



## Product Information Sheet

### Human HSP27 ELISA Kit

**Catalog No.** EK0881  
**Size** 96T  
**Range** 62.5pg/ml-4000pg/ml  
**Sensitivity** < 5pg/ml

**Specificity**

No detectable cross-reactivity with any other HSP .

**Storage**

Store at 2-8°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 HSP27 in sera, body fluids, tissue lysates or cell culture supernates.

**Principle**

Human HSP27 ELISA Kit was based on standard sandwich enzyme-linked immune-sorbent assay technology. Human HSP27 specific-specific monoclonal antibodies were precoated onto 96-well plates. The human specific detection monoclonal 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 HSP27 amount of sample captured in plate.

**Kit Components**

1. Lyophilized recombinant human HSP27 standard: 10ng/tubex2.
2. One 96-well plate precoated with anti- human HSP27 antibody.
3. Sample diluent buffer: 30 ml
4. Biotinylated anti- human HSP27 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<sub>2</sub>O 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<sub>2</sub>HPO<sub>4</sub>

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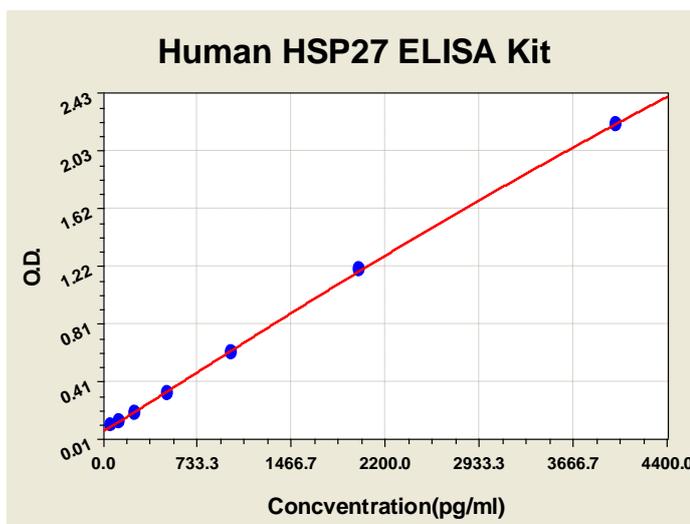
and 0.2g NaH<sub>2</sub>PO<sub>4</sub> to 1000ml  
distilled water and adjust pH to

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°C for 30 min before using.

## Human HSP27 ELISA Kit-1X96 Well Plate Image



## Background

**Hsp27** is a chaperone of the sHsp (small heat shock protein) group among ubiquitin,  $\alpha$ -crystallin, Hsp20 and others. The common functions of sHsps are chaperone activity, thermotolerance, inhibition of apoptosis, regulation of cell development, and cell differentiation. They also take part in signal transduction. Hickey et al. (1986) determined that the HSP27 gene has 3 exons. Hunt et al. (1997) mapped the mouse Hsp25 gene to chromosome 5 in a region homologous to 7q in the human. They also mapped the mouse Hsp105 gene to chromosome 5 but suggested that the human homolog is probably on 13q, not chromosome 7. The standard used in this kit is recombinant human HSP27, consisting of 205 amino acids with the molecular mass of 22.7kDa.

## Reference

1. Hickey, E., Brandon, S. E., Potter, R., Stein, G., Stein, J., Weber, L. A. **Sequence and organization of genes encoding the human 27 kDa heat shock protein.** Nucleic Acids Res. 14: 4127-4145, 1986. (Note: Erratum: Nucleic Acids Res. 14: 8230 only, 1986.)
2. Hunt, C. R., Goswami, P. C., Kozak, C. A. **Assignment of the mouse Hsp25 and Hsp105 genes to the distal region of chromosome 5 by linkage analysis.** Genomics 45: 462-463, 1997.

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