

ストレプトリジン O 毒素タンパク質 (溶血レンサ球菌毒素)

01-531 20 µg, 01-531-5 5 x 20 µg

保存: 4°Cで輸送、-20°C (長期; -80°C) で保存。SLOは酸素に不安定であるが、20 mM cysteine や 10 mM DTT 等の SH 還元剤によって再活性化させることができる (文献2)。コレステロールによって不可逆的に溶血活性が失われる。

用途

- 1) 抗 SLO 抗体 (ASO) の力価測定のための抗原 (臨床検査薬) WB, Dot blot, ELISA
- 2) 細胞膜の透過剤 (生細胞に低分子から高分子まで導入可能: 文献1)
- 3) 生きた細胞質内の ATP の放出 (文献3)

活性測定: 3%ヒツジ赤血球を 37°Cで30分処理して50%溶血させる活性を1HUと定義

純度: SDS-PAGE (CBB 染色) で98%以上がSLO(図1)

性状: 1 mg/ml SLO in PBS (-), 1 mM DTT, 50% glycerol, フィルター滅菌済。SLOは容器に吸着してロスが多いので、タンパク質低吸着のシリコン加工したチューブ等を用いることを推奨する。

背景: ストレプトリジン O (SLO) は溶血レンサ球菌が菌体外に産生する膜傷害毒素であり、その膜傷害活性は赤血球の溶血により測定されている。SLOは酸素に不安定で容易に失活するが、チオール化合物により再活性化することからチオール活性化膜傷害毒素 (thiol-activated cytolysin) とも呼ばれている。SLOはA群溶血レンサ球菌だけでなく、C群やG群の菌株によっても産生され、これらのアミノ酸配列は高度に保存されており相同性は98%以上である。

本品は、C群溶血レンサ球菌のSLOを遺伝子組換え技術により大腸菌で大量発現させ、高度に精製したものである。比活性は1,900,000 hemolytic units (HU)/mgと高く、細胞膜上に大きな孔を形成するため、生細胞中にタンパク質を導入することが可能である (文献1)。成熟型SLOは60.4 kDaで、本品はタグがついていて64.5 kDaである。

データリンク Swiss-Prot [Streptolysin O](#)

