刺激分解性ビオチン化試薬を用いた細胞内タンパク質送達

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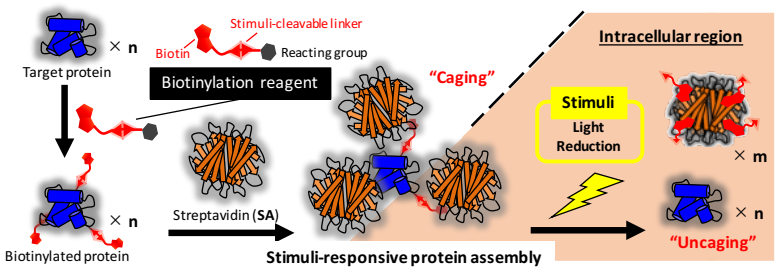
Intracellular protein delivery using stimuli-cleavable biotinylation reagents ([1]*Graduate School of Engineering, The University of Tokyo*, [2]*Research Center for Advanced Science and Technology*) ○Kazuho Yamamoto[1], Satoshi Yamaguchi[2], Akimitsu Okamoto[1,2]

Regulation of cellular functions in the specific cells can contribute to a variety of medical technologies. As methods for “on-demand” activation of protein, stimuli-responsive “caging” have attracted much attention[1)]. Our group has also developed photolytic protein assemblies consisting of biotinylated target proteins and streptavidin (SA) by using a photo-cleavable biotinylation reagent, and reported the light-induced release of target protein in the extracellular region[2)]. In this study, we aimed to develop two kinds of stimuli-responsive protein assemblies, photo-degradable assembly and redox-degradable assembly for intracellular protein release. Saporin (Sap), a ribosome-inactivating protein, was used as a target protein. As a result, we were able to confirm the dissolution of both assemblies upon stimulation. Furthermore, light-induced death of the cells transfected with photo-degradable assembly was achieved by non-toxic light irradiation. Also, redox-responsive death of the cells transfected with redox-degradable assembly was achieved responding to reductive environment in the cells.

Keywords : Drug Delivery System; Protein assembly; Protein bodies; Stimuli-degradable; Cancer Therapy

タンパク質による特定の細胞機能制御は幅広い医療技術に貢献する。タンパク質の「オンデマンド」活性化の手法として、刺激応答性のケージング法が注目されてきた[1)]。我々も、光分解性ビオチン化試薬を用いたビオチン化標的タンパク質とストレプトアビジン(SA)からなる光溶解性ナノ集合体を開発し、細胞外での光依存的な標的タンパク質の活性化を報告してきた[2)]。本研究では、光溶解型と還元溶解型の 2 種の刺激応答性複合体を作製し、細胞内での刺激に応じたタンパク質放出を試みた。標的タンパク質としてリボソーム不活化タンパク質であるサポリン(Sap)を用いたところ、どちらの複合体も刺激に応じた溶解が確認された。さらに、各々の複合体を細胞に導入した結果、外部からの光照射及び、細胞内還元環境に応じた細胞死誘導が観察された。



1) Lawrence, *et al.*, *Curr. Opin. Chem. Biol.* **2005**, *9*, 570; 2) Ishiwatari, *et al.*, *Adv. Healthcare Mater.* **2016**, *5*, 1002