INFLUENCE OF THE TEMPERATURE ON BIOHYDROGEN PRODUCTION BY PHOTOFERMENTATION

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ABSTRACT

The use of fossil fuels is one of the main causes of global warming, environmental degradation, and health problems. Furthermore, fossil fuel resources are limited and non-renewable, and their indiscriminate use will eventually lead to their extinction in the coming decades. From this perspective, hydrogen can be an alternative, as it is a source of low (or zero) carbon energy, depending on its production process. The current work verified the effect of the temperature on hydrogen production by photofermentation. For that, the purple non-sulfur bacteria *Rhodobacter capsulatus*, *Rhodospirillum rubrum*, and *Rhodopseudomonas palustres* (1:1:1) were used. In addition, glucose was used as a carbon source. Hydrogen was produced with a maximum productivity of 11.32 mmol H_2/(L.d).

Keywords: biohydrogen. photofermentation. purple non-sulfur bacteria.

1 INTRODUCTION

The increase in global energy needs, the depletion of fossil fuel reserves, and the environmental problems related to the continued use of these fuels define a new panorama for the 21st century and require new policies regarding energy sources. The alarming growth in demand for energy sources, associated with uncertainty regarding the availability and price of oil, leads to the adoption of practices for the development and exploration of new energy resources.[1] Therefore, hydrogen production emerges as a very attractive alternative source. Hydrogen production via a biological process can be carried out by direct biophotolysis from water using green algae, indirect photolysis from water by cyanobacteria, photofermentation of organic compounds by photosynthetic bacteria, anaerobic fermentation from organic compounds by fermentative bacteria, and hybrid systems involving photosynthetic and fermentative bacteria.[2,3] Photofermentation occurs through the action of purple non-sulfur photosynthetic bacteria that convert a carbon source (sugar or organic acid) to H_2 and CO_2.

Numerous factors influence the photofermentation, including light intensity, age of the inoculum, type of substrate, and temperature.[4] Temperature is one of the most relevant operational parameters in hydrogen production, as it alters the hydrogen-producing activity of bacteria.[5,6] An increase in temperature can lead to an increase in hydrogen production, although high temperatures result in a decrease in viability and performance by biological systems.[7] This study aims to investigate the influence of temperature on hydrogen production by photofermentation using photosynthetic bacteria such as *Rhodospirillum rubrum*, *Rhodobacter capsulatus*, and *Rhodopseudomonas palustris,* and glucose as a carbon source.

2 MATERIAL & METHODS

The strains *Rhodobacter capsulatus* DSM 1710, *Rhodospirillum rubrum* DSM 467, and *Rhodopseudomonas palustris* were acquired from the German Collection of Microorganisms and Cell Culture DSMZ. RCV medium was used.[8] The fermentation medium was RCV medium with glucose (Synth), as the carbon source, replacing malic acid. Assays were initially carried out in 50 mL disposable bioreactors with a working volume of 37 mL (with 32 mL of fresh medium and 5 mL of inoculum) and kept in a germination chamber at 31 and 35 °C or room temperature, under illumination of 2,500 lx by using LED lamps (9 W). Initial conditions were adjusted to 10 g/L glucose, 0.6 g_{vs}/L cell concentration, and pH 6.8. Hydrogen production was described by Cangani et al.[9]

The composition of the biogas was determined using a Shimadzu GC 17A chromatograph equipped with a thermal conductivity detector and capillary column Carboxen 1010. Argon was used as carrier gas. The quantification of organic acids and sugar was obtained by HPLC, a Shimadzu LC-20A Pronience chromatograph (SUPELCOGEL Ce610H column). The cell density was determined by a solution diluted from a sample of reaction medium. The absorbance of the solution was read in a Shimadzu spectrophotometer (660 nm).

