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Efficiency of *Rhodopseudomonas faecalis* in Increasing Digestion of Substrate in an Anaerobic Digester

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ABSTRACT: An anaerobic Purple Non Sulphur Bacteria (PNSB), *Rhodopseudomonas faecalis* was utilized as an inoculum to study the efficiency of a biogas digester. A 5 litre capacity anaerobic digester was prepared with 10% feed, 30 days of Hydraulic Retention Time (HRT) was studied using canteen waste as a substrate. Parameters like pH, Temperature, Chemical Oxygen Demand (COD), Total Solids (TS), Volatile Solids (VS) and gas production were analyzed and recorded on every day. It was found that pH was decreased and temperature was varying. Increase in the digestion of the substrates was due to the addition of the bacterium eventually causing decrease in COD, TS & VS, was observed. This has increased the production of biogas in the digester.

KEYWORDS: Biogas, Canteen waste, *Rhodopseudomonas faecalis,* COD, TS, VS.

INTRODUCTION

Anaerobic Digestion (AD) is a process, an efficient technology and a mechanism because it minimizes greenhouse gas emissions (Hidalgo D et al., 2012) and manages the organic waste that would otherwise be dumped into landfills to recover energy (Esteban-Gutiérrez, M et al.,2018). Besides, there is an increased interest in next level AD processes that enhances the biogas production and also reduce the use of fossil fuels (Arafat H.A et al.,2015). Moreover advanced AD processes are yet to be explored through techno-feasibility lenses and there have been a minimal industrial operations (García-Depraect, O et al.,2017).

Food waste is abundantly available waste biomass that contains high moisture and readily degradable organic matter. This type of characteristics make it an attractive feedstock for energy recovery from the AD process (Dhamodharan, K et al.,2015). Waste of food is actively connected to the waste of energy and production of greenhouse gases. An average hotel or restaurant business has a varied different waste pattern than any other business wherein 39 percentage is biodegradable kitchen and canteen waste (Emil Juvan et al.,2023) and 1% is other waste which is biodegradable. About 60% of waste is Non-Biodegradable (Fig 1). Various attempts are being made to optimize energy recovery from food waste through AD processes. Technically, AD is a matured technology that happens in the anaerobic condition through a series of processes viz., hydrolysis, acidogenesis, acetogenesis, and methanogenesis (Rivière, D et al.,2009). AD involves mesophilic, thermophilic and psychrophilic microorganisms which can survive at different temperatures corresponding to 20–45 ◦C, >50 ◦C and 4–25 ◦C respectively (Schober G et al., 1999).

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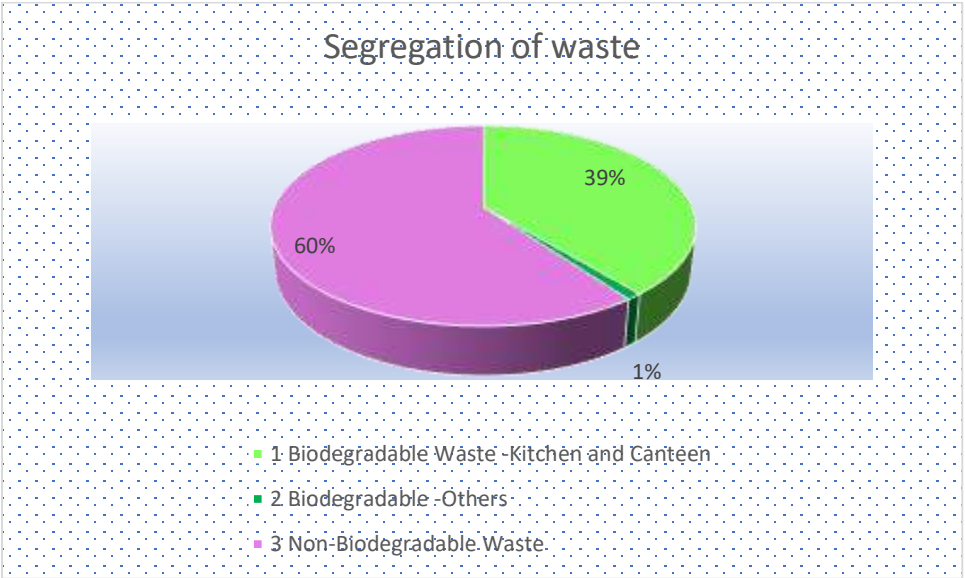

