Poll Res. 39 (4) : 1034-1037 (2020)

ISSN 0257–8050

BIOMETHANATION OF HIGH SOLID CONTAINING DISTILLERY SPENTWASH USING DEVELOPED ACCLIMATIZED MICROBIAL CONSORTIA

RAGHUNATH VISHNU BURASE[1]*, SANJAY VASANTRAO PATIL[1] AND RAJENDRA D. JOSHI[2]

[1] *Department of Alcohol Technology and Biofuels, Vasantdada Sugar Institute, Manjari (Bk.), Tal. Haveli, Dist. Pune 412 307, Maharashtra, India*

[2] *Department of Microbiology, Yogeshwari Mahavidyalaya, Ambejogai 431 517, Dist. Beed, Maharashtra, India*

(Received 2 March, 2020; Accepted 12 April, 2020)

ABSTRACT

Biomethanation of spent wash (SW) is now a well established technology in distilleries. At present in distilleries average COD reduction could be achieved around 60-65% for SW having COD in the range of 100000-130000 mg/L. But SW obtained from integrated evaporation system has COD 230000-250000 mg/L. However, biomethanation of concentrated SW (20-30 °brix) is problematic due to relatively poorer digestion efficiency and intolerance of consortia of microbes to some inhibitors. The objective of the current study is to understand the acclimatization of methanogenic consortia to concentrated SW. The sludge containing methanogenic bacterial consortia from biogas plant was used for acclimatization. The developed acclimatized consortia of methanogenic bacteria were used for biomethanation of concentrated SW. The performance of developed bacterial consortia for the production of methane was evaluated on bench scale (10 l). This study successfully demonstrates the application of acclimatized consortia of methanogenic bacteria for operation on bench scale with enhanced COD reduction in the range of 64 to 74% using 25 to 30 °brix SW. The control biodigester using un-acclimatized inoculum at 30°brix SW showed maximum 43% COD reduction and less gas production compared to experiment.

KEY WORDS : Biomethanation, Biodigester, COD reduction, Spentwash.

INTRODUCTION

Distilleries are considered as one of the tremendously polluting industries worldwide (Kim and Ihm, 2010). Hence Central Pollution Control Board, Government of India has decided distilleries as one of the major polluting industries (CPCB, 2003). Indian distilleries mainly use sugarcane molasses as a preferred substrate for alcohol production due to its easy availability. Distilleries utilize huge quantity of fresh water and create large volumes of spent wash (SW) which is highly polluted. Biomethanation is one of the preferred technologies used for SW treatment and a process utilized worldwide through anaerobic digestion (AD) for reduction of organic waste and production of biogas. A dark brown colored SW has pH 4.0-4.3; COD 100000-130000 mg/L and SW obtained from integrated evaporation system having COD in the range of 230000-250000 mg/L. Also SW contains high concentration of inorganics such as chlorides, sulfates, phosphates, sodium, potassium and calcium. At present, the performance of biogas plants in the distilleries shows an average COD reduction of 60-65% for SW having COD 100000 - 130000 mg/L. Recently integrated evaporation has emerged as a potential technology for reducing the SW volume from 14-15 l/l to 3.0-3.5 l/L of alcohol produced (PSA, 2014). Therefore, it is necessary to develop biomethanation process that can accept such high strength waste. There is a scope to develop biomethanation process to treat concentrated SW.

Microbes have a great capacity of adaptation to

the environmental adverse conditions. Hence the strategy of acclimatization was used for development of bacterial consortia responsible for biomethanation in the current research studies. It is reported that using swine waste, COD reduction was achieved by adapting microbial community to the continuing increase of total ammonium nitrogen (Esquivel-Elizonda, 2016).

The aim of this study was to operate biomethanation of high solid containing SW using acclimatized methanogenic bacterial consortia developed during this research work. Performance of developed bacterial consortia was evaluated on bench scale (10 l). Control experiment was also conducted using un-acclimatized consortia as inoculum.

MATERIALS AND METHODS

Acclimatization of bacterial consortia

The sludge containing the bacterial consortia was collected aseptically in a sterile container from the bottom of the biodigester of Pandurang Sahakari Sakhar Karkhana Ltd. Solapur, Maharashtra, India operating feed SW of 20°brix. The sludge of 0.270 l was acclimatized in a 10 l capacity biodigester by feeding 100 mL sterile SW (pH 7.0) per day starting from 21°brix up to 30°brix. After every ten days, concentration of SW was increased by 1°brix. Acclimatization was conducted at 37 ± 0.5°C anaerobically (Westerholm *et al.*, 2011).

