Academic Program [Oral B] | 16. Natural Products Chemistry, Chemical Biology：Oral B

Wed. Mar 26, 2025 1:00 PM - 3:30 PM JST | Wed. Mar 26, 2025 4:00 AM - 6:30 AM UTC
[A]A305(A305, Bldg. 1, Area 3 [3F])

[[A]A305-1pm] 16. Natural Products Chemistry, Chemical Biology

Chair: Ryo Okamoto, Daisuke Takahashi

English
1:00 PM - 1:20 PM JST | 4:00 AM - 4:20 AM UTC
[[A]A305-1pm-01]
Synthesis of glycopolymer-modified antibodies for cancer immunotherapy using sugar antigens

〇Zenya Naraoka[1], Sakura Kawahara[1], Ryohei Miyagawa[2], Yoshiyuki Manabe[2], Koichi Fukase[2], Tomonari Tanaka[1] (1. Kyoto Inst. Tech., 2. Osaka Univ.)

Japanese
1:20 PM - 1:40 PM JST | 4:20 AM - 4:40 AM UTC
[[A]A305-1pm-02]
Synthesis of *O*-linked glycan core structures using glycoside hydrolases

〇Shunsuke Nakada[1], Kisuke Tonomura[2], Takayuki Ohnuma[2], Hisashi Ashida[2], Katoh Toshihiko[3], Takane Katayama[3], Tomonari Tanaka[1] (1. Kyoto Institute of Technology, 2. Kindai Univ., 3. Kyoto Univ.)

English
1:40 PM - 2:00 PM JST | 4:40 AM - 5:00 AM UTC
[[A]A305-1pm-03]
Creation of a New Macrolide Antibiotic against Non-tuberculous *Mycobacterium* by Late-stage Boron-mediated Aglycon Delivery

〇Yuka Isozaki[1], Takumi Makikawa[1], Kosuke Kimura[1], Daiki Nishihara[2], Maho Fujino[2], Yoshikazu Tanaka[2], Chigusa Hayashi[3], Yoshimasa Ishizaki[3], Masayuki Igarashi[3], Takeshi Yokoyama[2], Kazunobu Toshima[1], Daisuke Takahashi[1] (1. Keio University, 2. Tohoku University, 3. Institute of Microbial Chemistry)

English
2:00 PM - 2:20 PM JST | 5:00 AM - 5:20 AM UTC
[[A]A305-1pm-04]
Elucidation of novel glycan functions that promote an α-helix formation of peptides

〇Intan Hawina Anjari[1], Kohtaro Hirao[1,2], Yuta Maki[1,2], Ryo Okamoto[1,2], Yasuhiro Kajihara[1,2] (1. Grad. Sch. Sci. Osaka Univ., 2. FRC, Grad. Sch. Sci. Osaka Univ.)

2:20 PM - 2:30 PM JST | 5:20 AM - 5:30 AM UTC
Break

English
2:30 PM - 2:50 PM JST | 5:30 AM - 5:50 AM UTC
[[A]A305-1pm-05]
Synthesis and properties of α-helical peptides doubly-crosslinked with isophthalic acid-based crosslinking agents

〇Tetsuya Yasukagawa[1], Junuya Chiba[1], Yuki Ohishi[1], Satoru Yokoyama[1], Zhou Yue[1], Masahiko Inouye[1] (1. The Univ. of Toyama)

Japanese

2:50 PM - 3:10 PM JST | 5:50 AM - 6:10 AM UTC

[[A]A305-1pm-06]

Development of Growth Inhibitor for Mushroom Based on Coprinoferrin

〇Tomohiro Tsutsumi[1], Haruka Suda[1], Chika Ando[2], Yuta Tsunematsu[2], Ichiro Hayakawa[1] (1. Graduate School of Integrated Basic Sciences, Nihon University, 2. Graduate School of Bioagricultural Sciences, Nagoya University)

English

3:10 PM - 3:30 PM JST | 6:10 AM - 6:30 AM UTC

[[A]A305-1pm-07]

Development of thiazole-containing cyclic peptide ligands by an mRNA-display-coupled post-translational chemoenzymatic modifications