Fig 1: Segregation of waste (Biodegradable and Non Biodegradable)

Anaerobic digestion is a complex biochemical process where microbes decompose organic matter and produce biogas. A number of parameters could influence the performance and biogas production for a semicontinuous and or continuous anaerobic digestion process (R. Chandra, H. Takeuchi, and T. Hasegawa, 2012) viz., substrate characteristics, organic loading rate (OLR), hydraulic retention time (HRT), temperature, and pH. Hydraulic Retention Time is an important operational parameter for anaerobic reactors or digesters which can regulate the conversion of volatile solids (VS) into biogas (M. A. Dareioti and M. Kornaros, 2014 ; D. Ho, P. Jensen, and D. Batstone,2014). Biogas is a renewable fuel produced from anaerobic digestion of organic material or feedstocks that includes municipal waste, food waste, energy crops and farm waste. Raw biogas typically consists of methane (50–75%), carbon dioxide (25–50%), and minor amounts of nitrogen (2–8%). Trace levels of hydrogen sulfide, hydrogen, ammonia and various volatile organic compounds are also present in biogas depending on the raw material (Wellinger A et al., 2013).

The main objective of this research was to check the efficiency of an anaerobic digestion of canteen waste in a 5 litre capacity biogas digester. The pH, Temperature, COD, TS, VS and gas production were selected as the evaluated factors to study the influence of OLR on HRT. Further a novel strain of *Rhodopseudomonas faecalis* was utilized as an inoculum in to the biogas digester. This has observed with the increase in efficiency of biogas production by degrading substrates efficiently.

METHODOLOGY

Substrate and Anaerobic Digester: A 5 litre working volume of Biogas digester was setup using HDPE plastic bottles. A canteen waste collected from various sources consisting of fruits, vegetables and cooked food etc., which is a semisolid slurry was grinded, sieved to remove large fibrous material and was poured in to the anaerobic digester. It is a closed system covered and is exposed to light at room temperature.

Organism: Previously, a novel strain of PNSB anerobic bacteria was screened from the samples collected from Sewage Treatment Plant (STP) of ALEAP Industrial Park, Hyderabad. It was isolated, purified and identified as *Rhodopseudomonas faecalis* through 16s RNA sequencing method. The cultures were maintained and 2.5% of inoculum was added in to a 5 litre capacity anerobic digester.

ANALYTICAL METHODS

The daily biogas production was measured by measuring cylinder through gas displacement method. Samples from the digester were daily collected for measurements of pH using pH meter and temperature with Thermometer. The Total Solid (TS) and Volatile Solid (VS) were analysed using standard methods (APHA 2005) where it was determined by measuring the weight difference of a

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sample. The sample was placed in an oven at 105 °C for 24 h. The weight was measured by taking the sample before and after it was oven-dried. The sample was kept in a desiccator after withdrawal to prevent the absorption of moisture while the crucibles were cooled. The VS of the samples was determined in a muffle furnace at 550 °C for 15 to 20 minutes. A 10% of Organic Loading Rate (OLR) and 30 days of HRT was maintained. COD was calculated using Titrimetric method (Burns E. R and Marshall C 1965). All measurements were carried out in triplicates. The conceptual flowchart of the research methodology was explained in the figure 2.

