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Isolation and Characterization of Biohydrogen-Producing Bacteria for Biohydrogen Fermentation Using Oil Palm Biomass-Based Carbon Source

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Abstract

The effectiveness of biohydrogen conversion from biomass sources is governed by the selection of ideal biohydrogen-producing bacteria to achieve high and consistent production performance. The aim of this research was to isolate and identify a biohydrogen producer in local soil samples, as well as to evaluate its fermentability in biohydrogen production from oil palm empty fruit bunches (OPEFB). To this end, preliminary identification was performed using morphological, phenotype, biological, and 16s rRNA analyses. The fermentability of the isolate was further evaluated in a serum bottle and then in a 1.5 L anaerobic column bioreactor (ACBR) to investigate the potential for biohydrogen production using two OPEFB-based carbon sources: hydrolysate of ammonia fiber expansion (AFEX)-pretreated OPEFB and molasses from dilute acetic acid (DAA)-pretreated OPEFB. The isolated strain, *Enterobacter* sp. KBH 6958, was found to be capable of producing biohydrogen from various carbon sources via the pyruvate:ferredoxin oxidoreductase (PFOR) pathway. The cumulative conversion of AFEX OPEFB hydrolysate was 45% higher than that observed in DAA OPEFB molasses fermentation in the production of biohydrogen. The biohydrogen yield after fermenting AFEX OPEFB hydrolysate with *Enterobacter* sp. KBH 6958 was 1.55 mol H₂/mol sugar, with a maximum productivity of 98.1 mL H₂/h (4.01 mmol H₂/L/h), whereas butyrate (10.6 mM), acetate (11.8 mM), and ethanol (4.56 mM) were found to be the major soluble metabolites. This study successfully demonstrated the biotechnological conversion of OPEFB into biohydrogen using a locally isolated strain, which not only solves environmental issues associated with the industry but may also offer a solution to the world's energy insecurity. © 2023 by the authors.

Author Keywords

anaerobic column bioreactor; biohydrogen; characterization; *Enterobacter* KBH 6958; isolation; OPEFB

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