Bioremediation and Bioprospecting of Cow Dung and Poultry Droppings Enriched with Sewage Water for Biogas Production

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Abstract

In this study, the biogas was produced by the gradual replacement of cow dung using sewage water and poultry dropping. Cow dung replacement with sewage water showed high utility of the total solids than the control bioreactor. In control bioreactor the total solid reduction is ranged between 10% and 9.8%, whereas in the experimental bioreactor which contained the gradual replacement of cow dung with sewage water, the total solid degradation occurred from 10% to 6.1%. The analysis of biogas production by gradual replacement of cow dung with poultry droppings and sewage water revealed the total solids degradation range from 7.2% to 6.7%. Total solid level reduction is considered to be one of the important parameter for biogas production. Regarding the production of biogas, the gradual replacement of cow dung with sewage water experimental reactor gave more biogas (1421lit/kg of dry matter/day) when compare to the control bioreactor (1007lit/k g of dry matter/day). The maximum gas production also occurred at 80% replacement with sewage water. The gradual replacement of cow dung with poultry droppings supplemented with sewage water revealed the high gas production (1952lit/kg of dry matter/day) than the cow dung replacement with sewage water and control bioreactors. During the gradual replacement of cow dung with poultry droppings and sewage water concluded that there is an excellent biogas production in the 100%replacement of cow dung which in turn indicated that poultry droppings contains more total solid level which can be easily degraded by methanogenic organisms than in cow dung and sewage water can be a good nutrient source for biogas production.

Keywords: Total solid level, biogas production, methanogenic organisms, sewage water.
INTRODUCTION

Agro-industrial residues, sewage water, large poultry and pig farms are often a major source of pollution in Asian countries. The production, distribution, use, misuse, disposal of sewage water have polluted the environment that threatens the health of humans, livestock, wildlife and indeed whole ecosystems. At the same time these pollutants constitute a large potential for biogas production through anaerobic digestion. The government, industry and the public have needed the effective alternatives to traditional physical and chemical methods for sewage management.

In India, the biogas technology is based on cattle dung as the main feed stock. Biogas has been produced traditionally from cattle dung but the reports about the need of cattle dung has been increased in years because of the reduced number of cattle. The alternate substrates like excreta of sheep, goat, pig and other animal wastes both in combination with cattle dung and alone are possible to produce biogas.

The raising cost and shortage of conventional fuel have generated renewed interest in producing methane from organic matter through anaerobic digestion. The production of energy from alternative sources becomes not only more desirable but economically more feasible. Biogas is one of the promising sources of alternative energy and the biogas technology of modern plant produces clean renewable with nutrient rich digested slurry.

Technologies for treating farm wastes along with sewage polluted environments have been a major concern over the last couple of decades. Research on anaerobic degradation of cellulosic wastes of cattle dung for enhanced biogas and ethanol production has shown the degrading ability of rumen microorganisms in biogas production. The successive biogas production occurred when poultry droppings, parthenium and eucalyptus leaves with donkey dung combination.

The temperature has a significant influence on methanogenic bacterial activity, bioremediation and stabilization efficiency in biogas production. The effect of temperature is independent of loading rate and retention time. The methanogenic activity, anaerobic biodegradability and toxicity are key parameters in the design and operation of anaerobic bioreactor. There was no significant inhibition of biogas production in the presence of salinity but this salinity along with ammonium nitrogen levels have an impact on biogas production.

Chemical oxygen demand, dry solids, volatile fatty acids and reactor volume occupied by the feed material are the important parameters in the biogas production. Effective bio conversion of organic matter in anaerobic digester depends on a diverse microbial population. The slurry from biogas plant after biogas production used as a nutrient source in agriculture. The bio digested slurries of biogas plant used as carriers for the preparation of carrier based inoculums acclaimed to play a vital role in modern agriculture. In this study, the sewage water which is consider to be the chief source of contamination used as supplementary feed and replacement source along with cow dung and poultry droppings for biogas production.

MATERIALS AND METHODS

Sample Collection

The present work was carried out using sewage water of Sewage effluent Treatment Plant (STP) of A.P.C. Mahalaxmi College campus, Thoothukudi. This is used as supplement for the production of biogas. The sewage water for STP is directly collected from hostel septic tanks, old hostel, new hostel and mess.

