Architecting pseudo-natural peptides by in vitro artificial biosynthesis

Yuki Goto
Department of Chemistry, Graduate School of Science, Kyoto University
E-mail: goto.yuki.4x@kyoto-u.ac.jp

Bioactive peptidic natural products often have unique non-proteinogenic structures, which are generally essential in their potent bioactivities. In the biosynthetic pathways of ribosomally synthesized and post-translationally modified peptides (RiPPs), the non-proteinogenic structures are generated by enzymatic structural modification on precursor peptides encoded in genetic information.

We have previously devised in vitro engineered biosynthesis systems for artificial RiPP analogs by combining a custom-made cell-free translation (Flexible In-vitro Translation; FIT) system with various RiPP modifying enzymes. In these systems, so-called FIT-RiPP, precursor peptides are expressed from synthetic DNA templates, and subsequently undergo posttranslational modifications by recombinant RiPP enzymes in a one-pot manner, allowing for high-throughput production of a variety of artificial RiPP analogs. One advantage of the FIT-RiPP strategy is its amenability to genetic code reprogramming, making it possible to yield RiPP analogs with multiple non-canonical amino acid residues. Another potential use of the FIT-RiPP system is an integration with in vitro display technology, enabling us to select artificial peptide ligands against protein targets of interest. In the present talk, we will discuss the latest advances in the FIT-RiPP strategy for the development of pseudo-natural RiPPs.