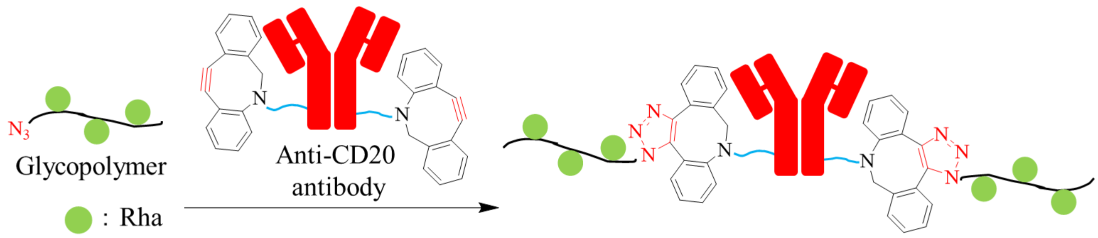
〇Akihiro Saito[1], Hiroaki Suga[2], Yuki Goto[1] (1. Kyoto University, 2. The University of Tokyo)

Synthesis of glycopolymer-modified antibodies for cancer immunotherapy using sugar antigens

([1]*Kyoto Inst. Tech.*, [2]*Osaka Univ.*) ○Zenya Naraoka,[1] Sakura Kawahara,[1] Ryohei Miyagawa,[2] Yoshiyuki Manabe,[2] Koichi Fukase,[2] Tomonari Tanaka[1]

α-Rhamnose (Rha) is a sugar antigen does not present in the human body and causes immune responses by an anti-Rha antibody.[1)] Recently, cancer immunotherapy using immune responses to sugar antigens has attracted much attention. Anti-cancer antibodies chemically modified with Rha are targeted by the specificity of the antibody and selectively kill cancer cells through an immune response to Rha.[2)] Furthermore, it has been reported that the introduction of Rha-bearing dendrimers onto an antibody strongly induces anti-Rha antibodies, resulting in the efficient immune response due to the glycosluster effect.[3)] The introduction of glycopolymers, synthetic polymers bearing saccharides, onto antibodies is also expected to strongly induce the anti-Rha antibodies. In this study, we synthesized glycopolymer-modified antibodies with numerous Rha moieties on the polymer side chain for application in cancer immunotherapy.

Polymers bearing activated esters in the side chains were synthesized by a reversible addition-fragmentation chain-transfer (RAFT) polymerization using an azide-containing chain transfer agent. Subsequently, glycopolymers with an azide group at the terminal group were synthesized by amidation reaction, a post-polymerization modification, with an amino-group-containing Rha derivative. Unreacted active esters were treated with excess 1-amino-2-propanol. After introduction of PEG linkers with a dibenzocyclooctine (DBCO) group onto Rituximab, anti-CD20 antibody, copper-free Huisgen cycloaddition reactions were performed to obtain the glycopolymer-modified antibodies (Scheme 1). The products were detected using MALDI-TOF MS analysis. Additionally, the results using strain-promoted inverse electron-demand Diels-Alder cycloaddition, called tetrazine click chemistry, and thiol-ene reaction will be reported.



Scheme 1 Synthesis of the glycopolymer-modified antibodies

Keywords: *α-Rhamnose; Cancer immunotherapy; Click Chemistry; Antibody; Glycopolymer*

1) W. Chen *et al.*, *ACS Chem. Biol.* **2011**, 6, 2, 185. 2) K. Zhou *et al.*, *J. Med. Chem.* **2022**, *65*, 323. 3) J. Sianturi *et al.*, *Angew. Chem. Int. Ed.* **2019**, *58*, 4526.

