Abstract

The soaring energy demand and the consequent emissions of greenhouse gases are encouraging researchers to consider H_2 as an alternative renewable energy source. Biological H_2 production processes are environment friendly and renewable. The present dissertation focuses on the evaluation of potential of dark fermentative H_2 production using a lab isolate, *Klebsiella pneumoniae* IIT-BT 08. Effect of different nutritional requirements such as nitrogen source, carbon source, metal ions, and vitamins were thoroughly investigated in the batch mode of operation. Among the various organic and inorganic nitrogen sources, yeast extract and di-ammonium phosphate supplements showed higher H_2 yield of 1.91 ± 0.06 and 1.68 ± 0.08 mol H_2 mol^{-1} glucose, respectively. Plackett–Burman design, the path of steepest ascent and Box-Behnken design were employed to study the effects of different metal ions and vitamins for H_2 production. A total cumulative H_2 production of 1810 ± 25 mL L^{-1} and a yield of 2.05 ± 0.05 mol H_2 mol^{-1} glucose was achieved using the optimized medium. Different organic wastes viz., cane molasses (CM), distillery effluent (DE), and starchy wastewater (SWW) have been explored as feedstock for H_2 production rather than pure substrates. Among different deoiled cakes, groundnut deoiled cake (GDOC) was found to be most suitable as both sole substrate and co-substrate for H_2 production. Using GDOC as supplement has increased the cumulative H_2 production by 1.06, 1.16, and 4.30 folds as compared to non-supplemented CM, DE, and SWW, respectively. To achieve a higher rate of H_2 production, different operational strategies have also been attempted. In case of regulated and intermittent adjustment of pH, a maximum H_2 yield of 15.8 ± 0.32 and 13.6 ± 0.28 mol H_2 kg^{-1} $COD_{removed}$, respectively was observed. The maximum H_2 production rate reached 2.7 ± 32 L L^{-1} h^{-1} at 0.6 h^{-1} dilution rate in a 20 L packed bed reactor. A study was conducted in 10 m^3 reactor to explore the feasibility of bioH_2 production process in pilot scale using co-digestion of CM (1% w/v) and GDOC (2.5% w/v). Cumulative H_2 production and energy recovery of 76.2 ± 2.5 m^3 and 37.9% were achieved, respectively from the 10 m^3 reactor. Thus, the present study indicates the possibility of commercial exploitation of dark fermentative H_2 production technology with dual benefit of bioremediation and green energy generation.

Keywords: *Klebsiella pneumoniae* IIT-BT 08; biohydrogen; dark fermentation; organic waste; groundnut deoiled cake; packed bed reactor; pilot scale.