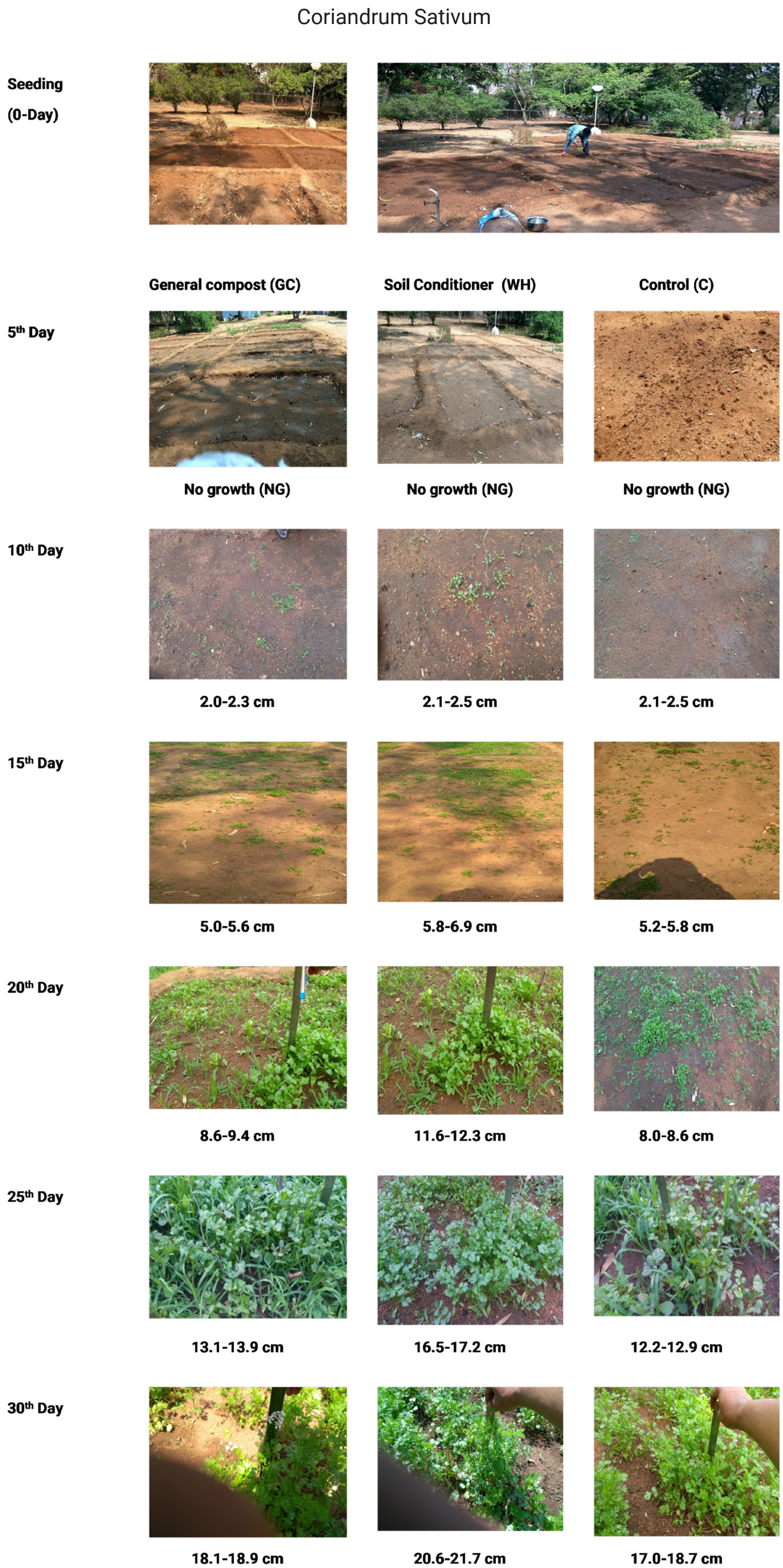

FIGURE 9: Comparison of Coriandrum Sativum plant growth estimation from 0 to 30th day upon application of general compost and soil conditioner over the control.

Control – without ferilizer Soil Conditioner

FIGURE 10: Application of soil conditioner produced from roots of WH on Hibiscus rosa-sinensis and Carica papaya plants.

ture farming (Fenugreek and Coriandrum Sativum) using soil conditioner produced from leaves of WH while that of roots was suitable for horticulture (Hibiscus rosa-sinensis and Carica papaya plants). Heavy metal presence in roots inhibits AD whereas whole plant material used for composting towards biofertilizer/ soil conditioner production made composting of WH more viable than AD. The pilot scale demonstration was found to be success with 1200 tons of WH removed from Kapra lake and converted to 120 tons of soil conditioner.

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