

Hycult biotechnology

HUMAN NEUTROPHIL ELASTASE ELISA Kit

Catalog No. HK319

Quantity : 2 x 96 determinations

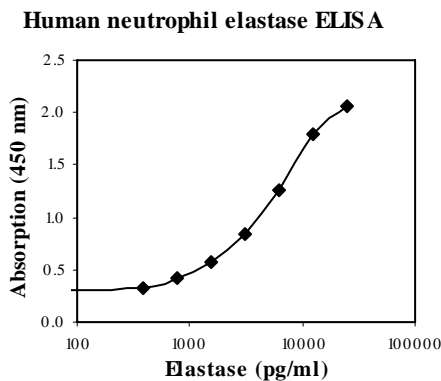
Description Neutrophil elastase, a major serine proteinase in man, is predominantly present in the azurophilic granules of neutrophils. Elastase has a broad range of extracellular matrix substrates including elastin, proteoglycans, collagen and fibronectin. The action of elastase is controlled by serine proteinase inhibitors. Elastase, when released during inflammation, is rapidly bound by its two main inhibitors, alpha1-PI and alpha2-macroglobuline to form elastase-inhibitor complexes. In addition, mucosa secretions may contain the locally secreted elastase inhibitors elafin/SKALP and SLPI. When secreted at sites of inflammation elastase can cause severe tissue damage. An important role has been suggested for elastase in various inflammatory disorders, including pulmonary emphysema, sepsis, arthritis, nephritis and certain skin diseases. Elastase induces the production of IL-8 in human bronchial epithelial, a proces that occurs in part through TLR4. In plasma of healthy individuals approx. 55 ng/ml (29 - 86 ng/ml) elastase is present.

Application The human neutrophil elastase ELISA has been developed for the quantitative measurement of free and bound natural human neurophil elastase in cell culture medium and plasma. In plasma samples human elastase can be measured accurately if plasma samples are diluted at least 20 times. Most reliable results are obtained if EDTA plasma is used. Please be aware that human neutrophil elastase is released from neutrophils into serum in the process of blood coagulation. This will lead to false positive and difficult to interpret results of serum samples. Therefore it is advised to use 'careful plasma'.

Features

- Minimum concentration which can be measured is 0.4 ng/ml neutrophil elastase.
- Measurable concentration range of 0.4-25 ng/ml.
- Working volume of 100 µl/well.

Typical standard curve



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Principle

- The human neutrophil Elastase ELISA is a ready-to-use solid-phase enzyme-linked immunosorbent assay based on the sandwich principle with a working time of 3½ hours.
- The efficient format of 2 plates with twelve disposable 8-well strips allows free choice of batch size for the assay.
- Samples and standards are captured by a solid bound specific antibody.
- Biotinylated tracer antibody will bind to captured human neutrophil elastase.
- Streptavidin-peroxidase conjugate will bind to the biotinylated tracer antibody.
- Streptavidin-peroxidase conjugate will react with the substrate, tetramethylbenzidine (TMB).
- The enzyme reaction is stopped by the addition of citric acid.
- The absorbance at 450 nm is measured with a spectrophotometer. A standard curve is obtained by plotting the absorbance (linear) versus the corresponding concentrations of the human neutrophil elastase standards (log).
- The human neutrophil elastase concentration of samples, which are run concurrently with the standards, can be determined from the standard curve.

Storage and stability

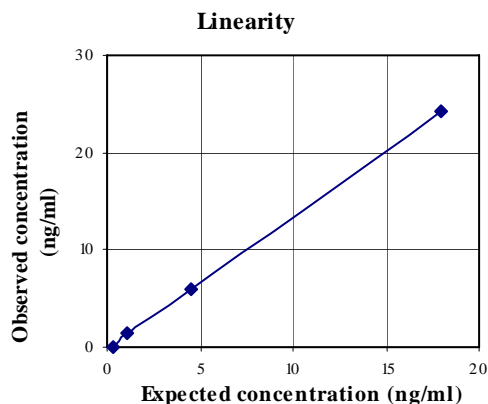
Product should be stored at 4°C. Under recommended storage conditions, product is stable for at least six months. After reconstitution the reagents are stable for 1 month if stored at 2-8°C. After reconstitution the standard is stable for 6 hours. For longer stability we recommend to store aliquots at -20°C.

Recovery

Normal human blood samples (plasma), containing baseline levels of human neutrophil elastase, were spiked with human neutrophil elastase in concentrations of 18 ng/ml. Samples with and without human neutrophil elastase were incubated for 1 hour at room temperature. Samples were measured using the ELISA. Values for human neutrophil elastase ranged between 93% and 111% (mean 104%).

Linearity

The linearity of the assay was determined by serially diluting a sample containing 18 ng/ml human neutrophil elastase. The diluted samples were measured in the assay. The line obtained a slope of 1.35 and a correlation coefficient of 0.999.



Precautions

For research use only. Not for use in or on humans or animals or for diagnostics. It is the responsibility of the user to comply with all local/state and Federal rules in the use of this product. Hbt is not responsible for any patent infringements that might result with the use of or derivation of this product.

References

1. Dentener, M et al; Bacterial/permeability-increasing protein release in whole blood ex vivo: strong induction by lipopolysaccharide and tumor necrosis factor-alpha. J Infect Dis 1996, 175: 108
2. Hiltermann, J et al; Ozone-induced inflammation assessed in sputum and bronchial lavage fluid from asthmatics: a new noninvasive tool in epidemiologic studies on air pollution and asthma. Free Radic Biol Med 1999, 27: 1448



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Also available	HK314	Human BPI ELISA kit, 2 x 96 determinations
	HK316	Human SLPI ELISA kit, 2 x 96 determinations
	HK318	Human Elafin/SKALP ELISA kit, 2 x 96 determinations
	HK324	Human MPO ELISA kit, 2 x 96 determinations
	HK325	Human Calprotectin ELISA kit, 2 x 96 determinations

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