Characteristics of sewage water

The following parameters were analyzed in sewage water both in control and experimental bioreactors. The Total Solids (TS), Volatile Fatty Acids (VFA) and Total Volatile Solids (TVS) were estimated as per the procedure given in MACs manual.

Bioreactor and loading Mode

In this experiment KVIC (Kadhi Village Industries Commission) model anaerobic bioreactors were used for biomethanation process. Semi continuous process was followed for biogas production. Anaerobic bioreactors were used. The total capacity of a bioreactor is 46 liters. Cow dung and poultry droppings were used as substrates. For experimental purpose these substrates were supplemented with sewage water and the biogas production was done in a separate
manner. The digesters were daily charged by these substrates for their stabilization. The experiment was carried out for the period of nearly 250 days continuously.

(i) In the first part of work, the control bioreactor was loaded with cow dung and ordinary water and the experimental bioreactor was loaded with cow dung supplemented with sewage water. The Retention Period (RT) for 30 days was maintained. After 30 days the gas output was measured by water replacement method.

(ii) In the second part of work, the cow dung amount was gradually reduced by sewage water in the order of 20%, 40%, 60%, 80% and 100%. Total Solid (TS) concentration was maintained at 10% level. The gas output was measured by water replacement method.

(iii) Another set of experiment setup was carried out in the above said same manner but the cow dung was replaced in the order of 20%, 40%, 60%, 80% and 100% level by poultry droppings and sewage water in the interval of 15 days. The sewage water level was maintained constantly. Here also the gas output was measured by water replacement method.

### Table 1. Characteristic features of sewage water

| TS(%) | TVS(%) | N(%) | P(%) | K(%) | C(%) |
|-------|--------|------|------|------|------|
| 9.360±3.207 | 60.290±2.472 | 1.12±2.039 | 0.32±1.074 | 0.28±1.937 | 28.2±3.008 |

### Table 2. Total Solids (TS), Total Volatile Solids (TVS) and Volatile Fatty Acids (VFA) in control bioreactor

| No of days | Initial TS (%) | Every 15 days TS (%) | Initial TVS (%) | Every 15 days TVS (%) | VFA (%) |
|------------|----------------|----------------------|----------------|----------------------|--------|
| 0th day    | 10.0±2.516     | 7.6±1.835            | 60.1±0.311     | 62.2±1.997           | 42.0±0.057 |
| 15th day   | 10.0±1.732     | 7.9±1.646            | 60.3±0.352     | 64.2±0.900           | 44.0±0.132 |
| 30th day   | 9.8±1.250      | 7.6±2.657            | 61.7±0.702     | 66.2±0.950           | 40.0±0.374 |
| 45th day   | 9.4±1.692      | 7.6±1.662            | 60.7±0.680     | 64.8±1.193           | 43.0±0.497 |
| 60th day   | 9.6±1.587      | 7.8±1.743            | 59.5±0.789     | 62.4±0.680           | 40.0±0.164 |
| 75th day   | 9.4±2.85       | 6.6±0.208            | 60.3±1.200     | 63.8±2.066           | 40.0±0.923 |
| 90th day   | 9.8±0.513      | 6.4±0.351            | 61.4±0.808     | 64.2±1.153           | 42.0±0.947 |

Values in Mean ± Standard Deviation.

### Table 3. Total Solids (TS), Total Volatile Solids (TVS) and Volatile Fatty Acids (VFA) in Experimental bioreactor (cow dung replacement with sewage water)

| No of days | Amount of cow dung replacement with sewage water | Initial TS (%) | Every 15 days TS (%) | Initial TVS (%) | Every 15 days TVS (%) | VFA (%) |
|------------|-----------------------------------------------|----------------|----------------------|----------------|----------------------|--------|
| 0th day    | 0%                                            | 10.0±0.100     | 6.3±1.053            | 57.4±0.585     | 65.8±1.504           | 52.0±1.230 |
| 15th day   | 20%                                           | 9.8±0.305      | 6.6±0.608            | 58.9±1.792     | 66.2±1.150           | 50.0±0.870 |
| 30th day   | 40%                                           | 9.4±0.493      | 6.9±0.550            | 57.3±0.642     | 66.8±2.954           | 58.0±0.043 |
| 45th day   | 60%                                           | 9.0±0.110      | 6.1±0.150            | 56.9±2.668     | 64.9±1.616           | 48.0±0.032 |
| 60th day   | 80%                                           | 8.8±0.152      | 6.8±0.519            | 61.9±2.193     | 72.8±1.101           | 48.0±0.743 |
| 75th day   | 100%                                          | 8.4±0.757      | 7.2±1.106            | 59.6±1.817     | 68.2±5.466           | 46.0±1.687 |
| 90th day   | 100%                                          | 8.1±1.156      | 7.1±0.251            | 59.5±2.569     | 68.8±6.086           | 52.0±1.1043 |

