

# Mouse IFN- $\gamma$ ELISA development kit

Product Code: 3321-1A-20

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CONTENTS, development kit for 20 plates:

**Vial 1 (green top)**

Monoclonal antibody AN18 (500  $\mu$ l)

Concentration: 1 mg/ml

**Vial 2 (yellow top)**

Biotinylated monoclonal antibody R4-6A2 (250  $\mu$ l)

Concentration: 1 mg/ml

**Vial 3 (white top)**

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

**Vial 4**

Recombinant mouse IFN- $\gamma$  standard (1  $\mu$ 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 mouse IFN- $\gamma$  in solution, e.g. cell culture supernatant and serum/plasma samples.

**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 0.002% Kathon CG.

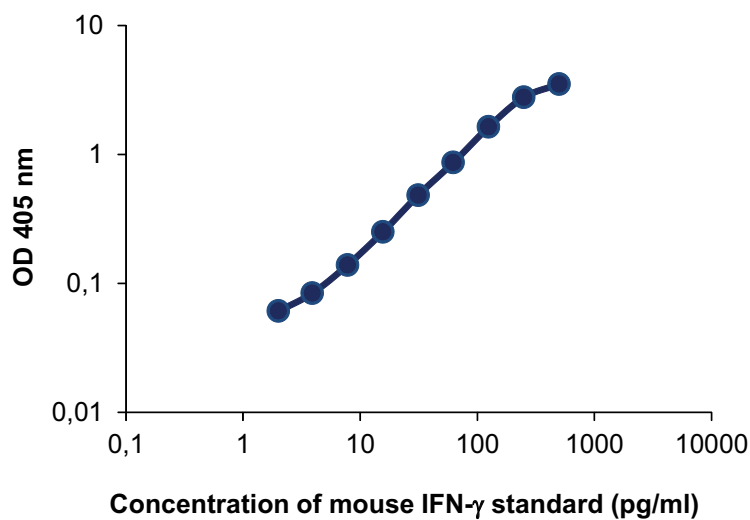
**Standard range:** 4-400 pg/ml

**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- $\gamma$ ELISA

- Day 1**
1. Coat a high protein binding ELISA plate with mAb AN18, diluted to 1  $\mu\text{g}/\text{ml}$  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 mouse IFN- $\gamma$  standard by reconstituting contents of vial 4 in 1 ml PBS with 1% BSA to a concentration of 1  $\mu\text{g}/\text{ml}$ . 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  $\mu\text{l}/\text{well}$  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  $\mu\text{l}/\text{well}$  of mAb R4-6A2-biotin at 0.5  $\mu\text{g}/\text{ml}$  in incubation buffer. 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.**

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