

Product Specification Sheet

Bacillus anthracis Edema Factor (EF) Recombinant Protein

Cat. # EF25-R

Bacillus anthracis EF Recombinant Protein

SIZE: 50 ug

After inhalation by mammals, *Bacillus anthracis* spores germinate in alveolar macrophages then migrate to lymph nodes where they multiply. The vegetative bacteria excrete the tripartite exotoxin which consists of three polypeptides: protective antigen (PA, 83 kDa), lethal factor (LF, 90 kDa) and edema factor (EF, 89 kDa). The two components (OF/EF and LF) of the toxin enzymatically modify substrates within the cytosol of the mammalian cells: The **OF/EF** is an adenylate cyclase that impairs the host defenses through a variety of mechanisms inhibiting phagocytosis. The **LF** is a zinc dependent protease that cleaves several mitogen activated protein kinase kinases (**MAPKK**) and causes lysis of macrophages. To intoxicate mammalian cells, the third component of the toxin **PA**, binds to a ubiquitously expressed cellular receptor, Tumor Endothelium Marker-8 (**TEM8**). Upon binding to TEM8, PA is cleaved into 20 and 63kDa fragments (PA20 and PA63) by furin or furin-like proteases. PA20 dissociates into medium and allows the PA63 fragment to heptamerize and bind LF and OF of the toxin. The resulting complex of **PA63** fragment with EF and/or OF binds to PA-receptor TEM8/ATR and internalized into endosomes followed by translocation of LF and OF into cytosol of the cells.

B. anthracis Edema Factor (EF) is one of the three proteins that comprise anthrax toxin, the others being lethal factor (LF), and protective antigen (PA). Edema factor is a calmodulin-dependent adenylate cyclase that is responsible for the swelling in cutaneous anthrax infections. The cellular cAMP levels increases in host cells, since the edema factor forms a complex with the host calmodulin which stimulates the activity of EF. The EF also inhibits the immune response by removing calmodulin from involvement in calcium triggered signaling.

Source of Protein

Recombinant EF was expressed and purified from *E. coli* using proprietary techniques (Purity: Single major band by SDS-PAGE, Mol wt ~90 kDa). It is provided as 50ug lyophilized solid. The lyophilized products should be reconstituted with 1.0 ml of sterile distilled water. It is recommended to add 0.1% BSA to enhance protein stability. It can then be used or aliquoted for storage in small aliquots at -70oC or below.

Endotoxin concn has been determined to be <3 EU/mg.

Biological activity

Recombinant EF biological activity is tested in CHO cells by testing cAMP stimulation using 1 ug/ml EF.

Special handling

Due to the nature of this product, it is recommended that this product be used by trained personnel and appropriate precautions taken to avoid contamination.

General References Tobias S et al (2003) JBC 10.1074, D. Borden Lacy et al (2002) JBC Vol. 227, 3006-3010; Kristina Cunningham et al (2002) PNAS Vol.99, 7049-7053; Jeremy Mogridge et al (2002) PNAS Vol.99, 7045-7048.

Storage

Short-term: unopened, undiluted vials for less than a week at 4oC.

Long-term: at -20C or below in suitable aliquots after reconstitution. Do not freeze and thaw and store working, diluted solutions.

Stability: 6-12 months at -20oC or below.

Shipping: 4oC for solutions and room temp for powder.

MSDS: Anthrax proteins (LF, PA, and EF) are produced by plasmids from a non-sporulating avirulent strain of *Bacillus anthracis* which lacks both of the wild type plasmids, pX01 and pX02. This host makes none of the anthrax toxin components and no polyglutamate capsule. Thus, the anthrax toxin components produced are single, pure proteins lacking all other virulence factors. Individually, each protein is non-toxic and presents no hazard during normal laboratory use. However, normal GLP procedures should be observed when handling this product and all areas cleaned after usage.

**This product is for In vitro research use only.*

Related materials available from ADI

Antibodies: Anthrax receptors ATR11-A, ATR12-A, ATR31-A, Protective antigen A, Spore Antigen Edema factor etc.

EF25-R Recombinant Anthrax toxin Edema Factor protein
 LF11-M Monoclonal Anti-Anthrax Lethal factor antigen IgG #1
 LF12-M Monoclonal Anti-Anthrax Lethal factor antigen IgG #2
 LF12-MB Monoclonal Anti- (LF) protein IgG biotinylated
 LF15-R Purified Recombinant Anthrax Lethal Factor protein
 LFPI-4 Lethal factor protease Inhibitor-1, Cell permeable,
 LFPS-1 Lethal factor Protease Substrate 1,
 LFPS-2 Lethal factor Protease Substrate 2, pNA derivative
 LFPS-3 Lethal factor Protease Substrate 3, AMC derivative
 PA11-M Monoclonal Anti-Anthrax Protective antigen IgG # 1
 PA12-M Monoclonal Anti-Anthrax Protective antigen IgG # 2
 PA12-MB Monoclonal Anti- Protective antigen (PA) IgG biotinylated
 PA63-R Purified Recombinant Anthrax Protective Antigen (63kD)
 PA83-R Purified Recombinant Anthrax Protective Antigen (83 kD)
 SA12-M Monoclonal Anti- Spore extract antigen IgG # 2, aff pure
 SA13-M Monoclonal Anti-Anthrax Spore extract antigen IgG # 3

ELISA kits to detect anti-LF, Anti-PA, Anti-EF antibodies in mouse, goat immunized with the recombinant proteins or vaccines
 EF25-R 80125A