

New

HBT ELISA TEST KITS FOR HUMAN PRE-ELAFIN/SKALP

The Hbt human pre-elafin/SKALP kit has been developed for the quantitative measurement of human pre-elafin/SKALP in culture medium, serum and plasma without interference of the 6 kDa form of elafin. Elafin is an epithelial proteinase inhibitor also known under various other names such as Skin-derived Anti leukoproteinase (SKALP) and Elastase-Specific Inhibitor (ESI). Elafin belongs to the Trappin gene family and was given the systematic name Trappin-2 in a recent classification. The Trappin family is defined by a N-terminal transglutaminase substrate domain and a C-terminal four disulphide core. Trappins have been suggested to play a role in the regulation of inflammation and in protection against tissue damage in stratified epithelia. Elafin is an inhibitor of leukocyte elastase and proteinase-3 and in addition it is a substrate for transglutaminases. The protein is constitutively expressed in various epithelia including those of hair follicles, oesophagus, vagina and oral cavity. Elafin is not present in normal human skin but is strongly induced during inflammation as in psoriasis and wound healing. The full-length protein is translated as a 12.3 kDa protein of 117 amino acids termed pre-elafin or Trappin-2. Cleavage of the signal peptide yields a mature protein with a molecular mass of 9.9 kDa. The 9.9 kDa secreted protein is the major form found in culture medium. In skin extracts a 6 kDa form comprising the 57 most C-terminal amino acids is present, which is the form of elafin originally described by Wiedow et al. In serum both the 9.9 and the 6 kDa form appear to be present. In urine only the short 6 kDa form is found. The HBT Elisa test kit is not suitable to measure the short 6 kDa form. Pre-elafin has been used as a quantitative marker for disease activity in severe psoriasis during treatment with cyclosporin.

In serum/plasma of healthy individuals approximately 10-50 ng/ml Elafin is present. Pre-elafin shows a 10-fold increase in concentration during psoriasis. The determination of pre-elafine is not disturbed by the presence of the short 6 kDa form.

PRINCIPLE OF THE TEST

The Hbt human pre-elafin/SKALP ELISA test kit is a solid-phase enzyme-linked immunosorbent assay based on the sandwich principle. Samples and standards are incubated in microtiter wells coated with antibodies recognizing human pre-elafin/SKALP. During this incubation human pre-elafin/SKALP is captured by solid bound antibody. Unbound material present in the sample is removed by washing. Next biotinylated second antibody (tracer) to human pre-elafin/SKALP is added to the wells. If pre-elafin/SKALP was present in the sample, the tracer antibodies will bind to the captured pre-elafin/SKALP. The excess tracer is removed by washing. Next a streptavidin-peroxidase conjugate is applied to the wells, this conjugate reacts specifically with the biotinylated tracer antibody bound onto the detected pre-elafin/SKALP. The excess streptavidin-peroxidase conjugate is removed by washing and substrate, tetramethylbenzidine (TMB) is added to the wells. Colour develops proportionally to the amount of human pre-elafin/SKALP present in the sample. The enzyme reaction is stopped by the addition of citric acid and the absorption at 450 nm is measured with a spectrophotometer. A standard curve is obtained by plotting the absorptions versus the corresponding concentrations of the known standards. The human pre-elafin/SKALP concentration of samples with unknown concentrations, which are run concurrently with the standards, can be determined from the standard curve.

SPECIAL FEATURES OF THE KIT

- Ready-to-use (i.e. pre-coated microwells).
- High specificity.
- High reproducibility
- High sensitivity. The minimum concentration which can be measured is 160 pg/ml of pre-elafin/SKALP.
- Large measurable concentration range. Standard curve from 156-10,000 pg/ml.
- Efficient format. Two plates with each twelve 8-well strips allow free choice of batch size for the assay.
- Simple, rapid procedure. Four pipetting steps are required to complete the assay. Working time 4½ hours.

LITERATURE

- Alkemade HAC, et al. Levels of skin-derived antileukoproteinase (SKALP)/Elafin in serum correlate with cyclosporin A. *J. Invest. Dermatol.* 1995; 104: 189-193.
- Kuijpers ALA, et al. The effects of oral liarozole on epidermal proliferation and differentiation in severe plaque psoriasis are comparable with those of acitretin. *Br. J. of Dermatology.* 1998; 139: 380-389.
- Vandermeeren M, et al. Development and application of monoclonal antibodies against SKALP/elafin and other trappin family members. *Arch. Dermatol. Res.* 2001; 293(7): 343-349.

AVAILABILITY

The Hbt human pre-elafin/SKALP test is available in a kit for 2 x 96 determinations.

PRODUCT NUMBER: HK318

Hbt human pre-elafin/SKALP ELISA Kit

For research purposes only.

Caution: Not for use in humans.