Product Information Sheet

Rat IL-6 ELISA Kit

Catalog No. EK0412

Size 96T

Range 62.5pg/ml-4000pg/ml

Sensitivity < 5pg/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 rat IL-6 in sera, plasma, body fluids, tissue lysates or cell culture supernates.

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Principle

Rat IL-6 ELISA Kit was based on standard sandwich enzyme-linked immune-sorbent assay technology. Rat IL-6 specific-specific polyclonal antibodies were precoated onto 96-well plates. The rat 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 rat IL-6 amount of sample captured in plate.

Kit Components

1. Lyophilized recombinant rat IL-6 standard: 10ng/tube×2.
2. One 96-well plate precoated with anti- rat IL-6 antibody.
3. Sample diluent buffer: 30 ml
4. Biotinylated anti- rat IL-6 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 and Automated plate washer.
2. Adjustable pipettes and pipette tips. Multichannel pipettes are recommended in the condition of large amount of samples in the detection.
3. Clean tubes and Eppendorf tubes.
4. 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.

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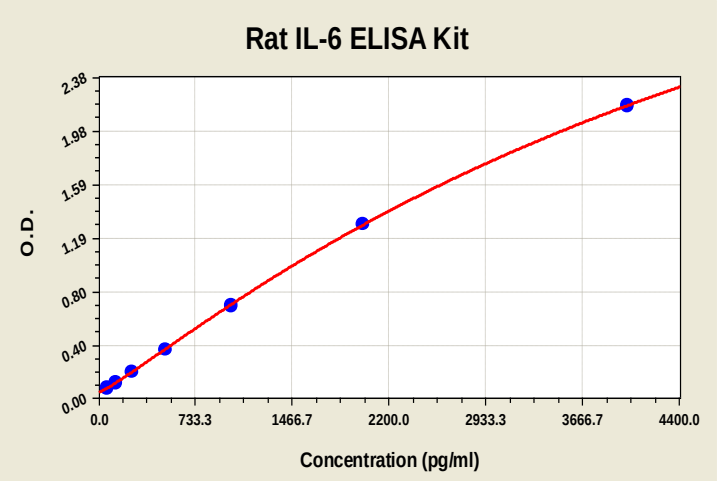
Notice for Application of Kit

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.

Rat IL-6 ELISA Kit-1X96 Well Plate Image



Background

Interleukin-6 (IL-6) is a protein that in humans is encoded by the IL6 gene.[1] Interleukin-6 (IL-6) is involved not only in the hepatic acute phase response but also in adipose tissue metabolism, lipoprotein lipase activity, and hepatic triglyceride secretion.[2] IL-6 is a pleiotropic cytokine that plays a critical role in bone resorption.[3] IL-6 has been suggested to stimulate the HPA axis during immune activation independent of the input of hypothalamic corticotropin-releasing hormone (CRH).[4] Interleukin-6 (IL-6) is an immunoregulatory cytokine that activates a cell-surface signaling assembly composed of IL-6, the IL-6 alpha-receptor (IL-6Ralpha), and the shared signaling receptor gp130.[5] The standard product used in this kit is recombinant rat IL-6, consisting of 188 amino acids with the molecular mass of 21.8kDa.

Reference

1. Ferguson-Smith AC, Chen YF, Newman MS, May LT, Sehgal PB, Ruddle FH (April 1988). "Regional localization of the interferon-beta 2/B-cell stimulatory factor 2/hepatocyte stimulating factor gene to human chromosome 7p15-p21". Genomics 2 (3): 203–8.
2. Fernandez-Real, J.-M., Broch, M., Vendrell, J., Richart, C., Ricart, W. Interleukin-6 gene polymorphism and lipid abnormalities in healthy subjects. J. Clin. Endocr. Metab. 85: 1334-1339, 2000.
3. Ferrari, S. L., Ahn-Luong, L., Garnero, P., Humphries, S. E., Greenspan, S. L. Two promoter polymorphisms regulating interleukin-6 gene expression are associated with circulating levels of C-reactive protein and markers of bone resorption in postmenopausal women. J. Clin. Endocr. Metab. 88: 255-259, 2003.
4. Venihaki, M., Dikkes, P., Carrigan, A., Karalis, K. P. Corticotropin-releasing hormone regulates IL-6 expression during inflammation. J. Clin. Invest. 108: 1159-1166, 2001.