

General Information

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| Synonyms | Human Tumor Necrosis Factor alpha, rTNFA; TNF-A; TNFalpha |
| Accession # | P01375 |
| Source | Human embryonic kidney cell, HEK293-derived human TNF-alpha protein |
| | Val77-Leu233 |
| Predicted Molecular weight | 17.4 kDa (Monomer) |
| Form/Structure | Trimer in solution |

Components and Storage

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| Formulation | Solution protein. |
| | Dissolved in sterile PBS buffer. |
| | This solution can be diluted into other aqueous buffers. Centrifuge the vial prior to opening. |

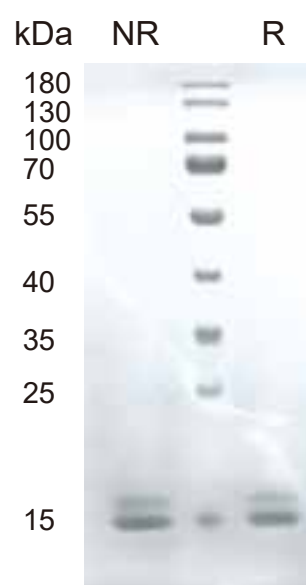
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| Storage and Stability | Avoid repeated freeze-thaw cycles. |
| | It is recommended that the protein be aliquoted for optimal storage. |
| | 12 months from date of receipt, -20 to -70 °C as supplied. |

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| Shipping | Shipping with dry ice. |
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Quality

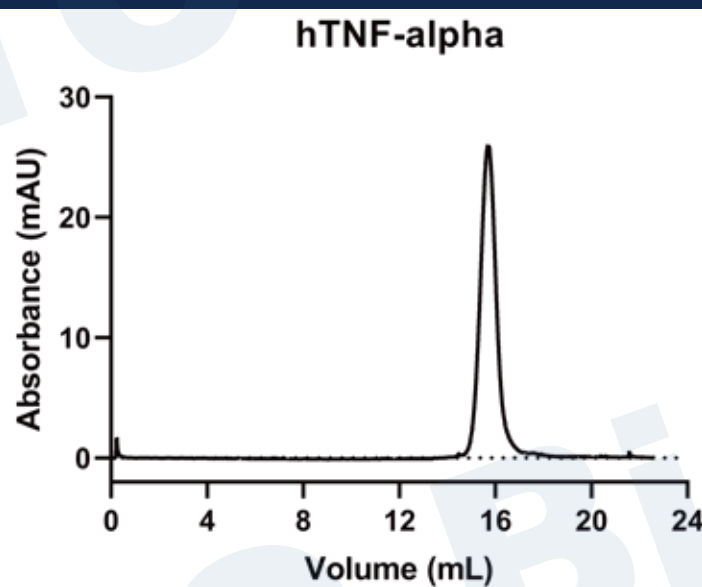
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| Purity | > 95%, determined by SDS-PAGE. |
| Endotoxin Level | <0.010 EU per 1 ug of the protein by the LAL method. |
| Activity | Measured in a cytotoxicity assay using L-929 mouse fibroblast cells in the presence of the metabolic inhibitor actinomycin D. The ED ₅₀ for this effect is 20-100 pg/mL. |

SDS-PAGE



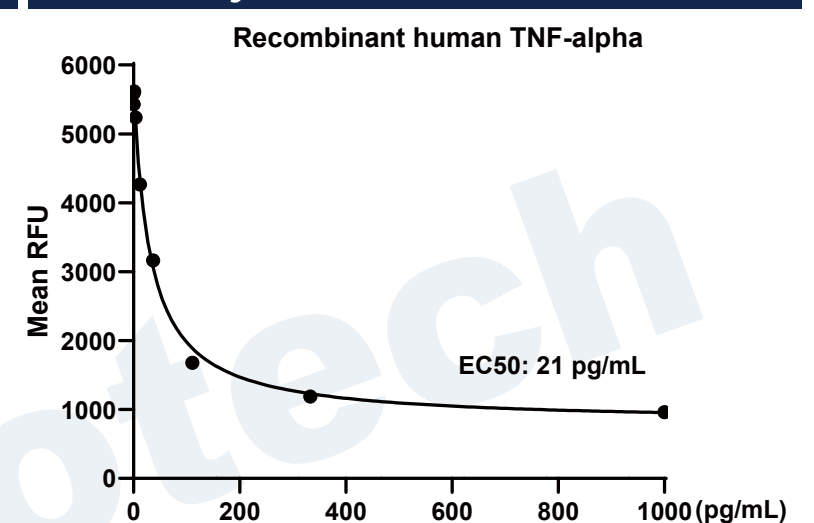
2 ug/lane protein was resolved with SDS-PAGE under non-reducing (NR) and reducing (R) conditions and visualized by Coomassie Blue staining.

Gel filtration



Size-exclusion chromatography of recombinant human TNF-alpha protein (280 nm absorbance)

Bioactivity



Recombinant human TNF-alpha (Catalog # HF-2015) induces cytotoxicity in the L-929 mouse fibroblast cell line in the presence of the metabolic inhibitor actinomycin D.

Background

Tumor necrosis factor alpha(TNF-alpha), is a pleiotropic pro-inflammatory cytokine secreted by various cells, including adipocytes, activated monocytes, macrophages, B cells, T cells and fibroblasts (1,2). It belongs to TNF family of ligands, and signals through two receptors, TNFR1 and TNFR2. Human TNF-alpha consists of a 35 amino acid (aa) cytoplasmic domain, a 21 aa transmembrane segment, and a 177 aa extracellular domain (ECD) (3). The ECD of human TNF-alpha shares 97% aa sequence identity with rhesus and 71%-92% with bovine, canine, cotton rat, equine, feline, mouse, porcine, and rat TNF-alpha. TNF-alpha is assembled intracellularly to form a noncovalently linked homotrimer which is expressed on the cell surface (4). Cell surface TNF-alpha can induce the lysis of neighboring tumor cells and virus infected cells, and it can generate its own downstream cell signaling following ligation by soluble TNFR1 (2, 5). Shedding of membrane bound TNF-alpha by TACE/ADAM17 releases the bioactive cytokine, a 55 kDa soluble trimer of the TNF-alpha extracellular domain (6-8). TNF-alpha binds the ubiquitous 55-60 kDa TNFR1(9, 10) and the hematopoietic cell-restricted 80 kDa TNFR2 (11, 12), both of which are also expressed as homotrimers (1, 2, 13).

Reference

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| 3. Pennica, D. et al. (1984) Nature 312:724. | 10. Loetscher, H. et al. (1990) Cell 61:351. |
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| 5. Perez, C. et al. (1990) Cell 63:251. | 12. Smith, C.A. et al. (1990) Science 248:1019. |
| 6. Black, R.A. et al. (1997) Nature 385:729. | 13. Loetscher, H. et al. (1991) J. Biol. Chem. 266:18324. |
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