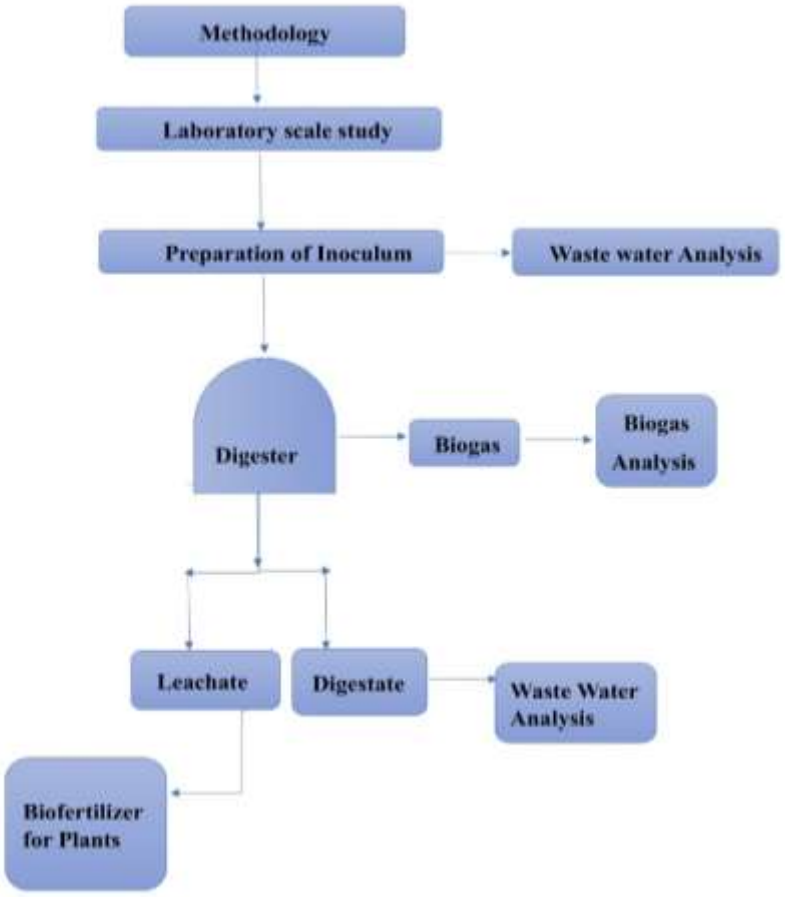

Fig 2 : Conceptual flowchart of research methodology

RESULTS & DISCUSSION

The experiment was carried out for the period of 30 days (HRT) for anaerobic digester of 5L capacity with 10% canteen waste as a substrate. The following results were observed for pH, Temperature, COD, TS, VS and Gas production. The graphs represent the control and test where the test results were recorded after the addition of inoculum of *Rhodopseudomonas faecalis* in the bioreactor where the efficiency of the digester was enhanced by considerable degradation of substrates with respect to TS and VS.

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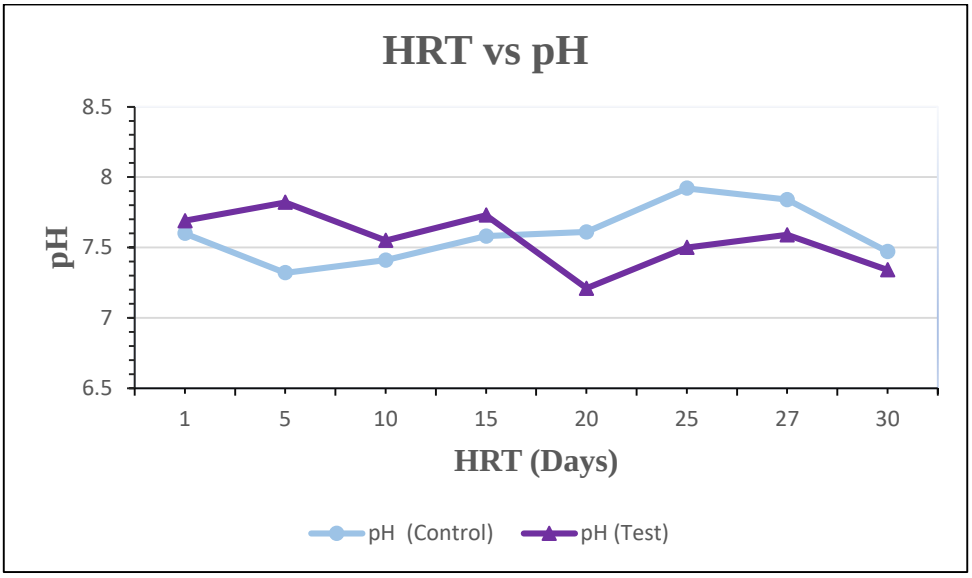

Fig 3: HRT vs pH showing Control and Test

The graph between Hydraulic Retention Time and pH shows that the pH value was decreased in test (7.21) when compared to control (7.32) and this decrease was because of the production of volatile fatty acids as shown in Figure 3.

