Tarım Makinaları Bilimi Dergisi (Journal of Agricultural Machinery Science)
2008, 4 (3), 269 - 272

Comparisons of the Methane Production Yield Coefficient from Anaerobic Microbial Assemblages in the Anaerobic Treatment of Wastewater

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Abstract: Mixed-microbial assemblages from a septic tank, leachate, sediment samples from a coastal area, sludge from the digester of a brewery wastewater treatment plant and acidic sulfate soil samples were compared on the basis of methane yield coefficient under anaerobic condition. The biomass inoculum of all these mixed culture was started up at ambient temperature (30±1^{0}C) by a synthetic glucose-based substrate in a semi-continuous-flow mode. These acclimated cultures were used as the parent culture in this study. Five batch reactors containing different kinds of these mixed cultures at the same content were fed with the glucose-based synthetic waste of 20000 mg COD/L for each reactor. Experiments were performed for around 2 month period with initial parameters of pH 7 and at room temperature of 30°C. The levels of COD and methane were followed through the experiment. The biogas production due to these cultures per a gram of organic waste removed was evaluated. The methane yield coefficient of various cultures was 317, 315, 226, 324 and 6.5 ml (at STP)/gram of COD removed for mixed-microbial assemblages from a septic tank, leachate, sediment samples from a coastal area, sludge from the digester of a brewery wastewater treatment plant and acidic sulfate soil samples, respectively. Bacteria originated from a brewery wastewater treatment plant showed the highest CH_4 production yield coefficient of 324 ml (at STP)/gram of COD removed. Anaerobic bacteria originated from a brewery wastewater treatment plant was found to be the most appropriate culture for the treatment of wastewater due to their methane gas production yield coefficient per gram of organic waste removed.

Key words: Anaerobic digestion, Gas production, Methane, Mixed culture, Wastewater treatment

INTRODUCTION and LITERATURE REVIEW

The current approach to biological treatment of wastewater relies primarily on the use of bacterial cultures. Many industrial waste treatment processes favor anaerobic processes because they usually require fewer resources. The use of appropriate microorganism combinations that for the biological treatment of wastewater can be a cost-effective process for treating waste streams. The main contributions of anaerobic bacteria to the integrated process are the structural transformation of organic waste and the production of methane. Major environmental problems are encountered with organic wastewaters such as those from beverage industries, edible oil and potato starch processing, as well as pulp and paper manufacturers (Yoda *et al.*,1987; Rinzema and Lettinga, 1988). In order to achieve these wastewater management advantages, the selection of appropriate bacteria that are able reduce organic substrate and produce methane gas effectively is a prerequisite. Therefore, the goal of the project was to find the most effective anaerobic mixed culture by comparing the methane yield coefficient of different mixed cultures obtained from different sources. Knowledge of the amount of methane produced by different mixed cultures will provide an advantage in biotechnological applications.

MATERIAL and METHOD

Mixed-microbial assemblages which were obtained from five different locations, i.e., mixed liquor in a septic tank, leachate, sediment samples from a coastal area at Pattaya (Chonburi, Thailand), sludge from the digester of a brewery wastewater treatment plant and that from acidic sulfate soil samples (Ongkarak, Nakornnayok Province, Thailand) were used as seed. A controlled environment was needed to ensure that the microorganisms had a proper medium in which to grow and that there could be comparability among reactors. Environmental conditions including pH, temperature regulation, nutrient and trace-element addition, initial loading factor (COD/biomass), anaerobic conditions, and proper mixing were carefully controlled.

Bioreactors

Five reactors were constructed from 6-liter capacity PET bottles. Each was equipped with two outlet ports, one for liquid sample withdrawal and the other for gas venting. The reactor was connected to a gas collection system, which was based on water displacement by the exiting gases (Figure 1).

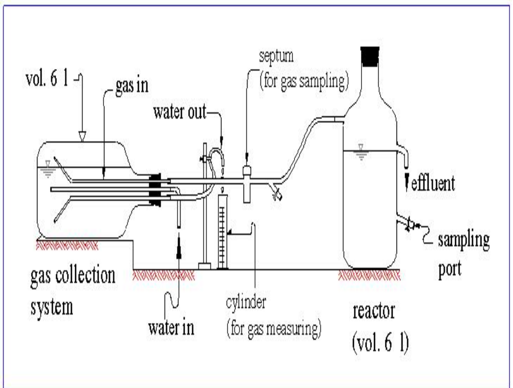

Figure 1. Bioreactor for the studies of biogas production and COD removal.

