



Product Information Sheet

Human ACE ELISA Kit

Catalog No.	EK0557
Size	96T
Range	156pg/ml-10,000pg/ml
Sensitivity	< 5 pg/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 ACE in sera, plasma, body fluids, tissue lysates or cell culture supernates.

Principle

Human ACE ELISA Kit was based on standard sandwich enzyme-linked immune-sorbent assay technology. Human ACE 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 ACE amount of sample captured in plate.

Kit Components

1. Lyophilized recombinant human ACE standard: 10ng/tubex2.
2. One 96-well plate precoated with anti- human ACE antibody.
3. Sample diluent buffer: 30 ml
4. Biotinylated anti- human ACE 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 if there is a large amount of samples for 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 H₂O 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 Na₂HPO₄ and 0.2g NaH₂PO₄ to 1000ml distilled water and adjust pH to 7.2-7.6. Finally, adjust the total volume to 1L.

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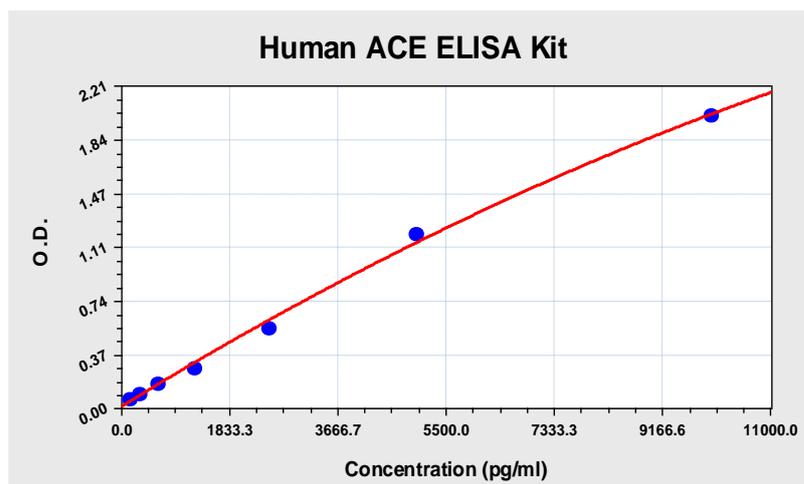
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Notice for Application of Kit

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°C for 30 min before using.

Human ACE ELISA Kit-1X96 Well Plate Image



Background

Angiotensin-converting enzyme (ACE) is a zinc-containing dipeptidyl carboxypeptidase widely distributed in mammalian tissues and is thought to play a critical role in blood pressure regulation. The predicted protein is identical, from residue 37 to its C terminus, to the second half or C-terminal domain of the endothelial ACE sequence. The protein sequence inferred consists of a 732-residue preprotein including a 31-residue signal peptide. The mature polypeptide has a molecular weight of 80,073.¹ Although ACE has been studied primarily in the context of its role in blood pressure regulation, this widely distributed enzyme has many other physiological functions. The ACE gene encodes two isozyms. The somatic isozyme is expressed in many tissues, including vascular endothelial cells, renal epithelial cells, and testicular Leydig cells, whereas the testicular or germinal angiotensin-converting enzyme is expressed only in sperm.² The standard product used in this kit is recombinant human ACE, consisting of 30-1261 amino acids with the molecular mass of 120KDa.

Reference

1. Ehlers, M. R. W.; Fox, E. A.; Strydom, D. J.; Riordan, J. F. Molecular cloning of human testicular angiotensin-converting enzyme: the testis isozyme is identical to the C-terminal half of endothelial angiotensin-converting enzyme. *Proc. Nat. Acad. Sci.* 86: 7741-7745, 1989.
2. Ramaraj, P.; Kessler, S. P.; Colmenares, C.; Sen, G. C. Selective restoration of male fertility in mice lacking angiotensin-converting enzymes by sperm-specific expression of the testicular isozyme. *J. Clin. Invest.* 102: 371-378, 1998.

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