JSR Publications

Available Online

J. Sci. Res. **2** (2), 351-361 (2010)

JOURNAL OF SCIENTIFIC RESEARCH

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Conversion of Waste Agriculture Biomass to Bioethanol by Recombinant *Saccharomyces cerevisiae*

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Received 23 July 2009, accepted in final revised form 13 March 2010

Abstract

Agricultural waste biomass has already been transferred to bioethanol and used as energy related products, although many issues such as efficiency and productivity still to be overcome. In this study, the protein engineering was applied to generate enzymes with completely reversed coenzyme specificity and developed recombinant yeasts containing those engineered enzymes for construction of an efficient biomass-ethanol conversion system. Recombinant yeasts were constructed with the genes encoding a wild type xylose reductase (XR) and the protein engineered xylitol dehydrogenase (XDH) (with NADP) of *Pichia stipitis*. These recombinant yeasts were characterized based on the enzyme activity and fermentation ability of xylose to ethanol. The protein engineered enzymes were expressed significantly in *Saccharomyces cerevisiae* as judged by the enzyme activity in vitro. Ethanol fermentation was measured in batch culture under anaerobic conditions. The significant enhancement was found in Y-ARS strain, in which $NADP^+$-dependent XDH was expressed; 85% decrease of unfavorable xylitol excretion with 26% increased ethanol production, when compared with the reference strain expressing the wild-type XDH.

Keywords: Agricultural waste biomass; Protein engineering; Xylitol dehydrogenase; Xylose-fermentation; Eethanol production.

DOI: 10.3329/jsr.v2i2.2882 J. Sci. Res. **2** (2), 351-361 (2010)

1. Introduction

The more efficient use of biomass is demanded to solve the global crisis such as depletion of fossil fuel and global warming. Woody biomass, including agriculture residues, wood chips, municipal solid wastes, paper wastes, etc., has already been transferred to

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