糖質加水分解酵素を用いた *O*-結合型糖鎖コア構造の合成

(京工繊大院工芸 [1]・近大院農 [2]・近大アグリ技研 [3]・近大生物理工 [4]・京大院生命 [5])
○中田 竣介 [1]・殿村 規介 [2]・大沼 貴之 [2,3]・芦田 久 [4]・加藤 紀彦 [5]・片山 高嶺 [5]・田中 知成 [1]

Synthesis of *O*-Linked Glycan Core Structures Using Glycoside Hydrolases ([1]*Graduate School of Science and Technology*, *Kyoto Institute of Technology*, [2]*Graduate School of Agriculture*, *Kindai University*, [3]*Agricultural Technology and Innovation Research Institute, Kindai University*, [4]*Biology-Oriented Science and Technology*, *Kindai University*, [5]*Graduate School of Biostudies*, *Kyoto University*) ○ Shunsuke Nakada[1], Kisuke Tonomura[2], Takayuki Ohnuma[2,3], Hisashi Ashida[4], Toshihiko Katoh[5], Takane Katayama[5], Tomonari Tanaka[1]

O-Linked glycans binding onto the hydroxyl groups of serine and threonine residues in proteins are frequently found as mucin glycoproteins and have been revealed to protect digestive organs and provide nutrients for symbiotic bacteria. There are eight types of core structures in *O*-linked glycans. Core 1, 2, 3 and 4 structures are found in intestinal mucins. Many chemical and glycosyltransferases-catalyzed synthesis of core oligosaccharides have been reported. , there are few reports of core oligosaccharides synthesis using glycoside hydrolases. In this study, we synthesized core 6 disaccharide (GlcNAc*β*1-6GalNAc) using a *β*-*O*-*N*-acetylglucosaminidase (OGA), which is classified under glycoside hydrolase family 84 (GH84), and a sugar oxazoline derivative. Furthermore, core 3 disaccharide (GlcNAc*β*1-3GalNAc) was synthesized and Core 4 trisaccharide (GlcNAc*β*1-3(GlcNAc*β*1-6)GalNAc) using a GH20 OGA. Additionally, core 2 trisaccharide (Gal*β*1-3(GlcNAc*β*1-6)GalNAc) was synthesized using the core 6 disaccharide, 4,6-dimethoxy triazinyl galactoside, and GH35 *β*-1,3-galactosidase.

Keywords : glycosidase, transglycosylation, N-acetylglucosaminidase, galactosidase, O-linked glycan

O-結合型糖鎖とはタンパク質のセリンまたはスレオニン残基の水酸基に結合した糖鎖である。この糖鎖は主にムチン糖タンパク質として存在し、消化器官の保護や共生細菌の栄養源になっていることが明らかになってきている。*O*-結合型糖鎖には八種類のコア構造が存在し、腸内ムチンでは主に Core 1～4 構造が多く見られる。*O*-結合型糖鎖コア構造オリゴ糖の合成法として有機合成法や糖転移酵素を用いた酵素合成法がよく研究されているが、糖質加水分解酵素を触媒とするグリコシル化反応を用いた合成例はほとんどない。本研究では、糖質加水分解酵素ファミリー84（GH84）に分類される *β*-*O*-*N*-アセチルグルコサミニダーゼ (OGA)と糖オキサゾリン誘導体を用いたグリコシル化反応によって、*O*-結合型糖鎖の Core 6 二糖(GlcNAc*β*1-6GalNAc)を合成した。さらに、GH20 OGA によるグリコシル化反応によって Core 3 二糖(GlcNAc*β*1-3GalNAc)と Core 4 三糖(GlcNAc*β*1-3(GlcNAc*β*1-6)GalNAc)を合成した。また、合成した Core 6 二糖と 4,6-dimethoxy triazinyl 糖、GH35 *β*-1,3-ガラクトシダーゼを用いて Core 2 三糖(Gal*β*1-3(GlcNAc*β*1-6)GalNAc)を合成した。

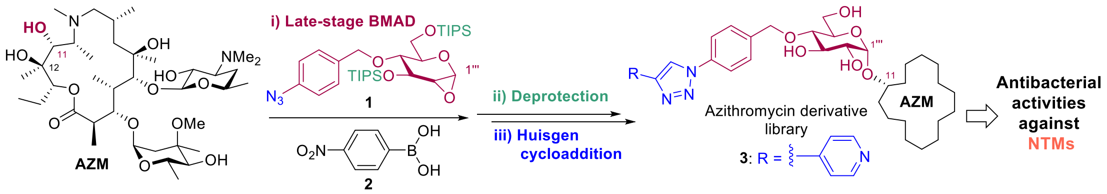
Creation of a New Macrolide Antibiotic against Non-tuberculous *Mycobacterium* by Late-stage Boron-mediated Aglycon Delivery

