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Biofilm production by *Zymomonas mobilis* enhances ethanol production and tolerance to toxic inhibitors from rice bran hydrolysate

Research Paper

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Microorganisms play a significant role in bioethanol production from lignocellulosic material. A challenging problem in bioconversion of rice bran is the presence of toxic inhibitors in lignocellulosic acid hydrolysate. Various strains of *Zymomonas mobilis* (ZM4, TISTR 405, 548, 550 and 551) grown under biofilm or planktonic modes were used in this study to examine their potential for bioconversion of rice bran hydrolysate and ethanol production efficiencies. *Z. mobilis* readily formed bacterial attachment on plastic surfaces, but not on glass surfaces. Additionally, the biofilms formed on plastic surfaces steadily increased over time, while those formed on glass were speculated to cycle through accumulation and detachment phases. Microscopic analysis revealed that *Z. mobilis* ZM4 rapidly developed homogeneous biofilm structures within 24 hours, while other *Z. mobilis* strains developed heterogeneous biofilm structures. ZM4 biofilms were thicker and seemed to be more stable than other *Z. mobilis* strains. The percentage of live cells in biofilms was greater than that for planktonic cells (54.32 ± 7.10% vs. 28.69 ± 3.03%), suggesting that biofilms serve as a protective niche for growth of bacteria in the presence of toxic inhibitors in the rice bran hydrolysate. The metabolic activity of ZM4 grown as a biofilm was also higher than the same strain grown planktonically, as measured by ethanol production from rice bran hydrolysate (13.40 ± 2.43 g/L vs. 0.432 ± 0.29 g/L, with percent theoretical ethanol yields of 72.47 ± 6.13% and 3.71 ± 5.24% respectively). Strain TISTR 551 was also quite metabolically active, with ethanol production by biofilm and planktonically grown cells of 8.956 ± 4.06 g/L and 0.0846 ± 0.064 g/L (percent theoretical yields were 48.37 ± 16.64% and 2.046 ± 1.58%, respectively). This study illustrates the potential for enhancing ethanol production by utilizing bacterial biofilms in the bioconversion of a readily available and normally unusable low value by-product of rice farming.

Introduction

Rising costs of fossil fuels in the last decades have made bioethanol an attractive alternative energy source. Biomass is a useful energy source because it can play a significant role in reducing greenhouse gas emission [20]. However, commercial bioethanol is commonly derived from the fermentation of edible food crops, such as starch based ethanol and sugarcane-based ethanol, resulting in increases in food prices and insufficient production to meet the increasing demands for bioethanol. Consequently, the future prospect is to use lignocellulosic materials for the production of bioethanol and to bring production toward the industrial scale [9]. Rice bran, a major lignocellulosic agricultural waste from the rice milling

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