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Product Information

Anti-Tumor Necrosis Factor Soluble Receptor I
produced in goat, affinity isolated antibody

Catalog Number **T2190**

Product Description

Anti-Tumor Necrosis Factor Soluble Receptor I (TNF sRI) is produced in goat using as immunogen recombinant mouse tumor necrosis factor soluble receptor I, expressed in *E. coli* (Gene ID 21937). The antibody is purified using mouse TNF RI affinity chromatography.

Anti-Tumor Necrosis Factor Soluble Receptor I (TNF sRI) may be used to detect mouse TNF sRI by immunoblotting. By immunoblotting, the antibody shows < 5% cross-reactivity with recombinant human TNF sRI. The antibody can interact with the cell surface mouse TNF RI (55 kDa) and exhibits TNF agonist activities on the mouse cell line L929.

TNF RI (CD120a) is a 55 kDa transmembrane glycoprotein that is expressed by virtually all nucleated mammalian cells.[1-3] Among the numerous cells known to express TNF RI are hepatocytes,[4] monocytes and neutrophils,[5] cardiac muscle cells,[6] endothelial cells,[7] and $CD34^+$ hematopoietic progenitors.[8] Both TNF-α and TNF-β bind to TNF RI. Soluble TNF-α binds with a Kd in the range of 20-60 pM,[9, 10] while TNF-β binds with a Kd equal to 650 pM.[9] TNF RI relative to TNF RII seems to be the more physiologically-relevant receptor, whereas TNF-R2 appears to play a direct role in only a limited number of TNF responses.[11,12] Soluble TNF RI, which blocks TNF-α activity, has been identified in both urine and blood (1-3 ng/mL).[4, 13, 14] Serum levels of sTNF receptors increase dramatically in certain pathological situations. Soluble forms of two molecular weights (p60 and p80) have been identified and are believed to be generated by proteolytic cleavage.[3, 15-17]

Human and mouse TNF RI have 64% amino acid sequence identity (70% in the extracellular region). Human TNF RI binds human and mouse TNF-α with equal affinity.[18, 19]

The extracellular region of TNF RI has four cysteine-rich motifs, the first of which is suggested to be required for binding.[9] The intracellular portion of TNF RI contains a “death domain” of about 70 amino acids that is required for the signaling of apoptosis and NF-6B activation.[20,21] TNF binds to the extracellular domain of TNF RI and induces receptor trimerization.[22] Then, the aggregated death domain of TNF RI recruits the adapter protein TRADD.[21] TRADD, in turn, recruits FADD, TRAF2 and RIP to form the TNF RI signaling complex and activate signaling cascades leading to apoptosis,[23, 24] JNK/SAPK activation,[23, 25] and NF-6B activation[26, 27] respectively. TNF RI self-associates and signals independently of ligand when overexpressed. This apparent paradox may be explained by silencer of death domains (SODD), a widely expressed ~60 kDa protein that was found to be associated with the death domain of TNF RI.[28]

Reagent

Supplied lyophilized from a 0.2 μm filtered solution in phosphate buffered saline containing 5% trehalose.

Precautions and Disclaimer

This product is for R&D use only, not for drug, household, or other uses. Please consult the Material Safety Data Sheet for information regarding hazards and safe handling practices.

Preparation Instructions

To one vial of lyophilized powder, add 0.5 mL of 0.2 μm-filtered PBS to produce a 0.2 mg/mL stock solution of antibody. If aseptic technique is used, no further filtration should be needed for use in cell culture environments.

Storage/Stability

Prior to reconstitution, store at –20 °C. Reconstituted product may be stored at 2-8 °C for at least one month. For prolonged storage, freeze in working aliquots at –20 °C. Avoid repeated freezing and thawing.

Product Profile
The antibody is tested for its agonist activity in a cytotoxicity assay in the presence of the metabolic inhibitor, actinomycin D, using mouse L929 cells.[28]

Immunoblotting: a working concentration of 0.1-0.2 µg/mL is determined using recombinant mouse TNF RI at 1 ng/lane under non-reducing conditions and 5 ng/mL under reducing conditions.

Immunohistochemistry: a working concentration of 5-15 µg/mL is determined to detect TNF RI/TNFRSF1A in perfusion fixed frozen sections of mouse intestine.

Flow Cytometry: a working concentration of 2.5 µg/10^6 cells is determined to detect TNF RI/TNFRSF1A in mouse L929 cells.

Note: In order to obtain the best results in various techniques and preparations, we recommend determining optimal working dilutions by titration.

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