3 RESULTS & DISCUSSION

Hydrogen production was evaluated using a bacterial system of *R. capsulatus, R. rubrum*, and *R. palustris* (1:1:1). The test lasted 144 h, and the fermentable medium used was modified RCV containing glucose at 10 g/L. The concentration of volatile solids corresponds to the concentration of cell biomass (Figure 1 a and b), reaching 0.76 and 0.77 g_{vs}/L after 144 h. The growth

represented an increase in the cell concentration of 1.4-fold at 31°C and 1.3-fold at 35°C about the initial value, with the peak value at 96 h after it started to decrease. Note in Figure 2 that in 96 h, there was maximum hydrogen productivity of about 1.33 and 0.80 mmol H_2/(L.d) and maximum hydrogen yield of 1.45 and 1.38 mmol H_2/mol substrate consumed, at 31 and 35° C respectively. The production of organic acid was also evaluated (Figure 3 a and b).

Organic acids can also induce a microbial community shift in favor of acid-resistant species that could favor or disfavor H_2 production.[10] The production of acetic and butyric acid is associated with a high yield of H_2, while lactic acid and propionic acid are associated with low H_2 yields.[11] The production of acetic, butyric, and propionic acids was similar. The production of lactic acid was lower at 31°C, consequently, it is inversely proportional to hydrogen productivity and cell concentration. A pilot operation shows the increase in temperature results in an increase in microbial activity. Increasing the temperature from 25 to 35°C increased ethanol production as for acetic acid, and from 35°C to 55°C these concentrations decreased, suggesting the behavior may be related to some change in metabolism for hydrogen production[12].

In addition, experimental assays were carried out at room temperature, therefore covering a wider temperature range. The biomass varied from 0.20 to 0.29 g/L, while the hydrogen productivity and yield increased 4-fold and 3-fold, respectively. The hydrogen productivity reached 11.32 mmol H_2/ (L.d). In comparison with the previously mentioned results, H_2 productivity strongly increased. In addition, organic acids behavior showed that lactic acid (1.05 g/L) is predominantly produced while acetic (0.17g/L) and propionic acids (0.30 g/L) were verified, data not shown. Although hydrogen production has been low, an improvement in the operability of the process can result in better performance in the target product synthesis by bacteria.

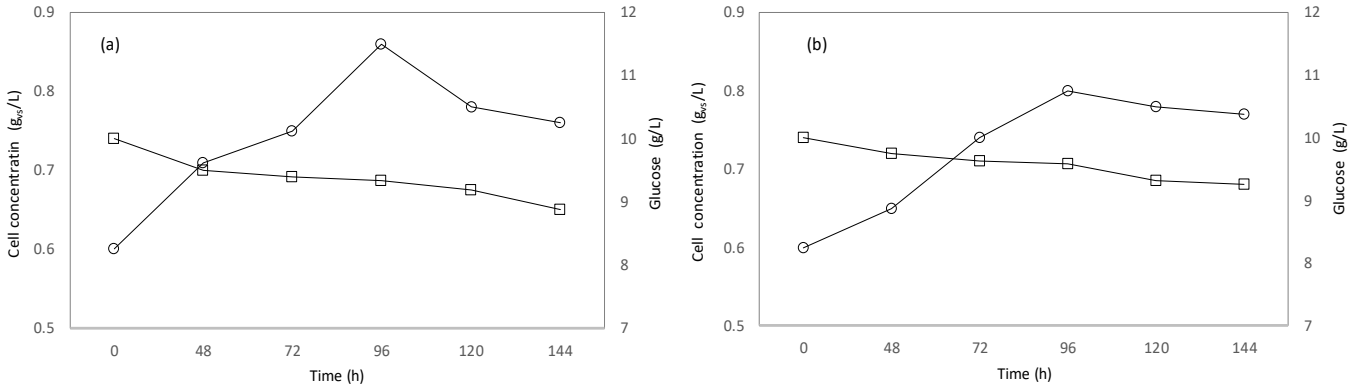

Figure 1. Profiles of cell concentration (○) and glucose (□): (a) 31°C and (b) 35°C.

