# Rat IFN-γ ELISA development kit

Product Code: 3220-1H-6

## CONTENTS, development kit for 6 plates:

### Vial 1 (green)

Monoclonal antibody rIFNγ-I (150 μl)

Concentration: 1 mg/ml

# Vial 2 (yellow top)

Biotinylated monoclonal antibody rIFN $\gamma$ -II (80  $\mu$ l)

Concentration: 1 mg/ml

## Vial 3 (white top)

Streptavidin-Horseradish Peroxidase (80 µl)

#### Vial 4

Recombinant rat IFN-γ standard (20 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

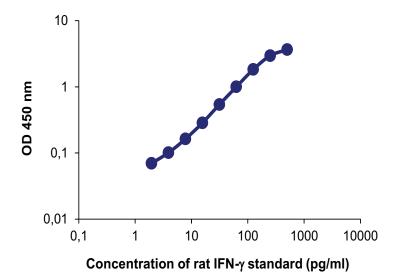
**Intended use:** For quantitative determination of native rat IFN- $\gamma$  in solution, e.g. cell culture supernatant and serum/plasma samples.

**Reagents:** Antibodies are supplied in sterile-filtered (0.2  $\mu$ m) PBS with sodium azide (0.02%). Streptavidin-HRP is supplied in PBS with 1% BSA and 0.002% Kathon CG.

Standard range: 3-300 pg/ml.

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

**Standard calibration:** No international standard exists for calibration.



# Guidelines for Rat IFN-γ ELISA

- **Day 1** 1. Coat a high protein binding ELISA plate with mAb rIFN $\gamma$ -I, 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  $\mu$ l/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 rat IFN-γ standard by reconstituting contents of vial 4 in 200 μl 5mM Tris pH 8.0+100mM NaCl to a concentration of 0.1 mg/ml. Dilute in PBS with 0.1% BSA to make up a stock solution of 10 μg/ml. 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  $\mu$ l/well of mAb rIFN $\gamma$ -II-biotin at 1  $\mu$ g/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 e.g. TMB, available from Mabtech product code 3652-F10.
  - 13. Measure the optical density in an ELISA reader after suitable developing time. If required stop the reaction first.



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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