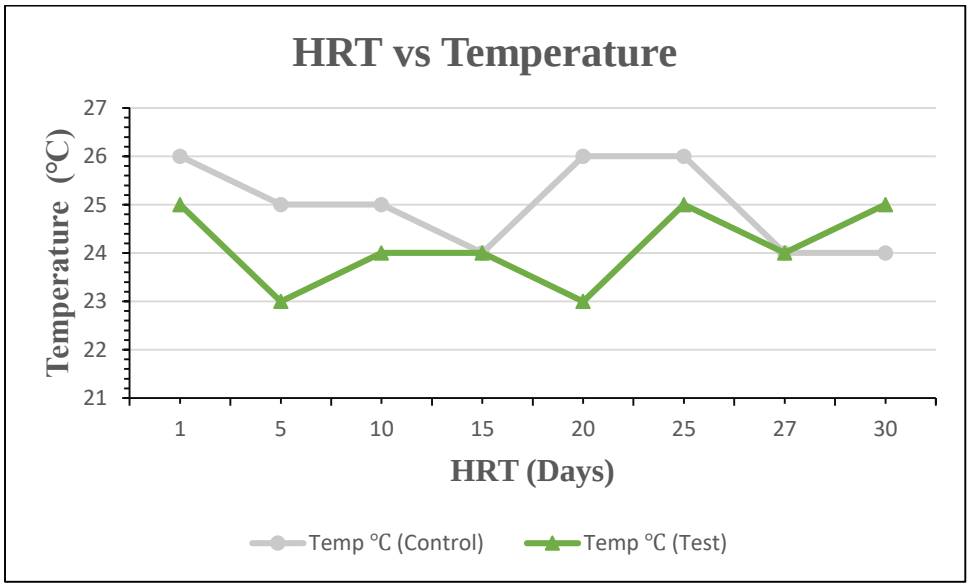

Fig 4: HRT vs Temperature showing Control and Test

The graph between Hydraulic Retention Time and Temperature shows that it was varied from 24° C to 26 ° C for the control whereas it was varied from 23° C to 25 ° C for test as shown in Figure 4.

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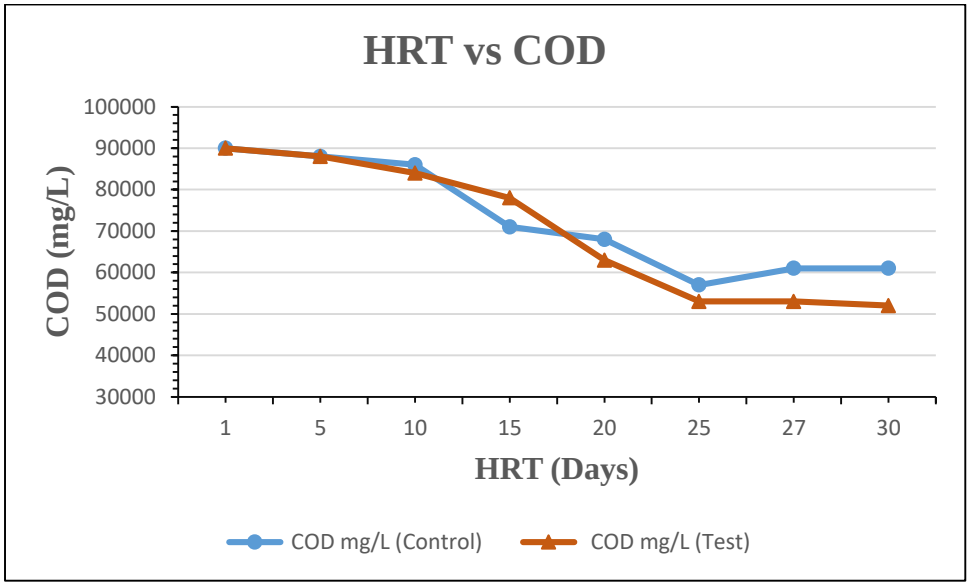