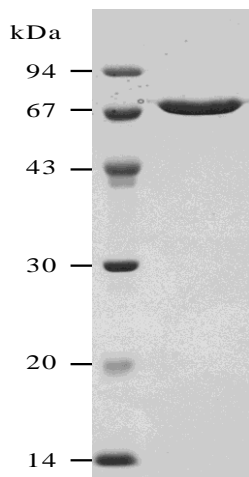


図1. SLOのSDS-PAGEによる解析

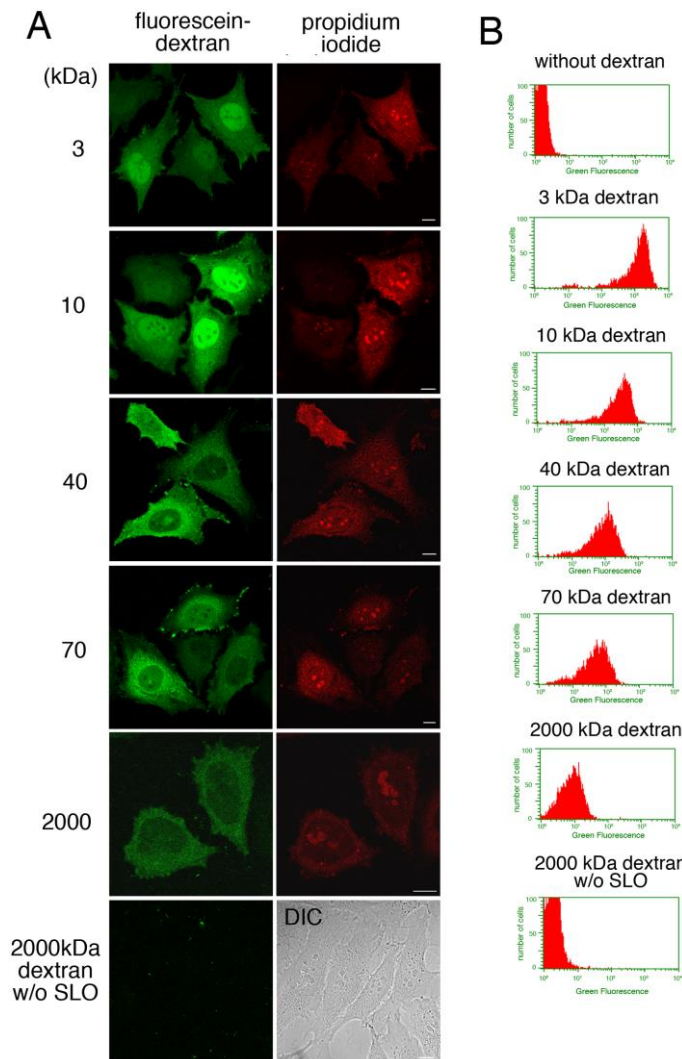


図2 種々の分子量の蛍光デキストランの再シールした細胞への導入

A. HeLa cells were incubated with or without (2000 kDa dextran w/o SLO) 0.13 $\mu\text{g/ml}$ SLO on ice for 5 min. After wash with PBS three times, the cells were further with transport buffer containing propidium iodide at 32°C for 5 min. Semi-intact HeLa cells were incubated with 1.5 mg/ml L5178Y cytosol, an ATP regenerating system, GTP, glucose, and 100 $\mu\text{g/ml}$ fluorescein-dextran of 3, 10, 40, 70, or 2000 kDa at 32°C for 15 min, and then were resealed by treatment with 1 mM CaCl_2 at 32°C for 5 min. After incubation with DMEM supplemented with FCS for 30 min, the cells were observed by confocal microscopy. Since the cells without SLO treatment did not contain the fluorescence of propidium iodide, differential interference contrast (DIC) image was shown. Bar = 10 μm . B. HeLa cells were treated as described in A, were trypsinized, and were subjected to flowcytometry. The histograms of fluorescein fluorescence of dextran with different molecular weight in PI-positive cells were shown.

Data from Kano F et al

A resealed-cell system for analyzing pathogenic intracellular events: perturbation of endocytic pathways under diabetic conditions. [PLoS One](https://doi.org/10.1371/journal.pone.0044127). 2012;7(8):e44127. doi: 10.1371/journal.pone.0044127. PMID::22952896

関連製品：[01-531](#) 抗ストレプトリジンO抗体、ウサギポリクロ

文献：本製品は文献3-12に使用されています。

1. Waley, I. *et al.* "Delivery of proteins into living cells by reversible membrane permeabilization with streptolysin-O." *PNAS* 98: 3185-3190 (2001) PMID: [11248053](#) 生きた細胞にタンパク質を導入する方法
2. Palmer, M. "The family of thiol-activated, cholesterol-binding cytolysins." *Toxicon* 39: 1681-1689 (2001) PMID: [11595631](#)

3. Maeda, Y. et al. GPHR is a novel anion channel critical for acidification and functions of the Golgi apparatus. *Nat. Cell Biol.* 10: 1135-45 (2008) PMID: [18794847](#) **Permeabilization of cells.**
4. Furukawa K. et al. Reduction-triggered fluorescent amplification probe for the detection of endogenous RNAs in living human cells. *Bioconj Chem.* 2009 May 20;20(5):1026-36. PMID: [19374406](#)
Introduction of probes for RNA into permeabilized human HL cells
5. Thiery J. et al. Perforin activates clathrin- and dynamin-dependent endocytosis, which is required for plasma membrane repair and delivery of granzyme B for granzyme-mediated apoptosis. *Blood* 2010 115:1582-1593. PMID: [20038786](#) **Promotion of endocytosis**
6. Kano F. et al. Hydrogen peroxide depletes phosphatidylinositol-3-phosphate from endosomes in a p38 MAPK-dependent manner and perturbs endocytosis. *Biochim Biophys Acta.* 2011 May;1813(5):784-801. PMID: [21277337](#). **Permeabilization of HeLa cells.**
7. Potez S. et al. Tailored protection against plasmalemmal injury by annexins with different Ca²⁺ sensitivities. *J Biol Chem.* 2011 May 20;286(20):17982-91. PMID:[21454475](#) **Permeabilization of HEK cells.**
8. Kano F. et al. A resealed-cell system for analyzing pathogenic intracellular events: perturbation of endocytic pathways under diabetic conditions. *PLoS One.* 2012;7(8):e44127. PMID: [22952896](#) **Introduction of molecules into HeLa cells.**
9. Imai A. et al. MADD/DENN/Rab3GEP functions as a guanine nucleotide exchange factor for Rab27 during granule exocytosis of rat parotid acinar cells. *Arch Biochem Biophys.* 2013 Aug 1;536(1):31-7. PMID: [23702376](#). **Introduction of antibody into cell**
10. Gao N and Lehrman MA. Mannose-6-Phosphate: A Regulator of LLO Destruction. : Inka Brockhausen (ed.), *Glycosyltransferases: Methods and Protocols*, 2013, Springer, Methods in Molecular Biology, vol. 1022, DOI 10.1007/978-1-62703-465-4_20. Link: springer.com/protocol/10.1007 **Introduction of mannose-6-phosphate into living cells.** (The authors specifically recommend BioAcademia streptolysin O for cell permeabilization)
11. Matsuto M et al. Reconstitution of the targeting of Rab6A to the Golgi apparatus in semi-intact HeLa cells: A role of BICD2 in stabilizing Rab6A on Golgi membranes and a concerted role of Rab6A/BICD2 interactions in Golgi-to-ER retrograde transport. *Biochim Biophys Acta.* 2015 Oct;1853(10 Pt A):2592-609. PMID: [25962623](#) **Introduction of protein (Rab6A) into permeabilized HeLa cells.**
12. Yasuga H. et al. Logic gate operation by DNA translocation through biological nanopores. *PLoS One.* 2016 Feb 18;11(2):e0149667. PMID: [26890568](#) **Nanopore formation in bilayer lipid membrane.**

