ELISA for Human IFN-α (subtype 2)

Product Code: 3423-1H-20

CONTENTS, development kit for 20 plates:

Vial 1 (red top)
Monoclonal antibody MT1 (500 µl)
Concentration: 1 mg/ml

Vial 2 (yellow top)
Biotinylated monoclonal antibody MT2 (250 µl)
Concentration: 1 mg/ml

Vial 3 (white top)
Streptavidin-Horseradish peroxidase (250 µl)

Vial 4
Recombinant human IFN-α standard, subtype 2c

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 IFN-α in solution, e.g. cell culture supernatant. The system will detect native and recombinant human IFN-α subtypes 2a, 2b and 2c. 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 µm) PBS with sodium azide (0.02%). Streptavidin-HRP is supplied in PBS with 1% BSA and 0.15% Kathon CG.

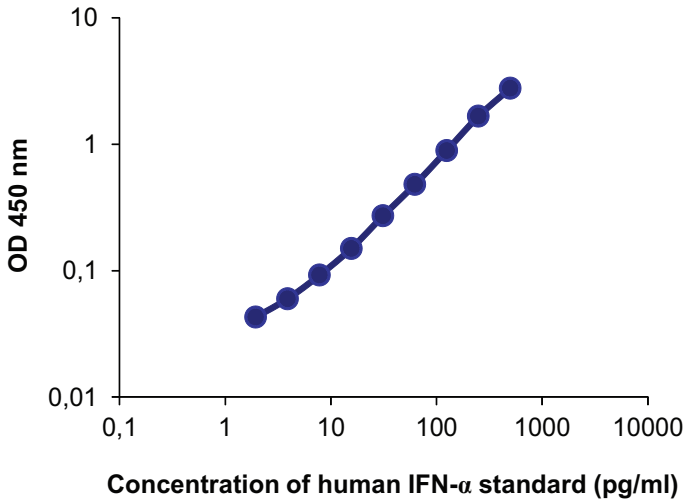
Standard range: 4-400 pg/ml

Limit of detection: 2 pg/ml

Intra-assay variation: < 4%

Standard calibration: 1 ng of supplied standard equals 195 U 95/566 NIBSC*standard according to repeated calibrations. Calibration is batch-specific.

*National Institute for Biological Standards and Control, UK.



Guidelines for Human IFN-α (subtype 2) ELISA

Day 1

1. Coat a high protein binding ELISA plate with mAb MT1, 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% Tween.
5. Prepare hIFN-α 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 MT2-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-HRP diluted 1:1000 in incubation buffer. Incubate for 1 hour at room temperature.

Please note that sodium azide used in buffers will inhibit HRP activity.

11. Wash as in step 4.
12. Add 100 µl/well of appropriate substrate solution.
13. Measure the optical density in an ELISA reader after suitable developing time. If required stop reaction.

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.

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