ELISA for Mouse IFN-γ

Product Code: 3321-1H-20

CONTENTS, development kit for 20 plates:

Vial 1 (green top)
Monoclonal antibody AN18 (500 µl)
Concentration: 1 mg/ml

Vial 2 (yellow top)
Biotinylated monoclonal antibody R4-6A2 (250 µl)
Concentration: 1 mg/ml

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

Vial 4
Recombinant mouse IFN-γ 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 mouse IFN-γ in solution, e.g. cell culture supernatant and serum/plasma samples.

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.

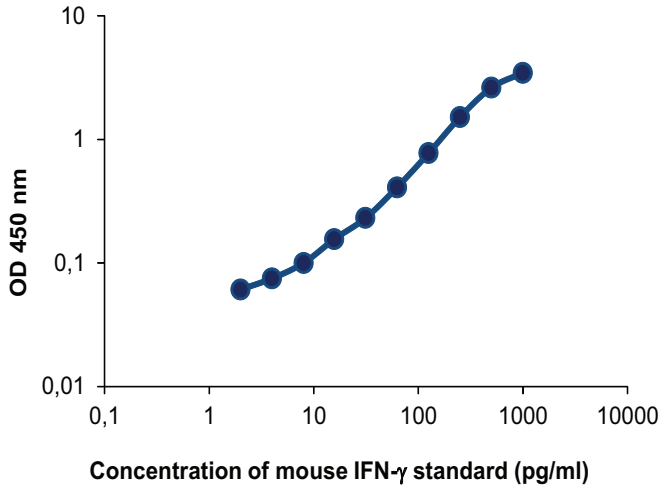
Standard range: 4-400 pg/ml

Limit of detection: 2 pg/ml

Intra-assay variation: < 4%

Standard calibration: 1 ng of supplied standard equals 5 U of Gg02-901-533 NIAID*-standard according to repeated calibrations. Calibration is batch-specific.

*National Institute of Allergy and Infectious Diseases, USA.



Guidelines for Mouse IFN-γ ELISA

Day 1 1. Coat a high protein binding ELISA plate with mAb AN18, diluted to 1 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 mouse IFN-γ standard by reconstituting contents of vial 4 in 1 ml PBS with 1% BSA 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 and incubate for 2 hours at room temperature.

7. Wash as in step 4.

8. Add 100 μl/well of mAb R4-6A2-biotin at 0.5 μg/ml in incubation buffer. 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 the reaction first.

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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