Bench scale biodigester experiments using acclimatized and un-acclimatized inoculum

1. A wide mouth glass bottle of 10 L capacity with a working volume of 9 l was used as the biodigester. The gas was measured using water displacement method by monitoring water level displaced in the cylinder twice in a day and average value was considered to quantify the gas (Pham *et al.*, 2013). Three different experiment sets were conducted using acclimatized and un-acclimatized consortia on bench scale. The experiment sets A, B and C were performed using SW of 15, 20 and 30°brix respectively. The developed consortium of microbes (30%, v/v) was fed to the experiment biodigester as inoculum for set C only (Thiyagu and Sivarajan, 2018). The biomethanation was operated at 37 °C, correspondingly hydraulic retention time was of 24 days (Metcalf and Eddy, Inc. 1972). Hence 0.375 l SW of respective solid content was fed daily to the biodigester via peristaltic pump. The content of biodigester was mixed thoroughly at an interval of 6 hours at 150 rpm for 10 minutes magnetic stirring. The desired concentration of SW was obtained either by dilution with distilled water or concentrated by heating at controlled temperature. pH of SW was maintained close to 7 by using sodium bicarbonate as a buffer during first 24 days. After the process got stabilized, SW was fed without pH adjustment.

After the stabilization of biodigesters, the feed concentration of SW and organic loading rate was gradually increased per 3 days by 1°brix. In case of A, B and C sets, initial COD of SW were 51000 mg/L. The final feed COD of SW in case of A was 125000 mg/L, B was 181000 mg/L and C was 242000 mg/L. The overflow of the biodigester was collected for analysis of various parameters. Control biodigester was inoculated using sludge of biogas plant (un-acclimatized inoculum). The feeding strategy of SW and experimental conditions were same for experiment set.

Analytical methods

1. The analytical methods used in order to monitor the performance of the biodigester, were performed using the methods given in the Standard Methods for the Examination of Water and Wastewater (Laura, 2012). COD digester (2015M, Spectralab, India) and autotitrator (CT15 Spectralab, India) used for estimation of COD.

RESULTS AND DISCUSSION

Acclimatization of bacterial consortia

The developed consortia of acclimatized methanogenic bacteria were adjusted to the environment of SW. Initially the volatile fatty acid (VFA) concentration was more, that indicates VFAs are produced by acid forming bacteria which may be utilized by acclimatized methanogens in a suitable mode. Accordingly stable pH range of 6.90-7.2 which is optimum for AD was developed. Therefore, inoculation of biodigester with developed acclimatized microbial consortia may enhance biodigester stability.

1. Structure and composition of *Archaea* may be changed after acclimatization. Acetoclastic and hydrogenotrophic methanogens play key roles in methane generation (Godina *et al.*, 2012).

Ultimately methane content of experiment biodigester was enhanced.

Performance of bench scale biodigesters

Set A: Lab scale trial using un-acclimatized consortia using 15°SW

The biodigester pH during initial 10 days was 4.30-4.50, afterwards increased to 6.85 within 18 days and reached its steady state values 7.10-7.20 within 35 to 45 days. At feed COD of 125000 mg/L, 60% COD reduction was achieved. On bench scale using 15°brix SW, maximum gas generation of 13.2 l/day was achieved after 45 days of biomethanation.

Part B: Lab scale biodigetser trial using un-acclimatized consortium using 20°SW

At feed COD of 181000 mg/L, the COD reduction achieved was 64%. Using 20°SW, maximum gas generation of 20 l/day was achieved within 60 days. It was confirmed that the un-acclimatized consortia was able to tolerate shock up to 20°SW.

Part C: Lab scale biodigetser trial using acclimatized consortia and un-acclimatized consortia for concentrated 30°SW

Performance of bench scale control biodigester using 30°SW

Bench scale control biodigester also showed consistent COD removal of above 60% after 45 to 65 days. However, as the concentration of feed SW increased from 21°brix, the biodigester started to disturb after 66 days. This may be due to sensitivity of microbial consortium to various inorganics of concentrated SW. VFA to alkalinity was raised from 0.06 to 0.17. However, in control biodigetser at feed concentration above 21°brix started to increase VFA from 1230 mg/L to 3150 mg/L at feed of 30°SW. COD reduction was noticed by the disturbance in the biodigetser (from 64% to 45%, maximum gas generation of 21.5 l/day) with increase in VFA concentration from 17200 mg/L to 18200 mg/L. These results are contradictory to the experiment biodigester.

