Functional screening for a novel D-xylose isomerase from the gut of a wood feeding beetle reveals efficient expression in *Saccharomyces cerevisiae*

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Renewable sugar rich feedstocks such as lignocellulosic hydrolysates remain one of the primary sources of potentially renewable fuel and bulk chemicals. The pentose sugar D-xylose is often present in significant amounts along with hexoses such as glucose and galactose. The yeast *Saccharomyces cerevisiae* can acquire the ability to metabolize D-xylose through expression of heterologous D-xylose isomerase (XI). This enzyme is notoriously difficult to express in *S. cerevisiae* and only fourteen genes have been reported to be active. We cloned a new XI from microorganisms in the gut of the wood feeding beetle *Odontotaenius disjunctus*. The new enzyme, 8454_2 XI, was functionally screened from a pool of enzymes with potential XI activity based on its sequence similarity to the XI from *Piromyces* sp. strain E2. A phylogenetic analysis revealed that the enzyme 8454_2 XI shares high identity with XIs from *Bacteroidia* class of the *Bacteroidetes* phylum, and all XIs from *Bacteroidia* screened in yeast so far have exhibited high activity. Cells carrying the new XI in D-xylose containing media as the sole carbon source showed higher growth and D-xylose consumption rates to those of XI of *Piromyces*. Remarkably, the 8454_2 XI also exhibited 2.6 times higher Vmax and 37 % higher affinity, and retained substantially higher relative activity at 30 ºC. The new XI is a useful addition to the molecular toolbox for genetic modification of *S. cerevisiae* for the metabolism of second-generation substrates.

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