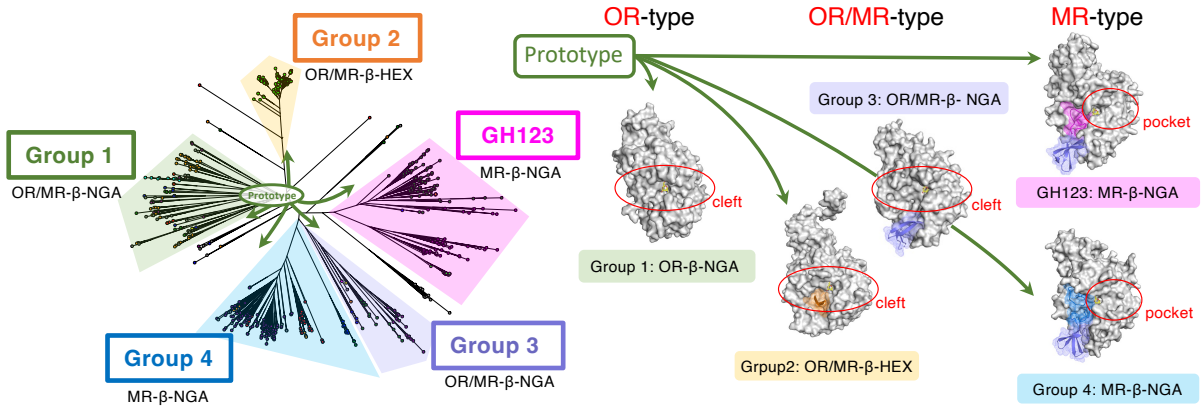
Principal Investigator: Tomomi Sumida
Grant Title: Screening and characterization of novel glycoside hydrolases from deep sea

Abstract
Glycans are widely distributed in nature as glycoconjugates of polysaccharides, glycolipids, glycoproteins, lipopolysaccharides and glycoRNA. The length, combination, and abundance of glycans are regulated by glycan-related enzymes. β-*N*-acetylgalactosaminiase (β-NGA), one of the glycan-related enzymes, hydrolyzes β-*N*-acetylgalactosamine (β-GalNAc) linkage of various glycans. Although β-GalNAc-containing glycans play important roles in biological function such as cell adhesion, interactions, signal transduction and immune responses, their biological significance is not yet fully understood. One of the reasons is that only one type of enzyme acting on β-GalNAc, the exo-β-NGA, has been found. In this study, to unveil the unprecedented functional diversity of β-NGA activity, we conducted sequence-based screening of β-NGA from deep-sea microbial assemblages. The deep-sea (below 200 meters depth) is an environment of darkness, low temperature, high pressure, and sometimes high temperature (for example, hydrothermal vents). The deep-sea environments are completely distinctive from those on land. However, due to the limited accessibility for sampling, microorganisms in the deep-sea have not been well studied. Therefore, the deep-sea is one of the frontiers to investigate undiscovered enzymes. In this study, based on the metagenomic analysis of deep-sea microbiota and subsequent domain searching, we identified four novel glycoside hydrolase groups and characterized eight novel β-NGAs and β-*N*-acetylhexosaminidase (β-HEX). Despite little sequence similarity, biochemical and structural characterization of these enzymes suggest that all enzymes share a prototype enzyme structure and have diversified substrate specificities (oligosaccharide-releasing (OR), oligosaccharide/monosaccharide (OR/MR)-releasing, and monosaccharide (MR)-releasing) through the accumulation of mutations and insertional amino acid sequences. The discovery and functional elucidation of these enzymes unveils the evolutional and functional machinery of glycan-related enzymes and will contribute to the understanding of the biological functions of β-GalNAc-containing glycans.



Genetic and functional diversity of novel β-*N*-actylgalactosamine-targeting glycosidases

Reference: T. Sumida et al., "Genetic and functional diversity of β-*N*-acetylgalactosamine-targeting glycosidases expanded by deep-sea metagenome analysis". *Nature Commun.*, 15, 3543 (2024)