

Abstract

In the present world, the energy demand is increasing due to rapid industrialization and urbanization. The non-judicious use of fossil fuels has dented the environment by escalating global warming. In order to develop clean energy, a holistic approach is quintessential towards achieving feedstock, environmental and economic sustainability. Present study embarks the idea of developing biohythane production from lignocellulosic wastes for gaseous energy generation with simultaneous bioremediation. Lignocellulosic wastes are widely available, renewable and non-toxic. Moreover, efficient and cost effective production of biohythane from lignocellulosic wastes depends on the selection of a suitable pretreatment technique and the type of inoculum. Pretreatment steps are required to reduce the lignin content and decrease the recalcitrant structure of lignocellulosic biomass. Acidogenic mixed consortia was developed from cow dung after various inoculum pretreatments. Experiments were design to enumerate the variation in hydrogen production using different pretreated inoculum. The heat shock pretreatment (100 °C, 10 min) showed significant improvement in H₂ production. The process was validated using modified Gompertz model ($R^2 > 0.99$). The suitable pH, temperature, YE concentration and C/N ratio were determined in batch system. At suitable physico-chemical parameters (pH 6.5, 37 °C, YE 3 g/L and C/N ratio 26), the maximum cumulative H₂ production and yield of 3220 ± 40 mL/L and 2.6 ± 0.05 mol/mol glucose, respectively were observed. Statistical analysis using Taguchi design showed that C/N ratio had the most influential parameter towards H₂ production. The PCR-DGGE analysis indicated the dominance of *Clostridium* sp. in the mixed culture. The maximum methane production and yield of 1641 ± 36 mL/L and 1.86 ± 0.04 mol/mol acetic acid were observed using methanogenic mixed consortia at suitable process conditions (pH 7.5, 35 °C and A/B ratio of 2:1). Different lignocellulosic wastes viz. sugarcane bagasse, sugarcane top and water hyacinth were explored as feedstock for biohythane production. Chemical and biological pretreatment were used to reduce the lignin content of sugarcane bagasse and sugarcane top. The maximum lignin removal of 89 ± 3.5 % (w/w), 74.6 ± 1.7 % (w/w) and 60.4 ± 2.3 % (w/w) were observed using alkaline hydrogen peroxide (NaOH 1 % w/v, H₂O₂ 2.5 % v/v, 50 °C, 150 min), sodium hydroxide (0.5N, 50 °C, 60 min), and *Pleurotus pulmonarium* MTCC 1805 (21 d, SSF), respectively. Various analytical techniques viz. SEM, XRD, FTIR and confocal microscopy were used to confirm the removal of lignin. Further, potentiality of pretreated lignocellulosic biomass was evaluated towards biohythane production. The present study emphasised that, pretreated sugarcane bagasse and sugarcane top gave maximum hydrogen yield of 93.4 ± 2.2 mL/g-VS and 84 ± 5.4 mL/g-VS, respectively in batch process. The overall gaseous energy recovery as well as substrate conversion efficiency were improved using spent medium of dark fermentation for 2nd stage biomethanation process. Water hyacinth leaves are rich in proteins and microelements and was found suitable for the biohythane production. To achieve the higher rate of hydrogen and methane production, continuous biohythane production was studied in pack bed (H₂ stage) and continuous stirrer tank reactor (CH₄ stage). The maximum hydrogen and methane production of 15 ± 2.7 g/kg COD and 195 ± 10 g /kg COD, respectively were observed with COD removal of 89 %. This improved the total gaseous energy recovery (62 %). The material and COD analysis of biohythane production indicate the potentiality of the process for the gaseous energy recovery from lignocellulosic wastes.

Keywords: biohydrogen; biomethane; biohythane; dark fermentation; spent medium; mixed culture; lignocellulosic waste; pretreatment; medium optimization; Taguchi design; gaseous energy; material analysis.