

Optimization of culture condition for hydrogen production by *Clostridium butyricum* CWBI1009

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The dependence on fossil fuels as our primary energy source is a significant cause of global warming, environmental degradation, and health problems. Hydrogen (H₂) is a promising energy vector for the future since CO₂ is not released during its combustion. At present hydrogen is produced by chemical methods, such as steam reforming or partial oxidation of fossil fuels, which involve the release of large quantities of greenhouse gases into the atmosphere. Biological hydrogen production by "dark-fermentation" of organic waste or effluents is a promising mean of producing renewable energy from waste products.

The genus *Clostridium* is known to produce hydrogen from carbohydrates using various different metabolic pathways which are promoted or inhibited by the prevailing culture conditions. The metabolic pathway has a maximum yield of four mol of hydrogen per mol of glucose. Each pathway is characterized by the specific metabolite such as acetate, butyrate, ethanol, lactate or formate. The acetate and butyrate pathways are the only ones which involve the release of molecular hydrogen, i.e. 4 mol hydrogen per mol glucose with acetate production and 2 mol hydrogen per mol glucose with butyrate production. Although the *Clostridium* genus is promising for fermentative hydrogen production, few investigations have used pure strains to make a detailed study of the optimal conditions for hydrogen production.

Many authors have reported that pH, temperature and stirring has a marked effect on hydrogen production from carbohydrate substrates. Since most of these investigations were carried out using mixed cultures of microorganisms or using pure cultures without pH control, little is known about the precise impact of pH on the hydrogen production rate and yield and the metabolic pathways involved.

Investigations were carried out to determine the optimum culture conditions for the production of hydrogen with *Clostridium butyricum* CWBI and to identify some other related limiting factors. Two substrates were used, glucose, the most used substrate in the literature for the characterization of *Clostridium*, and starch an abundant, inexpensive and reliable raw material.

Keywords: biohydrogen, dark fermentation, *Clostridium* Sp., anaerobic fermentation