ELISA for Equine IFN-γ

Product Code: 3117-1A-20

CONTENTS, development kit for 20 plates:

Vial 1 (green top)
Monoclonal antibody bIFNγ-I (1000 µl)
Concentration: 0.5 mg/ml

Vial 2 (red top)
Biotinylated monoclonal antibody PAN (125 µl)
Concentration: 0.5 mg/ml

Vial 3 (white top)
Streptavidin-Alkaline Phosphatase (250 µl)

Vial 4
Recombinant equine IFNγ standard

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.

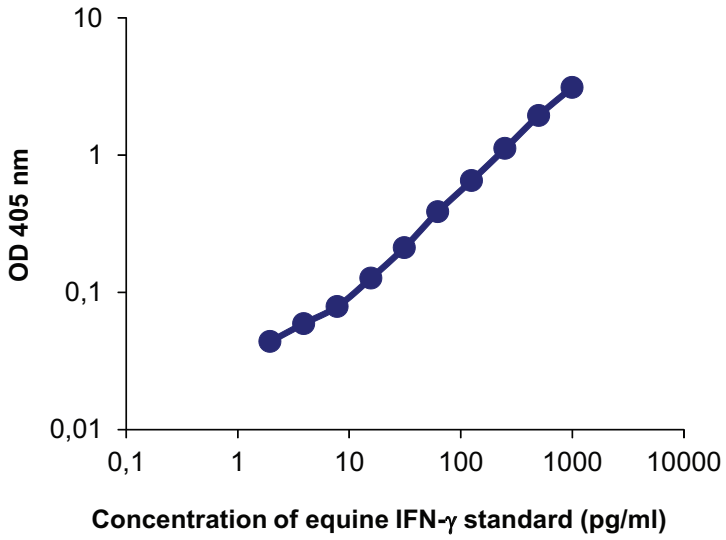
General

Intended use: For quantitative determination of native and recombinant equine IFN-γ in solution, e.g. cell culture supernatant and serum/plasma samples. The two mAbs cross react with native ovine IFN-γ and native and recombinant bovine IFN-γ.

Reagents: Antibodies are supplied in sterile-filtered (0.2 µm) PBS with sodium azide (0.02%). Streptavidin-ALP is supplied in 0.1 M Tris buffer with 0.15% Kathon CG.

Standard range: 8-800 pg/ml.

Standard calibration: No international standard exists for calibration.



Guidelines for Equine IFN-γ ELISA

Day 1

1. Coat a high protein binding ELISA plate with mAb bIFNγ-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 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 equine IFN-γ standard by reconstituting contents of vial 4 in 1 ml PBS with 1% BSA and leave at room temperature for 15 minutes, then vortex the tube and spin down. This gives a concentration of 0.2 µg/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 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 PAN-biotin at 0.1 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-ALP diluted 1:1000 in incubation buffer. Incubate for 1 hour at room temperature.
11. Wash as in step 4.
12. Add 100 µl/well of appropriate substrate solution e.g. p-nitrophenyl-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.

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