Performance of bench scale experiment biodigester

The concentration of alkalinity was 9900 mg/L after 25 days and gradually increased to 17500 mg/L within 85 days. The alkalinity was mainly increased due to the microbial biomass which had to be acclimatized. VFA to alkalinity ratio was 0.33 after about 25 days. Bench scale experiment biodigester showed steady state condition with consistent COD removal indicating successful acclimatization of methanogens to concentrated SW.

Rate of COD reduction was more than 60% after 45 days and reached maximum to 64% at feed COD of 242000 mg/L with actual gas generation of 28 l/day (theoretical gas generation-28.3 l/day).

CONCLUSION

Based on the bench scale studies, it can be concluded that the developed acclimatized consortia of methanogenic microorganisms is very effectively useful for utilization of concentrated SW. On bench scale, the control biodigester for feed SW of 20 to 30°brix shows maximum 43% COD reduction against 64% in experiment biodigester. As compared to experiment biodigester, the biomethanation process of control biodigester was disturbed and less gas was produced.

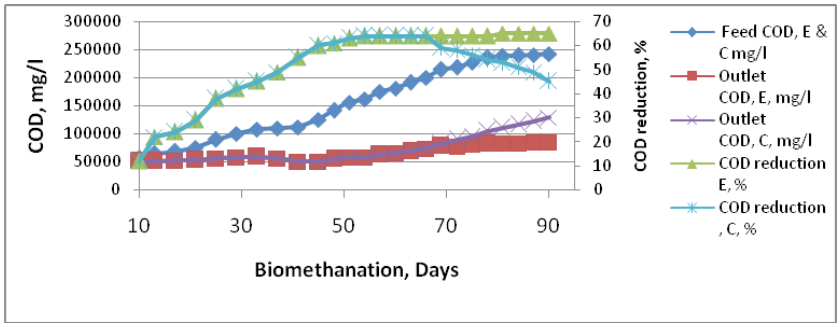
ACKNOWLEDGEMENTS

This work is the result of Ph. D. work of the first author. The authors are thankful to the Director General, VSI for providing the necessary facilities during the studies of this research.

REFERENCES

CPCB, Annual Report, Central Pollution Control Board, New Delhi, 2003.

Esquivel-Elizonda, S., Parameswaran, P., Delgado, A.G.,



Maldonado, J., Bruce, E. and Brown, E.R.K. 2016. Archaea and bacteria acclimate to high total ammonia in a methanogenic reactor treating swine waste, *Archaea :* Pp 1-10.DOI: doi.org/10.1155/2016/4089684

Godina, A., McLaughlinb, J. W., Websterc, K.L. and Packalen, M. 2012. Methane and methanogen community dynamics across a boreal peatland nutrient gradient. *Soil Biol. Biochem*. 48 : 96-105. DOI: http://dx.doi.org/10.1016/jsoilbio.2012.01.018.

Kim, K.H. and Ihm, S.K. 2010. Heterogeneous catalytic wet air oxidation of refractory organic pollutants in industrial wastewaters: a review. *J. Hazard. Mater*. 186 : 16-34.

Laura, P. 2012. *Standard Methods for the Examination of Water And Wastewater*, 22nd edition, American Public Health Association, Washington.

Metcalf & Eddy, Inc. 1972. *Waste Water Engineering*. McGraw-Hill, New York, N.Y. p. 605.

PSA- A report on opportunities for green chemistry initiatives: Molasses based distilleries, 2014. Office of Principal Scientific Adviser to the GoI, Vigyan bhawan annexe, New Delhi, 2014.

Pham, C.H., Triolo, J.M., Cu, T.T.T., Pedersen, L. and Sommer, S.G. 2013. Validation and recommendation of methods to measure biogas production potential of animal manure. *Asian Australas. J. Anim. Sci*. 26 (6) : 864-873. DOI: https:/doi.org/10.5713/ajas.2012.12623

Thiyagu, R. and Sivarajan, P. 2018. Influence of organic loading rate in acclimatization phase of hybrid upflow anaerobic sludge blanket (UASB) reactor treating distillery spentwash, *Nature Environmental and Pollution Technology.* 17: 223-227.

Westerholm, M., Moller, B., Arthurson, V. and Schnurer, A. 2011. Changes in the acetogenic population in a mesophilic anaerobic digester in response to increasing ammonia concentration. *Microbes Environ*. 26 (4) : 347-353. doi: 10.1264/jsme2.ME11123.