

# ELISA for Human IL-5

Product Code: 3490-1A-20

---

CONTENTS, development kit for 20 plates:

**Vial 1 (green top)**

Monoclonal antibody TRFK5 (500 µl)

Concentration: 1 mg/ml

**Vial 2 (yellow top)**

Biotinylated monoclonal antibody 5A10 (250 µl)

Concentration: 1 mg/ml

**Vial 3 (white top)**

Streptavidin-Alkaline Phosphatase (250 µl)

**Vial 4**

Recombinant human IL-5 standard (1 µg)

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 and recombinant human IL-5 in solution, e.g. cell culture supernatant.

**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  $\mu$ m) PBS with sodium azide (0.02%). Streptavidin-ALP is supplied in 0.1 M Tris buffer with 0.15% Kathon CG.

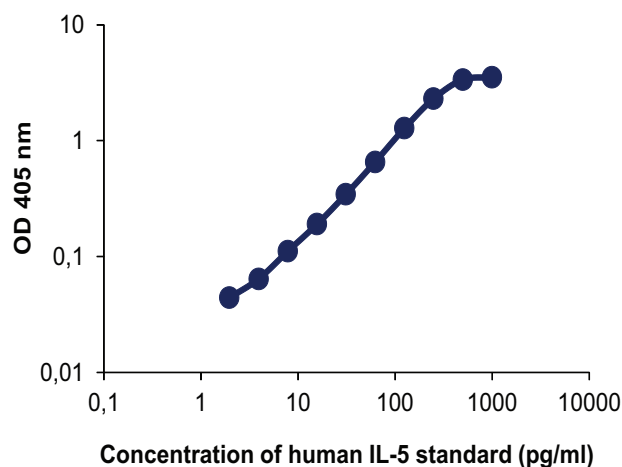
**Standard range:** 6-600 pg/ml

**Limit of detection:** 4 pg/ml

**Intra-assay variation:** < 5%

**Standard calibration:** 1 ng of supplied standard equals 14 U of 90/586 NIBSC\*-standard according to repeated calibrations. Calibration is batch-specific.

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



# Guidelines for Human IL-5 ELISA

- Day 1**
1. Coat a high protein binding ELISA plate with mAb TRFK5, diluted to 2 µg/ml in PBS, pH 7.4, by adding 100 µl/well. Incubate overnight at 4-8°C.
- Day 2**
2. Wash twice with PBS (200 µl/well).
  3. Block plate by adding 200 µl/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% Tween20
  5. Prepare hIL-5 standard by reconstituting contents of vial 4 in 1 ml PBS to a concentration of 1 µg/ml. Leave at room temperature for 15 minutes and then vortex the tube. 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 µl/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 µl/well of mAb 5A10-biotin at 1 µg/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 µl/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 µl/well of appropriate substrate solution e.g. p-nitrophenyl-phosphate (pNPP).
  13. Measure the optical density (405 nm for pNPP) in an ELISA reader after suitable developing time.

**NOTE; 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 therefrom.



MABTECH AB  
Box 1233  
SE-131 28 Nacka Strand  
Sweden  
Tel: +46 8 716 27 00  
Fax: +46 8 716 27 01  
E-mail: [mabtech@mabtech.com](mailto:mabtech@mabtech.com)  
[www.mabtech.com](http://www.mabtech.com)

MABTECH Inc  
M.E.B. 220  
3814 West Street  
Cincinnati, OH 45227  
USA  
Tel: +1 513 871 4500  
Fax: +1 513 871 7353  
E-mail: [mabtech.usa@mabtech.com](mailto:mabtech.usa@mabtech.com)

MABTECH AB Büro Deutschland  
Germany  
Tel: +49 40 4135 7935  
Fax: +49 40 4135 7945  
E-mail: [mabtech.de@mabtech.com](mailto:mabtech.de@mabtech.com)

2013-01-24

Developed and manufactured by MABTECH AB, Sweden, whose quality management system complies with the following standards:



MABTECH AUSTRALIA Pty Ltd  
resolvingIMAGES  
Unit 22, 196 Settlement Road  
Thomastown Victoria 3074  
Australia  
Tel: +61 3 9466 4007  
Fax: +61 3 9466 4003  
E-mail: [mabtech.au@mabtech.com](mailto:mabtech.au@mabtech.com)

MABTECH AB Bureau de liaison France  
BP 255, 1300 route des Crêtes  
06905 Sophia Antipolis  
France  
Tel: +33 (0)4 92 38 80 70  
Fax: +33 (0)4 92 38 80 71  
E-mail: [mabtech.fr@mabtech.com](mailto:mabtech.fr@mabtech.com)