Human soluble TNF Receptor Inhibitor/Fc chimera (TNFRSF1A)

Certificate of Analysis and Data Sheet

Source:	**Catalog No.**
Mouse Myeloma Cell Line (NSO)	CTK-309

Background:

Human soluble TNF Receptor Inhibitor/Fc chimera (sTNF RI/Fc chimera) is recombinant human soluble TNF Receptor Inhibitor fused with the Fc part of human IgG1. The recombinant mature sTNF RI/Fc is a disulfide-linked homodimeric protein. This dimeric human TNF RI/Fc has been shown to be an approximately 45-fold more potent inhibitor of TNF-a than the monomeric soluble TNF RI. Soluble TNF RI are truncated forms of high-affinity cell surface receptors, corresponding to TNFR-p60 Type B. Two types of soluble TNF receptors, TNFR-p60 Type B and TNFR-p80 Type A, have been identified in human serum and urine, which are shared by TNF-a and TNF-b and can neutralize the biological activities of TNF-a and TNF-b. In the new TNF superfamily nomenclature, TNF RI is referred to as TNFRSF1A. These apparent soluble forms of the receptors appear to arise as a result of shedding of the extracellular domains of the membrane-bound receptors. Although the physiological role of these proteins is not fully understood, it has been speculated that shedding of the soluble receptors in response to TNF release could serve as a mechanism to scavenge the TNF not immediately bound and thus localize the inflammatory response. It is also possible that the pool of TNF bound to soluble receptors could represent a reservoir for the controlled release of TNF.

Description :

Recombinant Human soluble TNF-RI produced in NSO is a disulfide linked homodimer, glycosylated, polypeptide chain having a molecular mass of 48Kda but as a result of the glycosylation, the recombinant protein migrates at 55-60Kda protein in SDS-PAGE. TNF-RI /Fc chimera (sTNF RI/Fc chimera) was fused to carboxy-terminal 6X histidine-tagged Fc part of human IgG1, via a linker peptide. Recombinant soluble TNFRI is purified by proprietary chromatographic techniques.

Physical Appearance:

Sterile Filtered White lyophilized (freeze-dried) powder.

Formulation:

The lyophilized protein (1mg/ml) was lyophilized from sterile solution containing PBS.

Solubility:

It is recommended that sterile PBS containing at least 0.1% human serum albumin or bovine serum albumin be added to the vial to prepare a stock solution of no less than 100 mg/mL.

Stability:

Lyophilized TNF-RI although stable at room temperature for 3 weeks, should be stored desiccated below -18° C. Upon reconstitution TNF-RI should be stored at 4° C between 2-7 days and for future use below -18° C. For long-term storage it is recommended to add a carrier protein (0.1% HSA or BSA).
Please avoid freeze-thaw cycles.

Purity:

Greater than 95.0% as determined by:
(a) Analysis by RP-HPLC.
(b) Anion-exchange FPLC.
(c) Analysis by reducing and non-reducing SDS-PAGE Silver Stained gel.

Amino acid sequence:

The sequence of the first five N-terminal amino acids was determined and was found to be Met-Glu-Glu-Val-Ser.

Dimers and aggregates:

Less than 1% as determined by silver-stained SDS-PAGE gel analysis.

Biological Activity:

This soluble TNF-RI is fully biologically active when compared to standard. The activity was measured by its ability to neutralize apoptosis of mouse L929 cells treated with 0.25 ng/mL rhTNF-a. The ED50 for this effect is typically 0.4-1 ng/mL.

Endotoxin:

Less than 0.1 ng/μg (IEU/μg) of TNF-RI.

Protein content:

Protein quantitation was carried out by two independent methods:

1. UV spectroscopy at 280.
2. Analysis by RP-HPLC, using a standard solution of TNF-RI as a Reference Standard.

Usage:

This material is offered for research, laboratory or further evaluation purposes.