

# Human IgE ELISA development kit

Product Code: 3810-1A-6

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CONTENTS, development kit for 6 plates:

**Vial 1 (green top)**

Monoclonal antibody 107 (150  $\mu$ l)

Concentration: 1 mg/ml

**Vial 2 (blue top)**

Biotinylated monoclonal antibody 182 (80  $\mu$ l)

Concentration: 1 mg/ml

**Vial 3 (white top)**

Streptavidin-Alkaline Phosphatase (80  $\mu$ l)

**Vial 4**

Lyophilised human IgE standard (2  $\mu$ g)

To ensure total recovery of stated quantity, vials have been overfilled.

**STORAGE:**

Shipped at ambient temperature. On arrival box 1 should be stored refrigerated at 4-8°C and box 2 should be stored frozen at -20°C.

# General

**Intended use:** For quantitative determination of native human IgE.

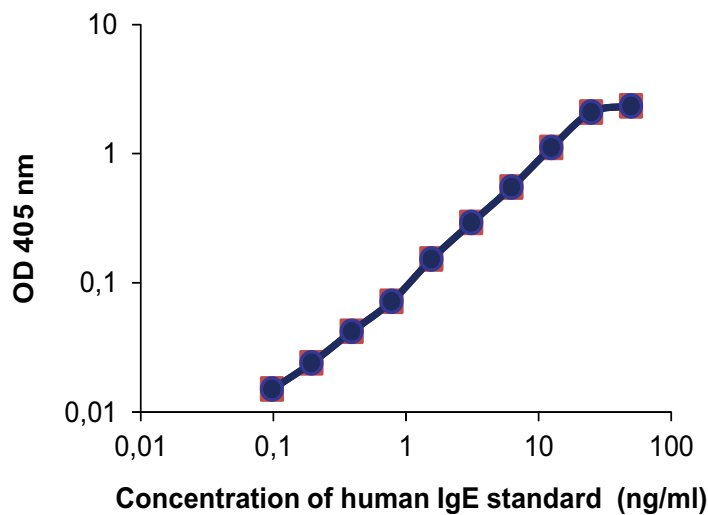
**Serum/plasma samples:** Please note that determination of analyte in human serum/plasma samples by this kit requires the use of ELISA diluent (product code: 3652-D2) for dilution of samples, standard and detection antibody. The diluent prevents false positive read-outs which may be caused by interference of heterophilic antibodies commonly found in human plasma and serum. The ELISA diluent has been validated using serum/plasma from normal healthy human blood donors. Please note that heterophilic antibody interference in samples from human subjects with various diseases or other conditions has not been assessed. Please contact Mabtech for further information.

**Reagents:** Antibodies are supplied in sterile-filtered (0.2  $\mu\text{m}$ ) PBS with sodium azide (0.02%). Streptavidin-ALP is supplied in 0.1 M Tris buffer with 0.002% Kathon CG.

**Standard range:** 0.3-30 ng/ml

**Standard calibration:** 1 ng of supplied standard equals 0.5 U of 75/502 NIBSC\* standard according to repeated calibrations. Calibration is batch-specific.

\* National Institute of Biological Standards and Control, UK.



# Guidelines for Human IgE ELISA

- Day 1**
1. Coat a high protein binding ELISA plate with mAb 107, diluted to 2  $\mu\text{g}/\text{ml}$  in PBS, pH 7.4, by adding 100  $\mu\text{l}/\text{well}$ . Incubate overnight at 4-8°C.
- Day 2**
2. Wash twice with PBS (200  $\mu\text{l}/\text{well}$ ).
  3. Block plate by adding 200  $\mu\text{l}/\text{well}$  of PBS with 0.05% Tween 20 containing 0.1% BSA (incubation buffer). Incubate for 1 hour at room temperature.
  4. Wash five times with PBS containing 0.05% Tween.
  5. Prepare hIgE standard by reconstituting contents of vial 4 in 1 ml PBS to make up a stock solution of 2  $\mu\text{g}/\text{ml}$ . The stock solution should be used immediately or stored in aliquots at -20°C for future use. We recommend the aliquots not be refrozen after initial use. For the test, prepare dilutions of the stock using the standard range as a guideline.
  6. Add 100  $\mu\text{l}/\text{well}$  of samples or standards diluted in incubation buffer or ELISA diluent for serum/plasma samples and incubate for 2 hours at room temperature.
  7. Wash as in step 4.
  8. Add 100  $\mu\text{l}/\text{well}$  of mAb 182-biotin diluted to 0.3  $\mu\text{g}/\text{ml}$  in incubation buffer or ELISA diluent for serum/plasma samples. Incubate for 1 hour at room temperature.
  9. Wash as in step 4.
  10. Add 100  $\mu\text{l}/\text{well}$  of Streptavidin-ALP diluted 1:1000 in incubation buffer. Incubate for 1 hour at room temperature
  11. Wash as in step 4.
  12. Add 100  $\mu\text{l}/\text{well}$  of appropriate substrate solution e.g. p-nitrophenyl-phosphate (pNPP), available from Mabtech product code 3652-P10.
  13. Measure the optical density (405 nm for pNPP) in an ELISA reader after suitable developing time.

Developed and manufactured by MABTECH AB, Sweden, whose quality management system complies with the standards ISO 9001:2015 & ISO 13485:2016.



**The products are for research use only.**

MABTECH shall not be liable for the use or handling of the product or for consequential, special, indirect or incidental damages there from.

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