([1]*Faculty of Science and Technology, Keio University*, [2]*Graduate School of Life Sciences, Tohoku University,* [3]*Institute of Microbial Chemistry*) ○Yuka Isozaki,[1] Takumi Makikawa,[1] Kosuke Kimura,[1] Daiki Nishihara,[2] Maho Fujino,[2] Yoshikazu Tanaka,[2] Chigusa Hayashi,[3] Yoshimasa Ishizaki,[3] Masayuki Igarashi,[3] Takeshi Yokoyama,[2] Kazunobu Toshima,[1] Daisuke Takahashi[1]

Keywords: Late-stage Glycosylation; Boron-mediated Aglycon Delivery; Macrolide Antibiotic; Non-tuberculous Mycobacteria; Drug-resistant Bacteria

Non-tuberculous mycobacteria (NTM) is a recently emerging pathogen causing the pulmonary NTM disease. The macrolide azithromycin (AZM) is the standard first-line antibiotic for the treatment of the disease. However, the rise of drug-resistant NTM necessitates the development of novel therapeutics. In this context, our laboratory has developed the late-stage boron-mediated aglycon delivery (BMAD), which can efficiently introduce a sugar moiety regio- and 1,2-*cis*-stereoselectively to unprotected glycosides under mild conditions.[1)] Herein, we report on the development of a late-stage modification method of AZM utilizing BMAD, and its application to the creation of a new lead compound with higher antibacterial activity not only against wild-type NTM but also against macrolide-resistant NTM.

Initially, we designed a new library of AZM derivatives that were expected to express high binding activity to the 23S rRNA of macrolide-resistant NTM, by introducing various functional groups via glucose at position C-11 of AZM. Next, BMAD reaction of AZM and **1** using a catalytic amount of boronic acid **2** was examined. It was found that the glycosylation proceeded regio- and stereoselectively, and the subsequent deprotection of the silyl groups and Huisgen cycloadditions with various acetylene compounds provided a library of AZM derivatives in good yields. Next, the antibacterial activities of the library against NTMs (*M. avium* and *M. intracellulare*) were evaluated by broth dilution method. As a result, it was found that AZM derivative **3** exhibited effective antimicrobial activity against not only wild-type NTM but also macrolide-resistant NTM, thus successfully creating a promising new lead compound against pulmonary NTM disease.[2)]



1) Review: Takahashi, D.; Toshima, K. *Adv. Carbohydr. Chem. Biochem.* **2022**, *82*, 79.
2) Isozaki, Y.; Makikawa, T.; Kimura, K.; Nishihara, D.; Fujino, M.; Tanaka, Y.; Hayashi, C.; Ishizaki, Y.; Igarashi, M.; Yokoyama, T.; Toshima, K.; Takahashi, D. *Submitted.*

Elucidation of Novel Glycan Function That Promotes an α-Helix Formation of Peptides

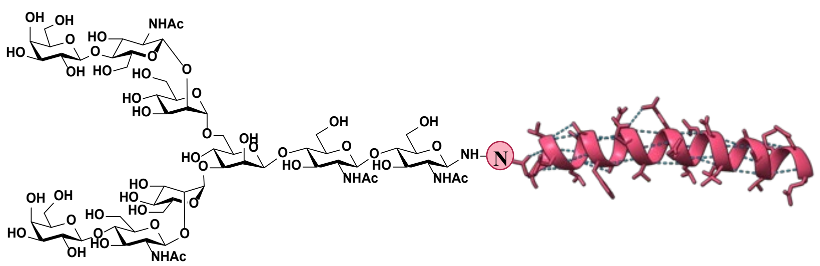
([1]*Graduate School of Science, Osaka University*, [2]*FRC, Graduate School of Science, Osaka University*) ○Intan Hawina Anjari,[1] Kohtaro Hirao,[1,2] Yuta Maki,[1,2] Ryo Okamoto,[1,2] Yasuhiro Kajihara[1,2]

