

Abstract

Expanded Toolbox for Directing the Biosynthesis of Macrocyclic Peptides in Bacterial Cells †

Jacob Iannuzzelli and Rudi Fasan

University of Rochester; jiannuz2@ur.rochester.edu (J.I.);
rudi.fasan@ur.rochester.edu (R.F.)

† Presented at the First Canadian Peptide and Protein Community Virtual Symposium, 27–28 May 2021; Available online: <https://cppc2021.sciforum.net/>.

Published: 27 May 2021

Abstract: The macrocyclization of recombinant polypeptides by means of genetically encodable non-canonical amino acids has recently provided an attractive strategy for the screening and discovery of macrocyclic peptide inhibitors of protein–protein interactions. Previously, our group has primarily utilized *O*-2-bromoethyl tyrosine (O2beY) for peptide macrocyclization. We report the development of an expanded suite of electrophilic unnatural amino acids (eUAAs) useful for directing the biosynthesis of genetically encoded thioether-bridged macrocyclic peptides in bacterial cells (*E. coli*). These reagents are shown to provide efficient access to a broad range of macrocyclic peptide scaffolds spanning from 2 to 20 amino acid residues, with the different eUAAs offering complementary reactivity profiles toward mediating short- vs. long-range macrocyclizations. Swapping of the eUAA cyclization module in a cyclopeptide inhibitor of streptavidin and Keap1, previously evolved using O2beY as the eUAA linker, led to compounds with markedly distinct binding affinity toward the respective target proteins, highlighting the effectiveness of this strategy toward tuning the structural and functional properties of bioactive macrocyclic peptides. These peptide cyclization strategies expand opportunities for the combinatorial biosynthesis of natural product-like peptide macrocycles in bacterial cells or in combination with display platforms toward the discovery of selective agents capable of targeting proteins and protein-mediated interactions.