ABSTRACT

Principal Investigator: Shinya Fushinobu

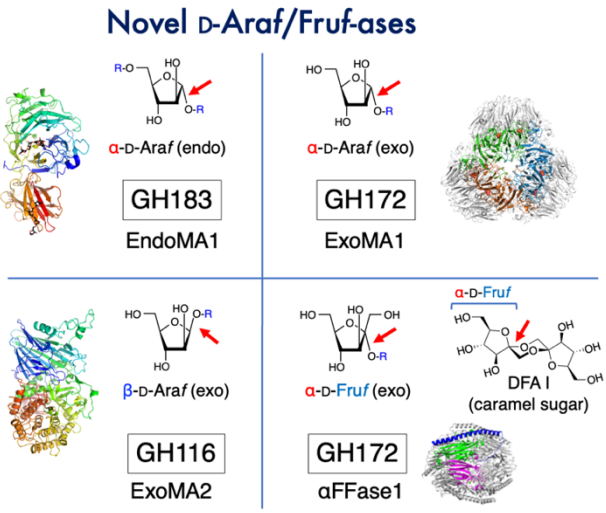
Grant Title: Structure-function analysis and functional exploration of novel furanosidase families

Purpose

We discovered novel glycoside hydrolase (GH) enzymes acting on α-D-fructofuranoside (α-D-Fru*f*), α-D-arabinofuranoside (α-D-Ara*f*), and β-D-arabinofuranoside (β-D-Ara*f*). Enzymes that cleave L-Ara*f* bonds, which are abundant as plant cell wall components, have been well studied, but enzymes that cleave D-Ara*f* bonds, which are present only in some pathogens, were only recently found. We aimed to explore additional novel enzymes related to these novel enzymes and to elucidate their functions and structures.

Methods

Novel genes for enzymes belonging to the GH families as above were found in the genome of an intestinal bacterium, and *E. coli* recombinant enzymes were characterized. X-ray crystallographic, NMR, cryo-EM, and QM/MM analyses of the D-Ara*f*-acting enzymes, and synthesis of probes necessary for these analyses were conducted.



Results

Three novel enzymes that can cleave α-D-Ara*f* and β-D-Ara*f* bonds were found. X-ray crystallographic, NMR, and QM/MM analyses of GH116 β-D-Ara*f*-ase ExoMA2 were performed. Cryo-EM structures of GH172 α-D-Ara*f*-ase ExoMA1 were also determined.

Reference

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2. A. Ishiwata, R. Fukushima, S. Fushinobu, K. Fujita, K. Tanaka, Y. Ito. Design and synthesis of the mechanism-based inhibitor probes against the glycoside hydrolase family 116 β-D-arabinofuranosidase. *Peptide Science*, in press (2024)