Synthesis and screening large libraries of cyclic peptides for addressing intracellular protein-protein interactions

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Our laboratory is engaged in the discovery and development of cyclic peptides for therapeutic application. In recent years, we have started to address the long-standing goal of developing target-specific peptides that are membrane permeable. Towards this end, we focus on generating cyclic peptides that have a rather small size (< 700 Da) and a limited polar surface so that they have a high chance of passively crossing membranes.

For generating such sub-kilodalton cyclic peptides that bind to disease targets of interest, we have established an approach based on nanomole-scale cyclic peptide synthesis and high-throughput screening (1-3). In brief, we produce thousands of peptides by solid phase peptide synthesis and diversify them combinatorially by reacting with a myriad of chemical building blocks. In this approach, the peptides and chemical building blocks are transferred in nanoliter volumes by acoustic dispensing and the reactions performed at a nanomole scale, allowing the synthesis and screening of ten-thousands of cyclic peptides in a short time.

In my talk, I will explain the cyclic peptide library synthesis and screening approach, show examples of libraries and their screening, present nanomolar ligands that we have developed against different proteins, including a protein-protein interaction target, and show data about the membrane permeability of the peptides.

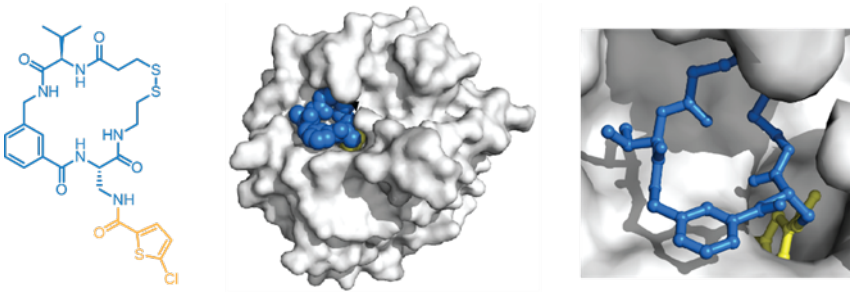

Figure: Chemical and X-ray structure of small cyclic peptide inhibitor of thrombin identified by synthesizing and screening ten-thousands of cyclic peptides at a nanomole-scale.

[1] S. Habeshian, et al., Cyclative release strategy to obtain pure cyclic peptides directly from the solid phase, *ACS Chemical Biology*, **2022**, *17*, 181–186.
[2] Z. Bognar, et al., Solid-phase peptide synthesis on disulfide-linker resin followed by reductive release affords pure thiol-functionalized peptides, *Organic & Biomolecular Chemistry*, **2022**, *17*, 5699-5703.
[4] S. Habeshian, et al., Synthesis and direct assay of large macrocycle diversities by combinatorial late-stage modification at picomole scale, *Nature Communications*, **2022**, *13*, 3823.