

ABSTRACT

In recent year, there has been an increasing interest in biohydrogen production. This is mainly due to growing global-scale environment concerns and energy insecurity. Being a sustainable and clean energy source, hydrogen is a promising alternative to fossil fuel which can be produced from a variety of renewable feed stock, such as derived sugar or other types of organic waste.

The Anaerobic Biohydrogen production by *Clostridium acetobutylicum* was studied in batch system using different substrates such as Cane molasses, Cane Juice, Corn Steep Liquor and Starch. Cane molasses was found to be the most suitable substrate. The fermentation conditions were optimized in test tubes and 500 ml autoclavable stoppered serum bottle. The optimized fermentation medium consisted of: (g/L) Cane molasses: 120.0 (6% sugar); $(\text{NH}_4)_2\text{SO}_4$: 3.0; Superphosphate, 0.7 MnCl_2 , 1.2 and CaCO_3 ; 3.0. The vegetative cell of *Clostridium acetobutylicum* was used as inoculum. The fermentation was carried out at strict anaerobic and aseptic conditions for 60 hours. The batch fermentation methodology was adopted and performance of culture was satisfactory. A temperature of 32 ± 1 °C was shown to be optimal for the hydrogen production. The work showed that there is a positive correlation of activity of *Clostridium acetobutylicum* and hydrogen production.