Product Information Sheet

Rat IGF-1 ELISA Kit

Catalog No. EK0377

Size 96T

Range 62.5pg/ml-4000pg/ml

Sensitivity < 5pg/ml

Specificity

Detectable cross-reactivity with IGF-2 < 1%.

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 IGF-1 in sera, body fluids, tissue lysates or cell culture supernates.

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Principle

Antagene's rat IGF-1 ELISA Kit was based on standard sandwich enzyme-linked immune-sorbent assay technology. Rat IGF-1 specific-specific monoclonal 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 IGF-1 amount of sample captured in plate.

Kit Components

1. Lyophilized recombinant rat IGF-1 standard: 10ng/tube×2.
2. One 96-well plate precoated with anti- rat IGF-1 antibody.
3. Sample diluent buffer: 30 ml
4. Biotinylated anti- rat IGF-1 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.

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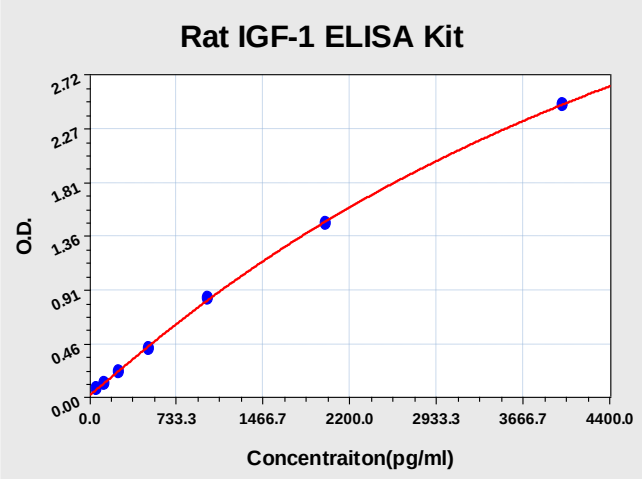
Notice for Application of Kit

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 IGF-1 ELISA Kit-1X96 Well Plate Image



Background

Insulin-like growth factor 1 (IGF-1) that was once called somatomedin C, is a polypeptide protein hormone similar in molecular structure to insulin. It plays an important role in childhood growth and continues to have anabolic effects in adults. Human IGF1 is a single chain 70-amino acid polypeptide cross-linked by 3 disulfide bridges, with a calculated molecular mass of 7.6 kD.[1] The IGF1 gene, mapped on 12q22-q24.1, contains 5 exons. Exons 1-4 encode the 195-amino acid precursor (IGF1B), and exons 1, 2, 3, and 5 encode the 153-residue peptide (IGF1A).[2] The structure of IGF1 resembles that of IGF2. And the IGF1 and IGF2 genes have complex structures with multiple promoters. The expression of both genes is regulated at the levels of transcription, RNA processing, and translation.[3] IGF-1 is produced primarily by the liver as an endocrine hormone as well as in target tissues in a paracrine/autocrine fashion. Moreover, approximately 98% of IGF-1 is always bound to one of 6 binding proteins (IGF-BP). Furthermore, IGF-1 is one of the most potent natural activators of the AKT signaling pathway, a stimulator of cell growth and multiplication and a potent inhibitor of programmed cell death.

Reference

1. Rinderknecht, E.; Humbel, R. E.: The amino acid sequence of human insulin-like growth factor I and its structural homology with proinsulin. J. Biol. Chem. 253: 2769-2776, 1978.
2. Rotwein, P.; Pollock, K. M.; Didier, D. K.; Krivi, G. G.: Organization and sequence of the human insulin-like growth factor I gene: alternative RNA processing produces two insulin-like growth factor I precursor peptides. J. Biol. Chem. 261: 4828-4832, 1986.
3. Sussenbach, J. S.; Steenbergh, P. H.; Holthuizen, P.: Structure and expression of the human insulin-like growth factor genes. Growth Regul. 2: 1-9, 1992.

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