Fig 5: HRT vs COD showing Control and Test

Figure 5 shows the graph between Hydraulic Retention Time and Chemical Oxygen Demand (COD). Treatment Efficiency (TE) of the digester with respect to COD is calculated by using initial and final COD values in mg/L. Percentage of Treatment Efficiency of control was 32.22% and whereas test was 42.22%.

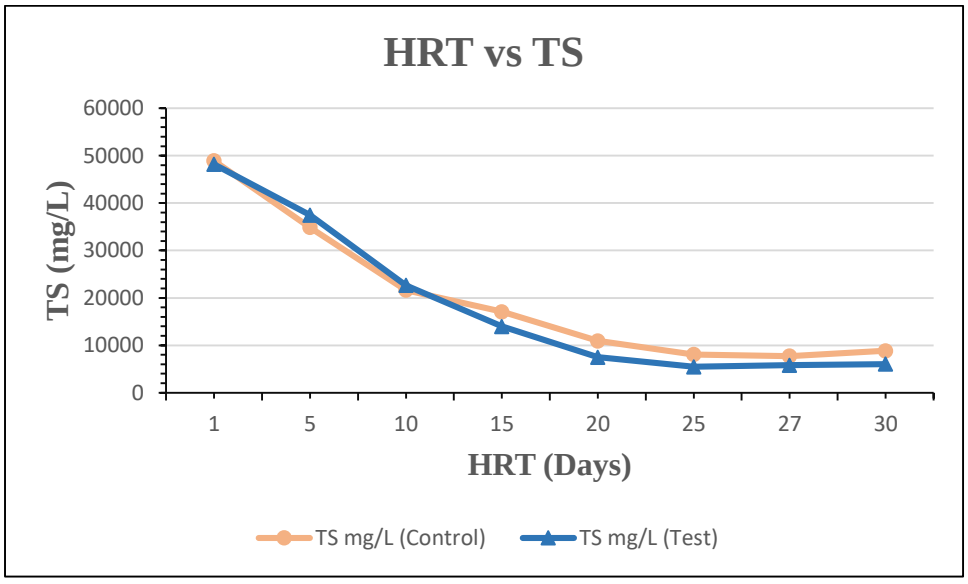

Fig 6: HRT vs TS showing Control and Test

Figure 6 shows the graph between Hydraulic Retention Time and Total Solids (TS) that, percentage of decrease in TS for control was 81.94% whereas TS for test it was 87.45%.

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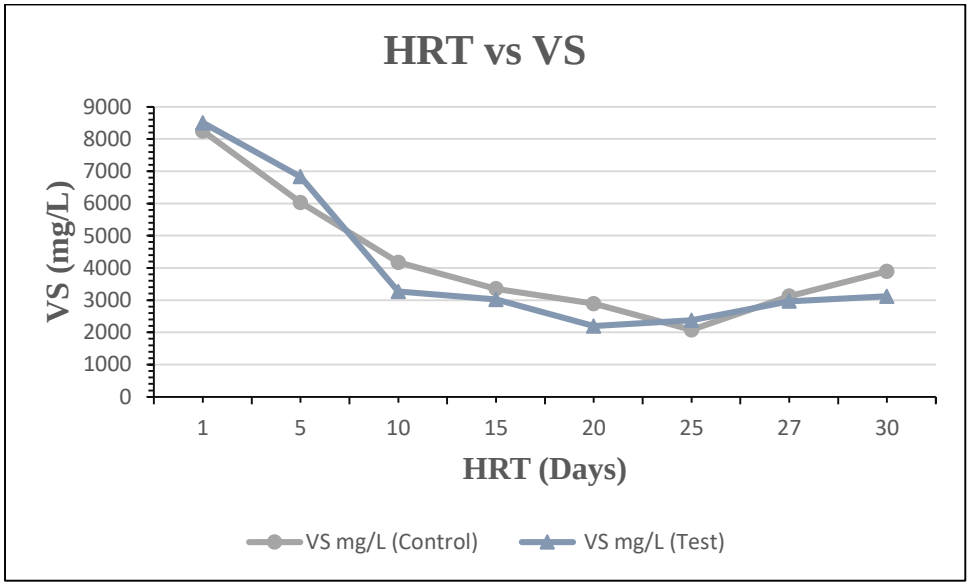