Keywords: Glycan; Glycopeptide; *α*-Helix; CD; NMR

Among protein modifications, glycosylation is one of the most abundant modifications in nature. Several glycan functions have been widely reported, particularly in relation to the stabilities, activities, and properties of proteins. Previously, Kajihara's group found that glycan can enhance the *α*-helix formation of glucagon and exenatide.[1,2] However, the underlying mechanisms of this phenomena remain unclear. Here, we conducted a detail study on the role of glycan in promoting the secondary structure of peptides, specifically focusing on the *α*-helix formation.

We have been investigating the glycan functions on glycopeptide using homogeneous glycopeptides obtained through chemical synthesis. For this purpose, several peptide fragments of proteins and their glycosylated forms consisting of less than 30 amino acids were chemically synthesized using Fmoc solid-phase synthesis (Fmoc-SPPS) method.

Using synthesized peptides and their glycosylated forms, the secondary structures were evaluated by circular dichroism (CD) spectroscopy and nuclear magnetic resonance (NMR) measurement. Particularly, the detailed comparisons were examined between peptides with glycan and without glycan. As a results, we found that glycans influence the secondary structure formation of peptides. In this presentation, we present a comprehensive discussion of this glycan function.



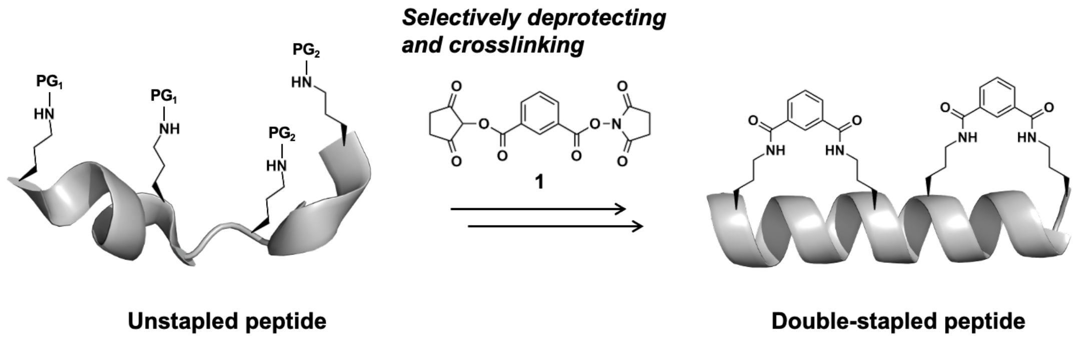
1) Liu, M. *et. al; Bioconjug. Chem.* **2021**, *32,* 2148-2153. 2) Chandrashekar, C. *et. al; Bioconjug. Chem.* **2023**, *34*, 1014-1018.

Synthesis and properties of α-helical peptides doubly-crosslinked with isophthalic acid-based crosslinking agents

(*Graduate School of Pharmaceutical Sciences, University of Toyama*)
○Tetsuya Yasukagawa, Junya Chiba, Yuki Ohishi, Satoru Yokoyama, Zhou Yue, Masahiko Inouye.
Keywords: Stapled Peptide; Protein-Protein Interaction; Proteolytic Stability; Protein Kinase A

Protein-Protein Interactions (PPIs) play a crucial role in various biological functions. We have developed an isophthalic acid-based crosslinking agent **1** for creating α-helical peptides. In the native peptides, two ornithine residues were placed at *i* and *i*+7 positions as a stapling point for the crosslinking. The crosslinked peptides with **1** retained high α-helical contents, presenting a simple helix-reinforcing strategy only using naturally occurring amino acids. In addition, our stapled peptides having the binding domain sequence of an apoptosis-inducing protein showed higher proteolytic stability than that of the unstapled counterpart. The stapled peptides exhibited internalization into HeLa and HCT-116 cells and induced their apoptosis.[1)]

