

Diphtheria Toxin, functional

01-517 200 μg

Shipping and Storage temperature: Shipp with ice pack or dry ice and store at -20°C or at -80°C (long period).

Applications:

- 1) Inhibition of protein synthesis in eucaryotic cells
- Negative selection agent for ES cells in construction of transgenic mouse (Yagi, T. *et al.* "A novel negative selection for homologous recombinants using diphtheria toxin A fragment gene." *Anal. Biochem.* 214:77-86,1993 PMID: <u>8250258</u>)
- 3) Putative drug for treatment of malignant tumors such as leukemia
- 4) Antigen for Western blotting and ELISA
- 5) As a marker of Diphtheria toxin in SDS-PAGE

Form: 2~5 mg/ml (depending on lot) in 20 mM Tris-HCl (pH 7.2), 150 mM NaCl, 10% Glycerol Purity: More than 95% purity by SDS-PAGE (see below; SDS-PAGE without mercaptoethanol) Activity test: Addition of 20 ~30 pg/ml of Diphtheria toxin in growth medium caused 50% lethality after 43 h in Vero cells. Nicking of Diphtheria toxin by trypsin by trypsin and reduction by dithiothreitol further increase the activity.

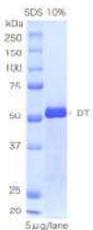
Background: This Diphtheria toxin was highly purified from the growth media of *Corynebacterium diphtheriae* strain PW8 as mostly unnicked form. Diphtheria toxin is a single polypeptide chain of 535 amino acids (58 kD) and nicked by cellular protease like furin to give fragments A (N-terminal, 21 kDa) and B (C-terminal, 37 kDa) which are linked by disulfide bridges. Binding to the cell surface of frgment B allows fragment A to penetrate the host cell. Fragment A catalyzes the ADP-ribosylation of eucaryotic elongation factor-2 (eEF2) by using NAD as a substrate, thus inactivating eEF2 and inhibiting protein synthesis.

Data Link: UniProtKB/TrEMBL Q5PY51 (Q5PY51_CORDI)

References:

 Uchida T *et al*' Diphtheria toxin and related proteins. 3. Reconstitution of hybrid "diphtheria toxin" from nontoxic mutant proteins." *J Biol Chem* 248:3851-3854 (1973) PMID: <u>4196586</u>

2. Pappenheimer A "Diphtheria toxin." *Annu Rev Biochem* **46**:69-94 (1977) PMID: 20040



* For research use only, not for human use.
*MSDS (Material Safety Data Sheet) is in the next page.

Fig.1 SDS-PAGE of Diphtheria toxin (DT)



Material Safety Data Sheet Diphtheria Toxin

Harzardous Ingradient

On a weight basis, Diphtheria toxin constitutes >95% of the total mass in 1 mg/ml protein solution.

Health Hazard Data

The MLD (Minimal Lethal Dose) in human is $\leq 100 \text{ ng/kg}$ when injected intramuscularly in an unimmunized adult.

Emergency Procedure

The toxin is less effective when it is administered orally, because it is unstable at acid pH in stomach. If skin pricking occurs accidentally, bleed and perform vigorous flushing of the area with large amounts of water. If injection occurs, seek a physician' attention immediately.

Handling

It should be handled carefully by persons with expertise in knowledge and techniques for the safe handling of Diphtheria toxin. Avoid mouth pipetting. Wear protective gloves when handling the toxin. Avoid contact with open wounds. Wash thoroughly any area of the body that makes contact with the toxin. It is recommended that persons who handle the toxin are immunized by diphtheria vaccine.

Inactivation

The toxin can be inactivated by exposing acids below pH 1 or pH above 12, followed by boiling for 30 min.