

A novel D-xylose isomerase from the gut of the wood feeding patent-leather beetle *Odontotaenius disjunctus*

Paulo César Silva^a, Javier Ceja-Navarro^b, Flávio Azevedo^a, Ulas Karaoz^b, Eoin L. Brodie^{bc} and Björn Johansson^a

(a) CBMA - Center of Molecular and Environmental Biology Engineering, University of Minho, Campus de Gualtar, Braga, 4710-057, Portugal. (b) Earth and Environmental Sciences, Lawrence Berkeley National Laboratory, Berkeley, California, USA. (c) Department of Environmental Science, Policy and Management, University of California, Berkeley, California, USA.

D-Xylose Isomerase (XI) is a key enzyme for the metabolism of D-xylose in renewable carbohydrate rich feedstocks such as lignocellulosic hydrolysates. The widely used industrial organism baker's yeast *Saccharomyces cerevisiae* can metabolize xylose upon heterologous expression of this enzyme. This enzyme is notoriously difficult to express in *S. cerevisiae* and only about ten active genes are known from prokaryotic and eukaryotic sources. We cloned a new XI from microorganisms in the gut of the wood feeding beetle *Odontotaenius disjunctus*. The new enzyme was functionally screened from a pool of enzymes with potential XI activity based on its sequence similarity to XI from *Piromyces* sp. strain E2. Interestingly, the newly identified enzyme and XI from *Piromyces* shared the highest sequence identity among the assayed enzymes. Cells carrying the new XI grew in media with D-xylose as the sole carbon source at a superior rate to that of XI from *Piromyces*, yet at a considerably inferior rate to that of the alternative xylose reductase–xylitol dehydrogenase pathway. Furthermore, optimal conditions of temperature and pH, kinetic parameters, and inhibition kinetics by xylitol were determined for the new enzyme. The physiological characterization of D-xylose fermenting *S. cerevisiae* expressing the new XI will be further discussed.