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Mouse TNF-α ELISA development kit

Product Code: 3511-1H-6

CONTENTS, development kit for 6 plates:

Vial 1 (green top)
Monoclonal antibodies MT1C8/23C9 (300 µl)
Concentration: 0.5 mg/ml

Vial 2 (yellow top)
Biotinylated monoclonal antibody MT11B10 (80 µl)
Concentration: 0.5 mg /ml

Vial 3 (white top)
Streptavidin-Horseradish Peroxidase (80 µl)

Vial 4
Recombinant mouse TNF-α standard (0.5 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

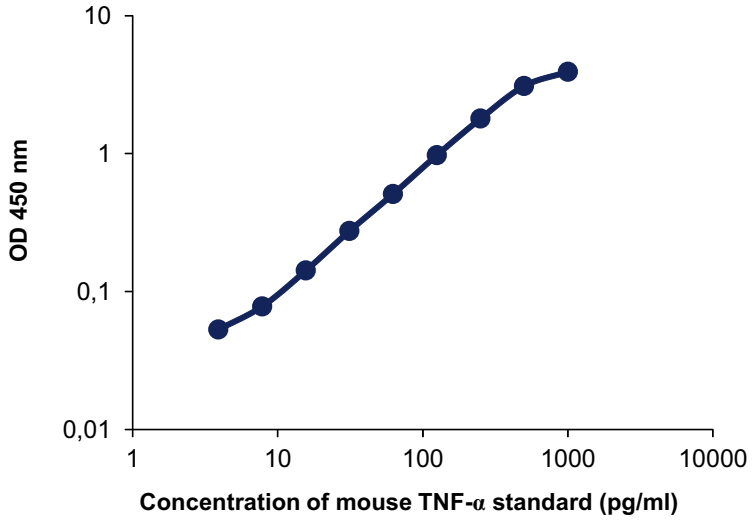
Intended use: For quantitative determination of native and recombinant mouse TNF-α in solution, e.g. cell culture supernatant.

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 0.002% Kathon CG.

Standard range: 6-600 pg/ml

Standard calibration: 1 ng of supplied standard equals 209 U of 88/532 NIBSC*-standard according to repeated calibrations. Calibration is batch-specific.

*National Institute of Biological Standards and Control, UK.



Guidelines for Mouse TNF-α ELISA

Day 1

1. Coat a high protein binding ELISA plate with mAbs MT1C8/23C9, 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% Tween20
5. Prepare TNF-α standard by reconstituting contents of vial 4 in 1 ml PBS to a concentration of 0.5 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 MT11B10-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.

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NOTE; for research use only.