In this time, we planned to build up double-stapled peptides by using **1** in order to further increase the proteolytic stability. We picked up a sequence of the reported R1AD peptide that selectively binds to protein kinase A regulatory subunit 1α (PKA-RIα). The R1AD peptide was optimized to disrupt PPIs between PKA-RIα and A-kinase anchoring proteins (AKAPs). However, the peptide was easily cleaved by ubiquitous proteases. Single- and double-stapled R1AD peptides were prepared in high yields by means of conventional solid-phase peptide synthesis (SPPS) with selective deprotection of ornithine residues and subsequent crosslinking with **1**. The double-stapled peptide showed significantly high helicity and high affinity for the target protein PKA-RIα (K_d = 0.11 nM). Furthermore, the double-stapled peptide displayed high proteolytic stability and efficient intracellular uptake compared to those of the unstapled and the single-stapled ones.



1) M. Inouye et al. *ChemBioChem* 2014, *15*, 2571–2576; *Chem. Commun.* 2017, *53*, 12104–12107.

日本化学会 第105春季年会 (2025)

コプリノフェリンに基づいたキノコ成長阻害剤の創製研究

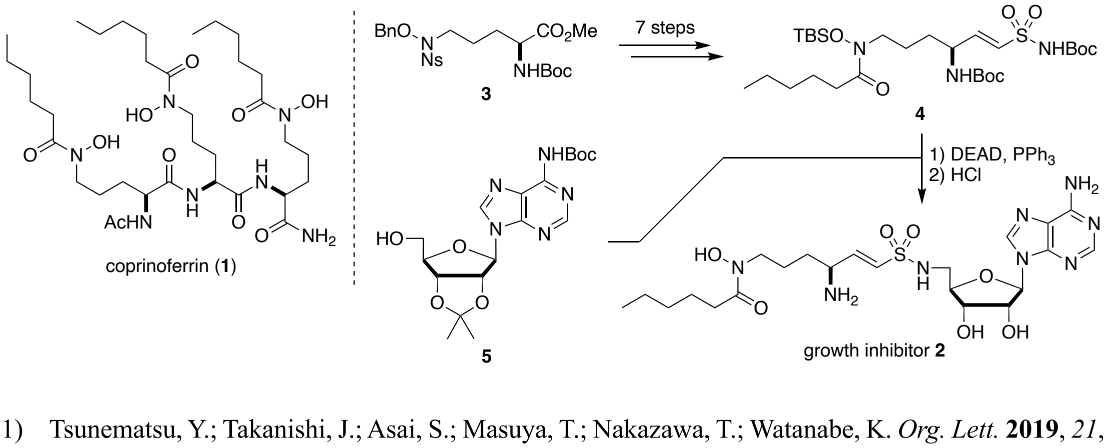
(日大院総合基[1]・名大院生命農[2]) ○堤 大洋[1]・須田 桜香[1]・安藤 知佳[2]・恒松 雄太[2]・早川 一郎[1]

Development of Growth Inhibitor for Mushroom Based on Coprinoferrin ([1]*Graduate School of Integrated Basic Sciences, Nihon University*, [2]*Graduate School of Bioagricultural Sciences, Nagoya University*) ○Tomohiro Tsutsumi,[1] Haruka Suda,[1] Chika Ando,[2] Yuta Tsunematsu,[2] Ichiro Hayakawa[1]

Coprinoferrin (**1**), a siderophore exists in various mushroom. **1** exhibits activity in cell growth and fluting body formation of mushrooms. In this study, we designed a novel mushroom growth inhibitor **2** containing a vinyl sulfonamide group, which acts as a covalent isostere of a thioester. We achieved the synthesis of **2** from an L-ornithine derivative **3** in nine steps. We will report design, synthesis, and biological activities of the growth inhibitor for mushroom.
Keywords : Coprinoferrin; Mushroom; Growth inhibitor