Operation

A glucose composition consisting of 20000 mgCOD/L and sufficient inorganics was used as the synthetic waste to maximize consistency for bacterial cell growth. The components followed those of Leighton and Forster (1997). The control conditions in the operation were: initial concentration of glucose based solution (20000 mg COD/L), initial content of mixed liquor volatile suspended solids (1000 mg MLVSS/L), pH (7), temperature (30±1^{0}C), and working volume (5 L).

The biomass inoculum was started up in 2-liter PET bottles that were maintained at ambient temperature (30±1^{0}C). The synthetic glucose-based substrate was gradually increased in concentration over a range from 2000-10000 mg/L in a semi-continuous-flow mode. The biogas produced was observed in order to monitor the performance of the bacterial growth. Final content of biomass (MLVSS) were determined in order to start the operation at the same content for each reactor.

The 5 batch reactors (Figure 1) containing different kinds of mixed-microbial were fed with the glucose-based solution. The batch reactions were operated for 2 months. Biogas was measured every 3 to 6 hours and collected to determine the amount of methane (CH_4) by a Shimadzu GC-14B GC (TCD). At selected times (10-15 days), liquid samples (5 ml) were withdrawn and analyzed for COD according to the procedure of Standard Method (APHA et al., 1992). The glucose synthetic waste utilization and methane production from the experiments were followed. The outcome of the bacterial work that showed the potential of the mixed bacterial groups from five different sources in producing methane from waste utilized was examined.

RESULTS and DISCUSSION

Regarding to biogas evolution, the gas occurred after 2 days of operation. The percentage of CH_4 was gradually increased but with differences by the bacterial culture in each reactor. During an experiment, the evolution of the CH_4 was followed.

We can represent product production as a conversion of a number of waste, dP/dS. Described formalism is useful as a first step in biotechnological studies. It gives estimate how much waste should be supplied to reactor to obtain required amount of target product. Where dP is the increase in product production consequent on utilization of the amount dS of waste, and dP and dS are respective infinitely small increments. It should be noticed that rigorous definition of $Y_{P/S}$ as derivative dP/dS stems from the fact that the CH_4 production yield coefficient ($Y_{P/S}$) can vary in time, the negative sign being introduced because of P and S vary in opposite senses. It expresses explicitly the waste utilization for product: how much mass units of particular waste should be consumed to produce one unit volume of CH_4. Computation table to determine the coefficients $Y_{P/S}$ was set (Data not shown) and the plot was shown in Figures 2-6.

A plot of the cumulative volume of CH_4 against the S utilized, (S_o-S_t), is a straight line of slope coinciding with the CH_4 production yield coefficient ($Y_{P/S}$). The CH_4 production yield coefficient is defined as the ratio of CH_4 produced in the experiment to the waste utilized. From this study, a slope of 317 (septic liquor), 315 (leachate), 226 (coastal sediment), 324 (brewery waste water treatment plant) and 6.5 (acidic sulfate soil), expressed in ml (at STP) per gram of biodegradable waste utilized, was found. Estimates of CH_4 production yield coefficient, $Y_{P/S}$, were compared for all selected types of bacteria, which exhibited a different yield.

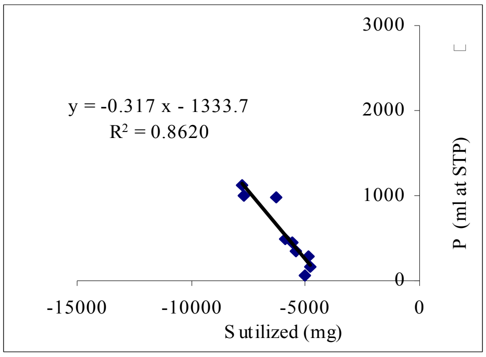

Figure 2. Graphical determination of the methane yield coefficient of mixed-bacteria from a septic tank.

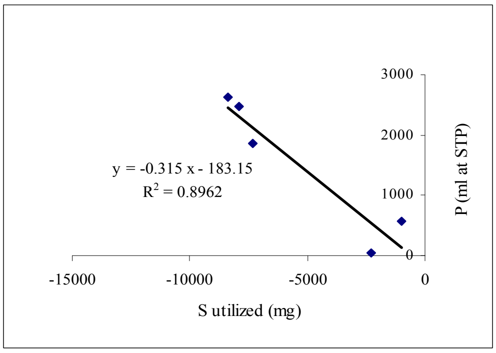

Figure 3. Graphical determination of the methane yield coefficient of mixed-bacteria from leachate.