Values in Mean ± Standard Deviation.
Characteristics of outlet slurry
The parameter analyses of outlet slurry were also done. The important parameters, pH and temperature were analyzed routinely both in control and experimental bioreactor. The temperature was ranged between 36°C to 38°C throughout the experimental period.

Statistical analysis
The characteristic feature of sewage water (Table 1), Total solids (TS), Total Volatile Solids (TVS) and Volatile Fatty Acids (VFA) in control bioreactor (Table 2) and experimental bioreactor of cow dung replaced with sewage water (Table 3) and all the experimental data were calculated by the average (mean) and standard deviation and given in the table as mean±SD by using Microsoft Excel. Then the values were tested by ANOVA (analysis of variance) which revealed that there is a significant difference between the control and experimental bioreactor in gradual replacement of cow dung with sewage water (Table 4). The Student’s ‘t’ test to explain the impact and individual analysis of TS, TVS & VFA for the biogas production in the case of gradual cow dung replacement with sewage water (Table 5). The observed values of temperature and pH of both control and experimental bioreactors

| Sources of Variation | Degrees of freedom | Sum of squares(x) | Mean sum of squares(x) | Mean sum of squares(y) | Table value of F |
|----------------------|--------------------|-------------------|------------------------|------------------------|-----------------|
| Between Means        | 2                  | 17016.28          | 8508.14                | 9634.61                | 3.55            |
| Between products     | 18                 | 13.55             | 0.75                   | 2.39                   |
| Total                | 20                 |                   |                        |                        |                 |

Calculated value of F for x and y are greater than that of the table value of F for(2, 18) degrees of freedom. There is a significant difference between the control and the experiment. Based on that the further analysis was worked out for the same separately by using student’s t-test.

Fig. 1. Biogas production in control and experimental bioreactor (cow dung replaced with sewage water)

Table 4. Analysis of variance (ANOVA) for the production of biogas between control and experimental bioreactor
which included the gradual replacement of cow dung with sewage water and gradual replacement of cow dung with poultry droppings and sewage water (Table 6) were recorded. Analysis of variance (ANOVA) for the production of biogas between control and experimental bioreactor of cow dung replacement with poultry droppings and sewage water is incorporated in Table 7. In Table 8, Student’s “t” test analysis of TS, TVS & VFA for biogas production by gradual replacement of cow dung supplemented with poultry droppings with sewage water was recorded and it revealed that they have very significant effect on producing TS, TVS and VFA. The Coefficient of variance analysis report revealed that experimental bioreactor is producing consistent biogas in the both experimental trials.

RESULTS

The study on the replacement of cow dung with sewage water showed high utility of the total solids from its initial level than the control bioreactor (Table 2 & 3). In the experimental bioreactor the total solid degradation occurred from 10% to 6.1%. From this it was observed that there was a significant gradual solid reduction in experimental bioreactor when compared to control.

The analysis of biogas production by gradual replacement of cow dung with poultry droppings and sewage water revealed that the total solids ranged between 7.2% and 6.7%. This indicated that there was a significant solid reduction in experimental bioreactor when compared to control. The utility of solids by microbes in turn indicated the high productivity of biogas. Also high utility of total volatile solids, volatile fatty acids were observed in both experimental bioreactors compared to the control bioreactor respectively. Throughout the experimental period the pH ranged between 7.0 and 7.3. The temperature changes both in control and experimental bioreactors were observed between 36°C and 38°C respectively (Table 6).

