Combinatorial Synthesis of Aeruginosin Derivatives and Development of Their Fluorescent Probes

○ Yoichiro Hoshina,[1] Hiroyuki Mogi,[2] Yoshifumi Yamada,[3] Takayuki Doi,[2] Hiroshi Tanaka,[2] Takashi Takahashi[2]
([1]Kaken Pharmaceutical Co., Ltd., [2]Tokyo Institute of Technology, [3]Olympus Corporation)

A combinatorial synthesis based on natural products is a powerful tool to cover a chemical space for drug candidates. We have studied solid-phase combinatorial syntheses for a variety of naturally occurring skeletons, such as steroids, trisaccharides, peptides, macrolides, alkaloids, and prostanoids. Aeruginosin 298-A, isolated from *Microcystis aeruginosa* (NIES-298), exhibited inhibitory activity for serine proteases. Aeruginosin 298-A consists of a *p*-hydroxyphenyllactic acid (D-Hpla), a D-leucine (D-Leu), an unusual 2-carboxy-6-hydroxyoctahydroindole (L-Choi), and an L-argininol (L-Argol). We report herein a combinatorial synthesis of aeruginosins and their analogues utilizing a silyl linker on a polymer-support and the evaluation of their biological activity. As a result, 24 aeruginosin derivatives were synthesized in excellent purity. In addition, it was found that one of the product (D-Hpla-D-Leu-L-Choi-Agma) was 300 times more potent than parent aeruginosin 298-A.

Fluorescent-labeled chemical probes are useful in high-throughput screening (HTS) assays of libraries of compounds. Fluorescence correlation spectroscopy (FCS) is a single-molecule detection technique using fluorescent-labeled chemical probes and enables the binding affinity of the chemical probes to proteins in solution to be estimated by simple manipulation. In this presentation, we report a design and synthesis of some fluorescent-labeled aeruginosins by using the solid-phase synthesis. To demonstrate the utility of the fluorescent probes, we used them in an FCS-based competitive binding assay of an aeruginosin library. One of an appropriate fluorescent-labeled derivative gave the results that were comparable to the protease inhibition assay.