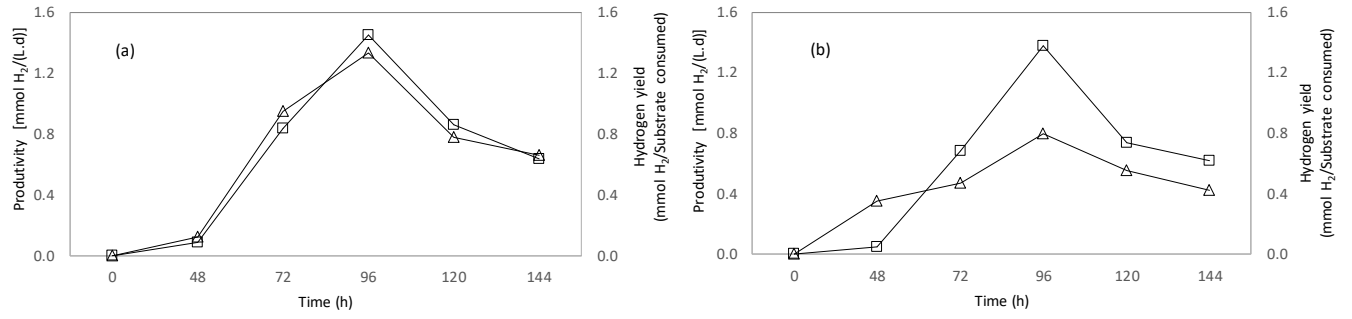

Figure 2. Profiles of hydrogen productivity(△) and hydrogen yield (□): (a) 31° C and (b) 35° C.

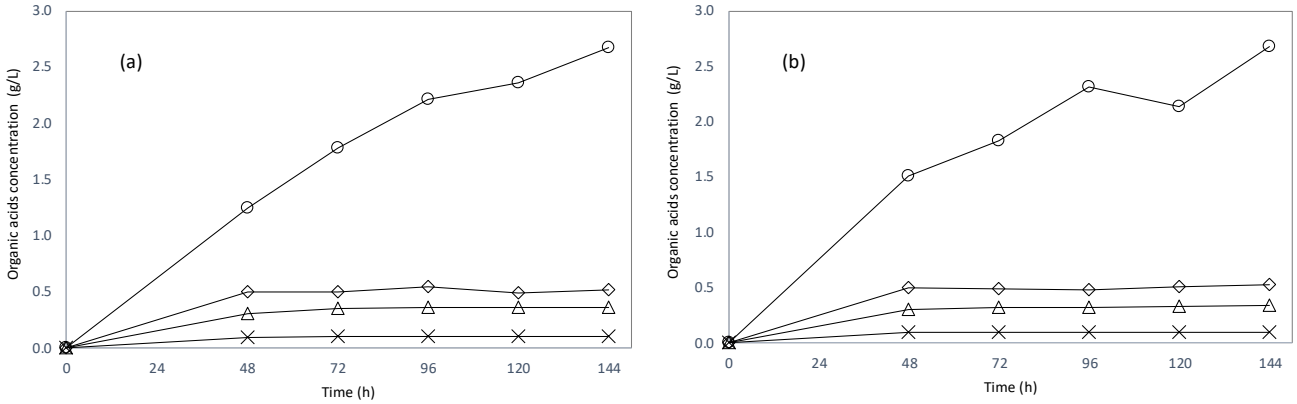

Figure 3. Profiles of acid organics (a) 31°C and (b) 35° C [latic acid (○), acetic acid (△), propionic acid (◊), and butyric acid (□)]

4 CONCLUSION

The current work showed that glucose and a combination of phototrophic bacteria (*R. capsulatus: R. rubrum: R. palustris*) for hydrogen production is feasible. Glucose, as a carbon source, was consumed by bacteria at an average of 60%, resulting in hydrogen production under light conditions. Maximum H_2 yield and H_2 productivity were 1.58 mol H_2/mol consumed glucose and 11.32 mmol H_2/(L d), respectively. The production of organic acids was an indicator of biofuel production. In contrast, the presence of propionic acid leads to a decrease in hydrogen synthesis.

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