Abstract

Global warming and climate change are the major issues faced by the current world. Due to rapid urbanization and population explosion, environmental pollution has amplified catastrophically. Moreover, the depletion of fossil fuel propagated the quest for a cleaner and sustainable alternative energy sources. Hydrogen being the cleanest fuel having the highest calorific value per unit mass as compared to other fuels could be the sustainable energy carrier. Dark fermentation process promises a viable route for biohydrogen production. However, the low substrate conversion efficiency hinders its commercial feasibility. Butanol is also a promising biofuel having better fuel characteristics as compared to bioethanol. Thus, present dissertation focuses on efficient concomitant hydrogen and n-butanol production from organic waste to enhance the net substrate energy recovery. A hyper solvent and hydrogen producer, *Clostridium saccharoperbutylacetonicum* has been used for the hydrogen and butanol production from biphasic fermentation in the present study. The impact of various process parameters such as pH, temperature, inoculum age, inoculum size, carbon source and nitrogen source has been comprehensively explored in batch process. Process parameters optimization revealed that pH of 6.5, temperature of 37 ˚C inoculum age of 4 h and inoculum size of 7 % (v/v) were suitable for obtaining maximum H_2 and butanol yields by *C. saccharoperbutylacetonicum*. Central composite design was employed and analysed using response surface methodology for the enhancement of H_2 and butanol production. Highest H_2 and butanol production of 3750 mL L^{-1} and 2.8 g L^{-1}, respectively were observed at optimized parameters with yield of 275 mL g^{-1} glucose and 0.2 g g^{-1} glucose, respectively. Starch and xylan were observed to be preferred carbon sources suggesting effective utilization of C_6 and C_5 sugars. The maximum H_2 yield of 264.3 mL g^{-1} starch and 216 mL g^{-1} xylan and butanol yield of 0.27 g g^{-1} starch and 0.24 g g^{-1} xylan were obtained with overall energy recovery of 85.61 % (starch) and 75.22 % (xylan), respectively. Overall carbon and energy balance has been performed and it asserted that most of the carbon was channelled towards butanol formation during the biphasic fermentation process. Butyric acid is one of the major byproducts of dark fermentation that affects butanol production. Hence, effect of butyric acid addition was also studied on hydrogen and butanol production using model substrate (glucose). Maximum H_2 and butanol production of 5.2 L L^{-1} and 5.5 g L^{-1} were obtained at 5 g L^{-1} butyric acid addition. This work indicates the potency of bi-phasic fermentation to boost energy recovery from starch/xylan based feedstock. For the practical realization of the process, utilization of organic residues in place of pure substrate is quintessential. Thus, co-digestion of organic wastewater such as cane molasses (CM), distillery effluent (DE) and starchy wastewater (SWW) and nitrogenous residues have been considered for H_2 and n-butanol production. The addition of co-substrate to organic wastewater has resulted in 12.2, 1.1 and 6.6 folds increase in hydrogen production in comparison to CM, DE and SWW, respectively as the sole substrate. Maximum energy conversion efficiency and positive net energy gain of 50.16 % and 7.29 kJ g^{-1} COD, respectively were achieved using SWW codigested with water hyacinth (WH). Amalgamation of

SWW and WH was found to be more suitable for H_2 and butanol production. To achieve disruption of complex structure of water hyacinth, different pretreatment methods were applied. Acid pretreatment of WH leads to higher H_2 and butanol production of 3160 mL L^{-1} and 4.5 g L^{-1}, respectively with 89 % energy conversion efficiency. To further enhance the H_2 and butanol productivity, continuous mode of fermentation was employed in suspended cell system as well as whole-cell immobilized system. Packed bed reactor showed higher H_2 and butanol productivity of 1900 mL L^{-1} h^{-1} and 5.04 g L^{-1} h^{-1}, respectively. This study shows the suitability of SWW with acid treated WH as feedstock for sustainable and efficient bioenergy generation with an additional advantage of waste management. The approach of the present study could escalate energy recovery from organic waste and helps in the development of decentralized bioenergy generation process.

Keywords: *Clostridium saccharoperbutylacetonicum*; biphasic fermentation; hydrogen; butanol; organic waste; packed bed reactor; energy conversion efficiency; net energy gain