

## **Certificate of Analysis**

Name: Streptolysin O (Hemolytic Streptococcus), Active Product Code: REC31958-20 Batch #: Date of Manufacture: Product Description: Recombinant streptolysin O, highly purified from E. coli over-expressing SLO of Group C hemolytic Streptococci (6xHis-tagged to the signal peptide removed N-terminal of SLO). Functional in membrane pore formation to introduce molecules into living animal cells. Expression System: E. coli Accession: Q54114 (TACY\_STREQ) **Amount:** 0.02 mg **Concentration:** 1.0 mg/ml Purity: >98% by SDS-PAGE Presentation: Liquid Buffer: PBS (-), 1 mM DTT, 50% glycerol, sterilized by filtration. No additive nor carrier protein. Applications: Functional studies. Reagent for membrane pore formation to introduce small-tomacromolecules into living cells. Antigen for measurement of anti-streptolysin O antibody (ASO), ELISA, Western blotting, Dot blotting, Immuno-chromatography, SDS-PAGE. Health & Safety: This product is an active protein and may elicit a biological response in vivo, handle with caution.

<u>Usage Guidelines</u> Short Term Storage: -20°C Long Term Storage: -80°C Stability: Not determined Freezing: Can be frozen



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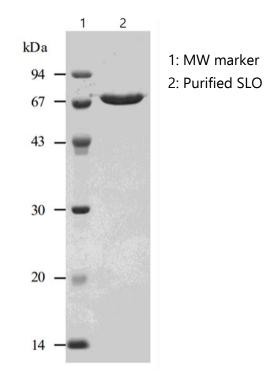
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**SDS-PAGE:** Purified streptolysin O (SLO) analysed by SDS-PAGE.





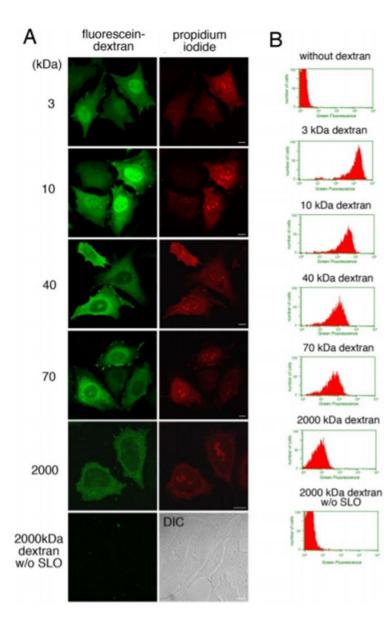
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## Introduction of fluorescein dextran of different molecular weights into resealed cells.

A. HeLa cells were incubated with or without (2000 kDa dextran w/o SLO) 0.13 µg/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 µg/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 CaCl2 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 =  $10 \mu m$ .

**B.** HeLa cells were treated as described in A, were trypsinized, and were subjected to flow cytometry. The histograms of fluorescein fluorescence of dextran with different molecular weight in PI-positive cells were shown.

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