TITLE: FUNCTIONAL EXPRESSION OF A NEW XYLOSE ISOMERASE IN *SACCHAROMYCES CEREVISIAE*

AUTHORS: COLOMBO, G.S.; MENDES, I.V.; SOUTO, B.M.; PARACHIN, N.; ALMEIDA, J.R.; QUIRINO, B.F.

INSTITUTION: EMBRAPA AGROENERGIA, BRASÍLIA, DF (PARQUE ESTAÇÃO BIOLÓGICA, ED. EMBRAPA AGROENERGIA, CAIXA POSTAL 40.315, CEP 70770-901, BRASÍLIA, DF) E UNIVERSIDADE DE BRASÍLIA, (CAMPUS UNIVERSITÁRIO DARCY RIBEIRO, BRASÍLIA, DF, CEP 70910-900).

ABSTRACT:

Second-generation bioethanol is an alternative for the global demand of biofuel, having the advantage of being made from lignocellulosic biomass, which can be crop waste. Plant biomass has three fractions, one of which is hemicellulose, composed of hexoses and pentoses sugars, such as glucose and xylose, respectively. The yeast *Saccharomyces cerevisiae,* which is routinely used for bioethanol production, is unable to convert the pentose xylose into ethanol. Therefore, for the hemicellulosic fraction of plant biomass to be completely used industrially, metabolic modifications of *Saccharomyces* by genetic engineering are needed. When expressed in *Saccharomyces*, the gene encoding the enzyme xylose isomerase, which isomerizes xylose to xylulose, an intermediary of the pentose pathway, confers yeast the ability to metabolize xylose. The present work aims to evaluate the expression of a gene coding for a xylose isomerase, isolated from a metagenomic library and named *XyBet*, by an optimized strain of *S. cerevisiae*, developed in prior work at Embrapa Agroenergia. *S. cerevisiae* was transformed by heat shock with a protocol optimized for yeast based on the LiAc/SS Carrier DNA/PEG method. The strain transformed with the gene *XyBet* was named LXB and compared to a positive control strain expressing a known xylose isomerase originated from an anaerobic fungus and a negative control strain transformed with the empty vector. To evaluate the expression of *XyBet* by the transformed strain LXB, comparative growth curves in YNB medium containing xylose as the sole carbon source were obtained for the three strains, each cultivated in triplicate at 30° C with agitation at aerobic conditions. Xylose and ethanol were measured by HPLC and will be reported. The potential application of these strains for ethanol production from xylose will be discussed.

Keywords: bioethanol, yeast, xylose, xylose isomerase.

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