Product Information Sheet

Human Soluble TNFR Ⅱ ELISA Kit

Catalog No. EK0530

Size 96T

Range 7.8pg/ml-500pg/ml

Sensitivity < 2 pg/ml

Specificity

No detectable cross-reactivity with any other cytokine.

Storage

Store at 4℃ for frequent use, at -20℃ for infrequent use.

Avoid multiple freeze-thaw cycles (Shipped with wet ice.)

Expiration

Four months at 4℃ and eight months at -20℃.

Application

For quantitative detection of human sTNFR Ⅱ in sera, plasma, body fluids, tissue lysates or cell culture supernates.

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Principle

Human sTNFR Ⅱ ELISA Kit was based on standard sandwich enzyme-linked immune-sorbent assay technology. Human sTNFR Ⅱ specific-specific polyclonal antibodies were precoated onto 96-well plates. The human specific detection polyclonal antibodies were biotinylated. The test samples and biotinylated detection antibodies were added to the wells subsequently and then followed by washing with PBS or TBS buffer. Avidin-Biotin-Peroxidase Complex was added and unbound conjugates were washed away with PBS or TBS buffer. HRP substrate TMB was used to visualize HRP enzymatic reaction. TMB was catalyzed by HRP to produce a blue color product that changed into yellow after adding acidic stop solution. The density of yellow is proportional to the human sTNFR Ⅱ amount of sample captured in plate.

Kit Components

1. Lyophilized recombinant human sTNFR Ⅱ standard: 10ng/tube×2.
2. One 96-well plate precoated with anti- human sTNFR Ⅱ antibody.
3. Sample diluent buffer: 30 ml
4. Biotinylated anti- human sTNFR Ⅱ antibody : 130μl, dilution 1:100.
5. Antibody diluent buffer: 12ml.
6. Avidin-Biotin-Peroxidase Complex (ABC) : 130μl, dilution 1:100.
7. ABC diluent buffer: 12ml.
8. TMB color developing agent: 10ml.
9. TMB stop solution: 10ml.

Material Required But Not Provided

1. Microplate reader in standard size.
2. Automated plate washer.
3. Adjustable pipettes and pipette tips. Multichannel pipettes are recommended in the condition of large amount of samples in the detection.
4. Clean tubes and Eppendorf tubes.
5. Washing buffer (neutral PBS or TBS).

   Preparation of 0.01M **TBS:** Add 1.2g Tris, 8.5g Nacl; 450μl of purified acetic acid or 700μl of concentrated hydrochloric acid to 1000ml H_2O and adjust pH to 7.2-7.6. Finally, adjust the total volume to 1L.

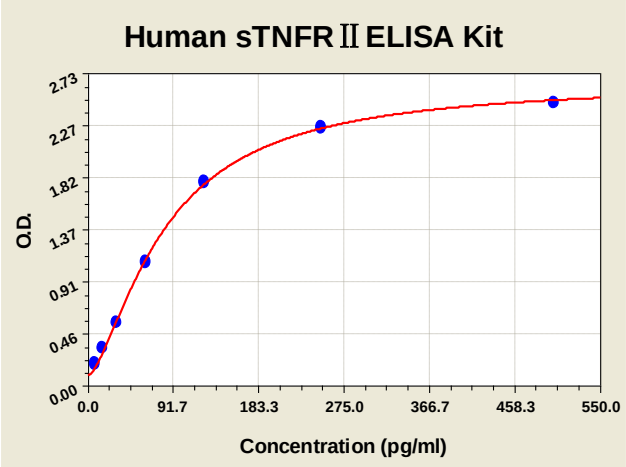
   Preparation of 0.01 M **PBS:** Add 8.5g sodium chloride, 1.4g Na_2HPO_4 and 0.2g NaH_2PO_4 to 1000ml distilled water and adjust pH to 7.2-7.6.

Finally, adjust the total volume to 1L.

Notice for Application of Kit

1. Before using Kit, spin tubes and bring down all components to bottom of tube.
2. Duplicate well assay was recommended for both standard and sample testing.
3. Don't let 96-well plate dry, dry plate will inactivate active components on plate.
4. In order to avoid marginal effect of plate incubation due to temperature difference (reaction may be stronger in the marginal wells), it is suggested that the diluted ABC and TMB solution will be pre-warmed in 37℃ for 30 min before using.

Human sTNFR Ⅱ ELISA Kit-1X96 Well Plate Image



Background

Tumor necrosis factor receptor p75 (TNF-R p75, or TNF-R2) is a 75-kDa type I transmembrane protein expressed predominantly on cells of hematopoietic lineage. TNF-R p75 belongs to the TNF receptor superfamily characterized by cysteine-rich extracellular regions composed of three to six disulfide-linked domains.[1] The tumor necrosis factor receptor Ⅱ (TNFR Ⅱ) gene localizes to 1p36. 2, a genomic region characteristically deleted in neuroblastomas and other malignancies. In addition, TNFR Ⅱ is the principal mediator of the effects of TNF on cellular immunity, and it may cooperate with TNFR Ⅰ in the killing of nonlymphoid cells.[2] The standard product used in this kit is recombinant human sTNFR Ⅱ, consisting of 24-206 amino acid sequence with the molecular mass of 20KDa.

Reference

1. Santee, S. M.; Owen-Schaub, L. B. Human tumor necrosis factor receptor p75/80 (CD120b) gene structure and promoter characterization. *J. Biol. Chem.* 271: 21151-21159, 1996.
2. Beltinger, C. P.; White, P. S.; Maris, J. M.; Sulman, E. P.; Jensen, S. J.; LePaslier, D.; Stallard, B. J.; Goeddel, D. V.; de Sauvage, F. J.; Brodeur, G. M. Physical mapping and genomic structure of the human TNFR2 gene. *Genomics* 35: 94-100, 1996.