コプリノフェリン(**1**)は、様々なキノコに普遍的に存在するシデロフォアであり、キノコの菌糸成長・子実体形成を促進することが報告されている[1]。今回我々は、**1** の生合成経路に着目し、ビニルスルホンアミド基を有するキノコ成長阻害剤 **2** を設計した。これまでに、当研究室では **1** の全合成を達成しており[2]、その合成中間体である L-オルニチン誘導体 **3** を用いて、**2** の合成を行った。

はじめに、**3** から 7 工程の変換でビニルスルホンアミド **4** を得た。得られた **4** に対して、光延反応によるアデノシン誘導体 **5** との縮合を行った後、全ての保護基を除去することで目的の **2** を合成した。現在、合成した **2** の活性評価を行っている。本発表では、キノコ成長阻害剤の設計、合成および活性評価の詳細を報告する。



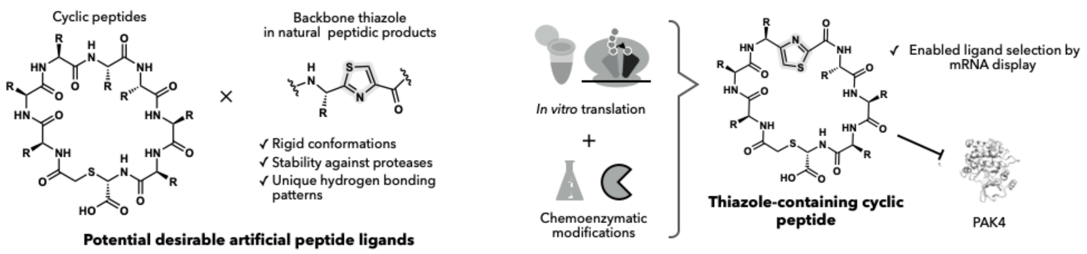
1) Tsunematsu, Y.; Takanishi, J.; Asai, S.; Masuya, T.; Nakazawa, T.; Watanabe, K. *Org. Lett.* **2019**, *21*, 7582.
2) Hayakawa, I.; Isogai, T.; Takanishi, J.; Asai, S.; Ando, C.; Tsutsumi, T.; Watanabe, K.; Sakakura, A.; Tsunematsu, Y. *Org. Biomol. Chem.* **2024**, *22*, 831.

Development of thiazole-containing cyclic peptide ligands by an mRNA-display-coupled post-translational chemoenzymatic modifications

([1]*Kyoto University,* [2]*The University of Tokyo*) ○Akihiro Saito[1], Hiroaki Suga[2], Yuki Goto[1]
Keywords: cyclic peptide; *in vitro* molecular selection; *in vitro* translation; post-translational modifications; genetic code reprogramming

Backbone thiazole moieties are widely found in peptidic natural products, possibly due to their rigid conformations, resistance to protease and hydrolysis[1], and unique hydrogen-bonding patterns[2]. These intrinsic attributes confer an advantage to the presence of backbone thiazoles in artificial cyclic peptides, enhancing their potential for ligand development. However, exploring *de novo* thiazole-containing peptide ligands with high efficiency and reliability has proven challenging. In this study, we focus on the synthesis of thiazole-containing peptides by *in vitro* post-translational chemoenzymatic modifications, which can synthesize diverse sequences under mild aqueous conditions. By applying this approach to mRNA display, we aim to establish a methodology to obtain cyclic peptide ligands containing backbone thiazoles. The synthetic method involves ribosomal incorporation of thioamides into peptides[3], spontaneous heterocyclization of thioamide and adjacent Cys, to form thiazoline, followed by thioether macrocyclization and enzymatic oxidation by GodE.

To achieve the specified goal, we first improved the synthesis method by focusing on the translation step in the post-translational chemoenzymatic modification method to increase the efficiency of desired product formation, making the method more versatile. We then constructed diverse thiazole-containing cyclic peptide libraries using the versatile synthetic method. Through *in vitro* selection of ligands using mRNA display, we obtained thiazole-containing cyclic peptide ligands with high binding affinities and inhibitory activities against p21-activated kinase 4 (PAK4), demonstrating their potential for drug development applications. This study established a selection system which expedite *de novo* discovery of desirable cyclic peptide ligands containing backbone thiazoles.



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