The results revealed that the cow dung replaced with sewage water (1421lit/Kg of dry matter/day) gave more biogas production when compared to control bioreactor (1007lit/Kg of

| No of days | Total solids (%) | Total Volatile Solids (%) | Volatile Fatty Acids (%) |
|------------|-----------------|---------------------------|-------------------------|
|            | Control | Experimental | Control | Experimental | Control | Experimental |
| 0          | 7.6     | 6.3           | 62.2    | 65.8           | 42.0    | 52.0           |
| 15         | 7.9     | 6.6           | 64.2    | 66.2           | 44.0    | 50.0           |
| 30         | 7.6     | 6.9           | 66.2    | 66.8           | 40.0    | 58.0           |
| 45         | 7.6     | 6.1           | 64.8    | 64.9           | 43.0    | 48.0           |
| 60         | 7.8     | 6.8           | 62.4    | 72.4           | 40.0    | 48.0           |
| 75         | 6.6     | 7.2           | 63.8    | 68.2           | 40.0    | 46.0           |
| 90         | 6.4     | 7.1           | 64.2    | 68.8           | 42.0    | 52.0           |
| Total      | 51.5    | 47            | 447.8   | 473.5          | 291.0   | 354.0          |
| Mean       | 7.357   | 6.714         | 63.971  | 67.643         | 41.5714 | 50.5714        |
| Variance   | 0.2622  | 4.4459        | 911.905 |                | t=2.179 |                |

Table value of t at 5% level of significance; Degrees of freedom =12;
*There is a significant difference between the control and experimental bioreactor (p<0.05). Therefore the amount of cow dung replacement with sewage water has a very significant effect on producing total solids, total volatile solids and volatile fatty acids.
dry matter/day) (Fig. 1) and the maximum gas production was observed during 80% replacement. The maximum biogas production also occurred in that particular level. During 100% replacement, the amount of biogas production becomes lower which in turn indicated that the importance of organic rigid supportiveness and methanogenesis in the biogas production.

In the replacement of cow dung with poultry droppings supplemented with sewage water work also gave high gas production (1952lit/Kg of dry matter/day) than the control bioreactor (1007lit/Kg of dry matter/day) (Fig. 2). And this is concluded that there is an excellent biogas production biogas production in the 100% replacement of cow dung which in turn indicated that poultry droppings contains more total solid level which can be easily degraded by methanogenic organisms and sewage water can be a good nutrient source for biogas production.

Table 6. Temperature and pH both in control and Experimental bioreactor

| Days of Days | Control bioreactor | Experimental bioreactor | Amount of cow dung replacement with sewage water | Experimental bioreactor | Amount of cow dung replacement with poultry droppings |
|--------------|--------------------|-------------------------|-----------------------------------------------|-------------------------|-----------------------------------------------------|
| Temp.(°C)    | pH                 | Temp.(°C)               | pH                                            | Temp.(°C)               | pH                                                  |
| 0            | 37                 | 37                      | 0%                                            | 36                      | 7.1                                                 | 0%                                                 |
| 15           | 38                 | 38                      | 20%                                           | 37                      | 7.4                                                 | 20%                                                |
| 30           | 37                 | 37                      | 40%                                           | 37                      | 7.3                                                 | 40%                                                |
| 45           | 37                 | 37                      | 60%                                           | 38                      | 7.5                                                 | 60%                                                |
| 60           | 37                 | 37                      | 80%                                           | 37                      | 7.4                                                 | 80%                                                |
| 75           | 36                 | 36                      | 100%                                          | 38                      | 7.6                                                 | 100%                                               |
| 90           | 37                 | 37                      | 100%                                          | 38                      | 7.7                                                 | 100%                                               |

Mean±SD 36±2.1°C for temperature and 7.2±0.2 for pH.
DISCUSSION

The experiment bioreactors produced more biogas than the control. The gradual replacement of cow dung with sewage water gave maximum gas production at 80% replacement. There are a large number of parameters which affect the net energy output from a digester. Retention time will have an effect on the final gas production as well as the total and volatile solids in the feed materials\textsuperscript{27-29}. In the present work the total solids ranged between 6.4% and 7.9% in control and between 6.1% and 7.2% in experimental bioreactor. The total solids value would be about 4% and do not exceed about 9%\textsuperscript{10}. The volatile solids do not exceed above 90% or drop below 40% and 73% could be chosen as a reasonable value. In the present study, the maximum volatile solids were 72.8%, it was observed in 80% replacement of cow dung with sewage water which gave more gas

