

# ELISA for Human IFN- $\alpha$ (pan specific)

Product Code: 3425-1A-20

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CONTENTS, development kit for 20 plates:

**Vial 1 (yellow top)**

Monoclonal antibodies MT1/3/5 a combination of three different antibodies (1000  $\mu$ l)

Concentration: 1 mg/ml

**Vial 2 (red top)**

Biotinylated monoclonal antibodies MT2/4/6 (250  $\mu$ l)

Concentration: 1 mg/ml

**Vial 3 (white top)**

Streptavidin-Alkaline Phosphatase (250  $\mu$ l)

**Vial 4**

Lyophilised native human IFN- $\alpha$  standard (0.5  $\mu$ 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 at -20°C.

# General

**Intended use:** For quantitative determination of native and recombinant human IFN- $\alpha$  in solution, e.g. cell culture supernatant. The system will detect native and recombinant human IFN- $\alpha$  subtypes 1/13, 2, 4, 5, 6, 7, 8, 10, 14, 16 and 17.

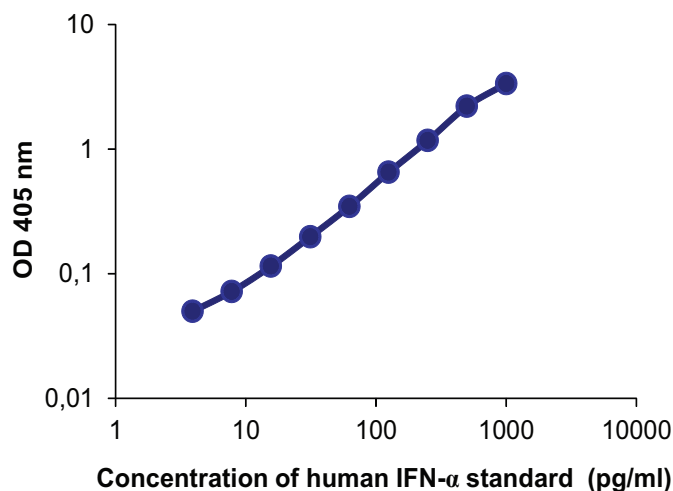
**Serum/plasma samples:** Please note that determination of analyte in human serum/plasma samples by this kit requires the use of ELISA diluent (product code: 3652-D2) for dilution of samples, standard and detection antibody. The diluent prevents false positive read-outs which may be caused by interference 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:** 7-700 pg/ml

**Standard calibration:** 1 ng of supplied standard equals 400 U of 94/784 NIBSC\*-standard according to repeated calibrations. Calibration is batch-specific.

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



# Guidelines for Human IFN- $\alpha$ (pan specific) ELISA

- Day 1**
1. Coat a high protein binding ELISA plate with mAbs MT1/3/5, diluted to 4  $\mu\text{g}/\text{ml}$  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% Tween20
  5. Prepare hIFN- $\alpha$  standard by reconstituting contents of vial 4 in 1 ml PBS with 0.1% BSA to make up a stock solution of 500 ng/ml. Use immediately or store in aliquots at -20°C for future use. We recommend the aliquots not to 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 mAbs MT2/4/6-biotin diluted to 1  $\mu\text{g}/\text{ml}$  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-nitro-phenyl-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)

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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)