

Ricin Toxoid, Recombinant with N-Terminal Histidine Tag

Catalog No. NR-4477

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Contributor and Manufacturer:

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Product Description:

Ricin toxin is a glycoprotein that can be isolated from the seeds of the castor bean plant *Ricinus communis*.¹ Structurally, ricin toxin consists of two polypeptide chains, A and B, that are linked by a disulfide bond. The A chain of ricin toxin catalytically inactivates the eukaryotic 28S ribosomal RNA subunit resulting in the inhibition of protein synthesis and death of the cell.² The ricin toxin B chain is a galactose-specific lectin that mediates the binding and delivery of the toxin to target cells.^{3,4} The sequence of the *R. communis* gene for the ricin toxin precursor protein has been reported (GenBank: X03179).⁵

NR-4477 is a genetically inactivated ricin toxoid expressed in *Escherichia coli* and purified by metal affinity chromatography. Catalytic Glu and Arg residues in the toxin active site have been mutagenized to create a non-toxic protein. The protein is expressed recombinantly as a single 62 kDa polypeptide containing both the A and B subunits. It also has an additional amino-terminal histidine-tag sequence.

This toxoid substance is prone to aggregation. The material precipitates at neutral pH but remains soluble in basic buffer of pH 8 or higher. The protein migrates through SDS-PAGE gel as a mixture of monomer, dimer, trimer and higher order species. Ricin toxoid is non-toxic. The predicted amino acid sequence of NR-4477 is shown in Table 1.

Material Provided:

Each vial of NR-4477 contains approximately 500 µg of ricin toxoid suspended in PBS, pH 8.4. The concentration, expressed as mg per mL, is shown on the Certificate of Analysis.

Packaging/Storage:

NR-4477 was packaged aseptically in plastic cryovials. The product is provided frozen on dry ice and should be stored at -20°C or colder immediately upon arrival. For long term storage, the contributor recommends -80°C or colder.

Functional Activity:

NR-4477 reacts specifically with monoclonal antibodies to ricin A chain (BEI Resources NR-843) as determined by western blot analysis.

Citation:

Acknowledgment for publications should read "The following reagent was obtained through BEI Resources, NIAID, NIH: Ricin Toxoid, Recombinant with N-Terminal Histidine Tag, NR-4477."

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publication: U.S. Department of Health and Human Services, Public Health Service, Centers for Disease Control and Prevention, and National Institutes of Health. Biosafety in Microbiological and Biomedical Laboratories, 5th ed. Washington, DC: U.S. Government Printing Office, 2009; see www.cdc.gov/biosafety/publications/bmbl5/index.htm.

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References:

1. Doan, L. G. "Ricin: Mechanism of Toxicity, Clinical Manifestations, and Vaccine Development. A Review." J. Toxicol. Clin. Toxicol. 42 (2004): 201–208. PubMed: 15214627.
2. Endo, Y. and K. Tsurugi. "RNA N-Glycosidase Activity of Ricin A-Chain. Mechanism of Action of the Toxic Lectin Ricin on Eukaryotic Ribosomes." J. Biol. Chem. 262 (1987): 8128–8130. PubMed: 3036799.
3. Chang, M. S., et al. "Cloning and Expression of Recombinant, Functional Ricin B Chain." Proc. Natl. Acad. Sci. U.S.A. 84 (1987): 5640–5644. PubMed: 3112772.
4. Olsnes, S., E. Saltvedt, and A. Pihl. "Isolation and Comparison of Galactose-binding Lectins from *Abrus precatorius* and *Ricinus communis*." J. Biol. Chem. 249 (1974): 803–810. PubMed: 4811904.
5. Halling, K. C., et al. "Genomic Cloning and Characterization of a Ricin Gene from *Ricinus communis*." Nucleic Acids Res. 13 (1985): 8019–8033. PubMed: 2999712. GenBank: X03179.

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Table 1 – Predicted Protein Sequence

1	MRGSHHHHHH	TDPM	I	FPKQY	PIINF	TTAGA	TVQSY	TNFIR	AVRGRL	TTGA
51	DVRHEIPVLP	NRVGLPINQR	FILVELSNHA	ELSVTLALDV	TNAYVVG	GYRA				
101	GNSAYFFHPD	NQEDAEAITH	LFTDVQNR	YTFAFGGNYDRL	EQLAGNLREN					
151	IELGNPLEE	AISALYYYST	GGTQLPTLAR	SFIICIQMIS	AAASFQYIEG					
201	EMRTRIRYNR	RSAPDPSVIT	LENSWGRLST	AIQESNQGAF	ASPIQLQRRN					
251	GSKFSVYDVS	ILIPIIALMV	YRCAPPPSSQ	FLLIRPVVP	NFNADVCM	DP				
301	EPIVRIVGRN	GLCVDVRDGR	FHNGNAIQLW	PCKSNTDANQ	LWTLKRDNTI					
351	RSNGKCLTTY	GYSPGVYVMI	YDCNTAATDA	TRWQIWDNGT	IINPRSSLVL					
401	AATSGNSGTT	LTVQTNIYAV	SQGWLPTNNT	QPFVTTIVGL	YGLCLQANS	G				
451	QVWIEDCSSE	KAEQQWALYA	DGSIRPQQNR	DNCLTSDSNI	RETVVKILSC					
501	GPASSGQRWM	FKNDGTILNL	YSGLVLDVRA	SDPSLKQIIL	YPLHGDPNQI					
551	WLPLF									

Amino-terminal histidine-tag and plasmid amino acids
Mutagenized catalytic Glu and Arg residues (E191A and R194S)