| No of days | *Total solids (%TS) | *Total Volatile Solids (%TVS) | Volatile Fatty Acids (%VFA) |
|------------|---------------------|---------------------------|-----------------------------|
|            | Control  | Experimental | Control  | Experimental | Control  | Experimental |
| 0          | 7.90     | 7.22         | 64.8     | 74.10         | 42.6     | 52.6          |
| 15         | 7.66     | 7.26         | 65.20    | 74.30         | 46.0     | 53.8          |
| 30         | 7.66     | 6.94         | 65.10    | 73.70         | 46.4     | 52.8          |
| 45         | 7.78     | 6.92         | 65.60    | 73.80         | 43.2     | 52.2          |
| 60         | 7.70     | 6.72         | 64.76    | 75.12         | 44.0     | 54.0          |
| 75         | 7.60     | 6.78         | 67.00    | 75.24         | 43.4     | 53.4          |
| 90         | 7.52     | 6.78         | 67.44    | 75.18         | 43.8     | 52.0          |
| Total      | 53.82    | 48.62        | 459.9    | 521.44        | 309.4    | 370.8         |
| Mean       | 7.689    | 6.946        | 65.7     | 74.49         | 44.2     | 52.971        |

Variance: 0.0310, 0.8129, 134.2857
Standard deviation: 0.1760, 0.9016, 11.5882
T: t=7.8959, t=18.2402, t=14.16

Table value of t at 5% level of significance; Degrees of freedom =12;
*There is a significant difference between the control and experimental bioreactor (p<0.05). Therefore the amount of cow dung replacement with sewage water has a very significant effect on producing total solids, total volatile solids and volatile fatty acids.

Table 7. Analysis of variance (ANOVA) for the production of biogas between control and experimental bioreactor

Table 8. Student’s "t" test analysis of TS,TVS&VFA for biogas production by gradual reduction of cow dung supplemented with poultry droppings with sewage water

Ho: There is significant difference between the production of TS,TVS and VFA in the control and in the experiment
production than other reduction concentration. The influence of temperature on the methanogenic bacterial activity, which inhibit the biodegradation and stabilization efficiency of substrates in bioreactor\textsuperscript{31}. In the present work also the mesophilic temperature was provided throughout the experimental period and optimum gas yield was also obtained from this temperature.

The temperature fluctuation showed that the bio gas production almost stop and total VFA, such as acetate and propionate are rapidly accumulated, accompanied by the fall in \( \text{pH} \). Temperature fall not only affected the methanogenesis but also the hydrolysis and acidification\textsuperscript{32}. The maximum gas production occurred in the study of the cattle waste when the total solids are completely degraded. It was true because in the present study, the maximum volatilization of total solid biodegradation occurred during 80\% replacement of cow dung by sewage water.

Among the various levels of solid concentration, total solid at 8\% registered a higher biogas production or productivity followed by total solid at 6\%\textsuperscript{33}. In the cow dung replacement with poultry droppings and sewage water study, the reduction of solid occurred up to 6.72\% The gas production was found to be maximum in 100\% replacement of cow dung with sewage water treated poultry droppings sample which was also in accordance the results of Bonmati et al., (2001)\textsuperscript{34}. Many series of experiments for biogas production using cattle, poultry and sewage sludge separately and combinations\textsuperscript{35}. Like that in this work, cow dung mixed with poultry droppings supplemented with sewage water gave comparatively maximum biogas yield than control which was loaded with cow dung and ordinary water. Among the various physical parameters, \( \text{pH} \) is the one which highly influences the microbial activity\textsuperscript{36}. The optimum \( \text{pH} \) for biomethanation ranged between 6.5 and 7.7\textsuperscript{37}. The results of the present finding of the experimental bioreactor also depicted the similar results with the maximum gas yield being around neutral \( \text{pH} \) (Table 8).

CONCLUSIONS

From this study it is well observed that the sewage water which is considered to be the chief source of contamination used as a source of nutrient along with cow dung and poultry droppings for biogas production. Because these animals excrete leads to many problems like odour nuisance, fly nuisance apart from causing serious problems like eutrophication. These substrates act as an alternative renewable energy sources for conventional energy and economically more feasible. After biogas production, the slurry from the biogas plant used as bioorganic fertilizer. Conclusively recycling of these waste materials is necessary to prevent pollution and to conserve natural resources.

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DATA AVAILABILITY

All datasets and statistical report analyses during this study are included in the manuscript.

ETHICS STATEMENT

This article does not contain any studies with human participants or animals.

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