



Product Information Sheet

Human EGFR ELISA Kit

Catalog No.	EK0327
Size	96T
Range	62.5pg/ml-4000pg/ml
Sensitivity	< 1pg/ml

Specificity

No detectable cross-reactivity with any other cytokine.

Storage

Store at 4°C for frequent use, at -20°C for infrequent use.

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

Expiration

Four months at 4°C and eight months at -20°C.

Application

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

Principle

Antagene's human EGFR ELISA Kit is based on standard sandwich enzyme-linked immune-sorbent assay technology. Human EGFR 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 EGFR amount of sample captured in plate.

Kit Components

1. Lyophilized recombinant human EGFR standard: 10ng/tubex2.
2. One 96-well plate precoated with anti- human EGFR antibody.
3. Sample diluent buffer: 30 ml
4. Biotinylated anti- human EGFR 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 H₂O and adjust pH to 7.2-7.6. Finally, adjust the total volume to 1L.

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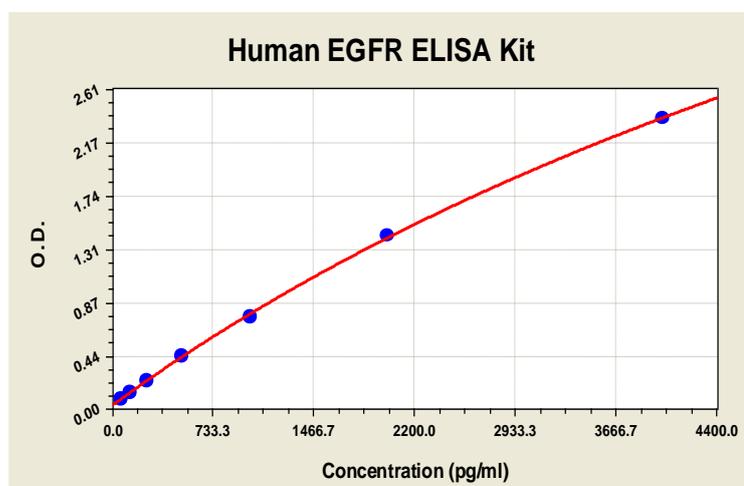
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Preparation of 0.01 M **PBS**: 1000ml distilled water and adjust pH to 7.2-7.6. Finally, adjust the total volume to 1L.
Add 8.5g sodium chloride, 1.4g Na_2HPO_4 and 0.2g NaH_2PO_4 to

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°C for 30 min before using.

Human EGFR ELISA Kit-1X96 Well Plate Image



Background

The epidermal growth factor receptor (EGFR; ErbB-1; HER1 in humans) is the cell-surface receptor for members of the epidermal growth factor family (EGF-family) of extracellular protein ligands.¹ It is a member of the ErbB family of receptors, a subfamily of four closely related receptor tyrosine kinases: EGFR (ErbB-1), HER2/c-neu (ErbB-2), Her 3 (ErbB-3) and Her 4 (ErbB-4). EGFR exists on the cell surface and is activated by binding of its specific ligands, including epidermal growth factor and transforming growth factor α (TGF α). EGFR and its ligands are cell signaling molecules involved in diverse cellular functions, including cell proliferation, differentiation, motility, and survival, and in tissue development.² Mutations that lead to EGFR overexpression (known as upregulation) or overactivity have been associated with a number of cancers, including lung cancer and glioblastoma multiforme. In this latter case a more or less specific mutation of EGFR, called EGFRvIII is often observed.³

Reference

1. Herbst RS (2004). "Review of epidermal growth factor receptor biology". *Int. J. Radiat. Oncol. Biol. Phys.* 59 (2 Suppl): 21–6.
2. Wang, K.; Yamamoto, H.; Chin, J. R.; Werb, Z.; Vu, T. H.: Epidermal growth factor receptor-deficient mice have delayed primary endochondral ossification because of defective osteoclast recruitment. *J. Biol. Chem.* 279: 53848-53856, 2004.
3. Kuan CT, Wikstrand CJ, Bigner DD (June 2001). "EGF mutant receptor vIII as a molecular target in cancer therapy". *Endocr. Relat. Cancer* 8 (2): 83–96.

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