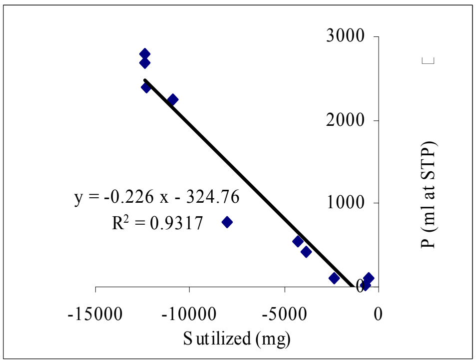

Figure 4. Graphical determination of the methane yield coefficient of mixed-bacteria from coastal sediment.

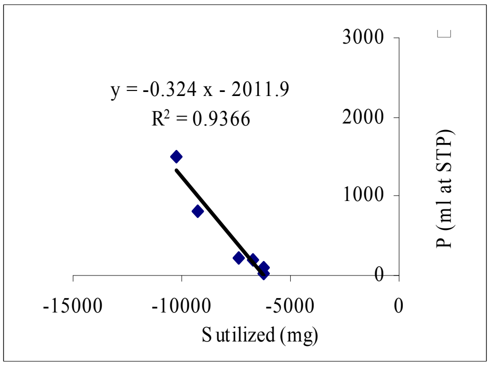

Figure 5. Graphical determination of the methane yield coefficient of mixed-bacteria from sludge from a brewery wastewater treatment plant.

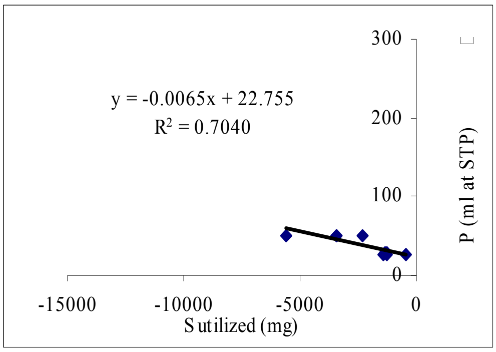

Figure 6. Graphical determination of the methane yield coefficient of mixed-bacteria from acidic soil sample.

These data show that sludge from the brewery wastewater treatment plant exhibited the highest value of CH_4 production yield coefficient, $Y_{P/S}$. This shows the highest performance of this mixed-bacterial culture to degrade environmental pollutant and yield the highest amount of CH_4. The CH_4 production yield coefficient, $Y_{P/S}$, of mixed cultures from a septic tank and leachate was comparable while that of cultures from acidic sulfate soil was much lower than all selected cultures. The theoretical amount of CH_4 of 350 ml at STP per a gram of glucose utilized is reported. (Tchobanoglous and Burton, 1991). However, it was observed from the experiments (Figures 2 and 5) involving the reactors that contained the bacterial cultures from a septic tank and from a brewery wastewater treatment plant, that the materials responsible for the COD had been initially used in some other biological activities (such as biomass or CO_2 production) before it began to be used to produce CH_4. On the other hand, the reactor that contained the bacterial culture from leachate (Figure 3), coastal sediment (Figure 4), and from acidic sulfate soil (Figure 6) produced CH_4 from the COD material from the beginning. The values of CH_4 production yield coefficient, $Y_{P/S}$, obtained from this study were summarized in Table 1.

Table 1. A summary of methane yield coefficient from various anaerobic mixed assemblages.

Bacterial Sources	**CH_4 yield coefficient, $Y_{P/S}$,** (ml CH_4 produced at STP / gram of glucose synthetic waste utilized)
1. Septic liquor	317
2. Leachate	315
3. Coastal sediment	226
4. Brewery wastewater treatment plant	324
5. Acidic sulfate soil	6.5

CONCLUSION

Sludge from the brewery wastewater treatment plant was found to be the most appropriate bacterial assemblages due to its ability to be used for application in anaerobic treatment. The properties in methane production in simultaneous lowering of COD make this culture an excellent candidate for technical application. The present investigation confirms the importance of the selection of the best populations that are able to effectively degrade environmental pollutants in order to produce the highest amount of methane.

ACKNOWLEDGEMENTS

This research was supported financially by Kasetsart University Research and Development Institute (KURDI), Kasetsart University, Bangkok, Thailand.

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