本品は研究用にのみご使用ください。ヒトには用いないで下さい。本品は人体に危険（マウス；LD50 8 ug/kg 静注）ですので、エキスパートが取り扱って下さい。MSDS 詳細は次ページ（英文）

Material Safety Data Sheet

Product name: Streptolysin O of group C streptococcus expressed as His6-tagged recombinant protein in E. coli and highly purified (>95%)

MSDS Date: May 3, 2012

MSDS Number: 01-531

Responsible Party

Company Name: BioAcademia Inc.

Address: North Building 3F, Research Institute for Microbial Diseases,

Osaka University, 3-1 Yamadaoka, Suita, Osaka, 565-0871, Japan

Phone: +81-6-6877-2335

Information about health hazards

Target: Cholesterol on human and animal cell membrane

Health Hazards: May be fatal if enters bloodstream.

LD50 - Lethal dose (50 percent kill) intravenous,

Rabbit, 1500 ng/kg (Ref :PHTHDT Pharmacology and Therapeutics. (Pergamon Press Ltd., Headington Hill Hall, Oxford OX3 0BW, UK) Vol.(Issue) 11, Page 661,1981)

Guinea pig, 12 ug/kg (Ref: BICMBE Biochimie. (SPPIF, B.P.22, F-41353 Vineuil, France, Vol.(Issue)55,Page 1187, 1973)

Toxicity is much less when introduced via other routes of entry like Interdermal injection

First Aid Measures

Ingestion: Wash out with large amount of water. When swallowed, get medical attention if any discomfort arises.

Eye contact: Wash with large amounts of water while lifting eye lids. Call medical doctor if irritation continues.

Skin contact: Wash off with soap and plenty of water.

Spill release: Wear glove and sweep up the spill and then wash spill site. All the contaminants should be autoclaved at 121°C for 20 min before disposal.

Handling and Storage

Handling and Storage Precautions: BIOHAZARD. DO NOT USE IF SKIN IS CUT OR SCRATCHED.

Other Precautions: CAUTION: SUBSTANCE NOT YET FULLY TESTED.

Exposure Controls/Personal Protection

Protective Gloves: COMPATIBLE CHEMICAL-RESISTANT GLOVES.

Eye Protection:ANSI APPROVED CHEMICAL WORKERS GOGGLES .

Other Protective Equipment:EYE WASH AND DELUGE SHOWER MEETING ANSI
DESIGN CRITERIA .

Work Hygienic Practices:WASH THOROUGHLY AFTER HANDLING.

Disposal Considerations

Waste Disposal Methods: Autoclave the waste at 121°C for 20 min.

Regulatory Information

Federal Regulatory Information:EUROPEAN INFORMATION: CAUTION; SUBSTANCE
NOT YET FULLY TESTED.