Fig 7: HRT vs VS showing Control and Test

The graph between Hydraulic Retention Time and Volatile Solids (VS) shows that, percentage of decrease in VS for control was 52.78% whereas for test it was 65.15% as shown in Figure 7.

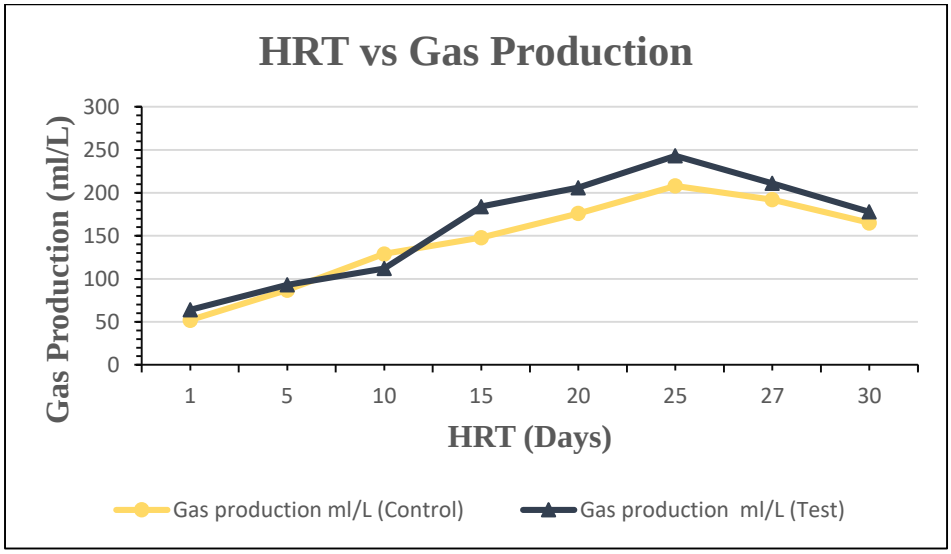

Fig 8: HRT vs Gas Production showing Control and Test

The graph between Hydraulic Retention Time and Gas production shows that it was 208 ml/L for control and it was increased to 243 ml/L for test which has *Rhodopseudomonas faecalis* as shown in Figure 8.

Table 1: Digestate analysis

S.NO	Parameters	Control (%)	Test (%,with culture)
1	COD (TE)	32.22	42.22
2	TS	81.94	87.45
3	VS	52.78	65.15
4	Gas Production at 25th Day	208 ml/L	243 ml/L

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Table 1 shows different operational parameters analyzed for the digestate with control and test (with culture) of a digester consisting of canteen waste as substate at 10% OLR and 30 days HRT.

CONCLUSION

The test consisting of *Rhodopseudomonas faecalis* as an inoculum in a 5 litre capacity biogas digester with 10% OLR and 30 days HRT for canteen waste as a substrate have shown results with a decrease in pH to 7.21 and temperature varied from 23° C to 25° C. Percentage of treatment efficiency of the digester with respect to COD of test was 42.22%, percentage of decrease in TS was 87.45%, for Volatile Solids it was 65.15% whereas gas production was increased to 243 ml/L. This shows that the addition of the novel strain of *Rhodopseudomonas faecalis* have increased the efficiency in digestion of the substrate of an anaerobic digester.

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