

Porcine TNF- α ELISA development kit

Product Code: 3135-1A-20

CONTENTS, development kit for 20 plates:

Vial 1 (red top)

Monoclonal antibody MT1P14 (1000 μ l)

Concentration: 0.5 mg/ml

Vial 2 (yellow top)

Biotinylated monoclonal antibody MT859 (250 μ l)

Concentration: 0.5 mg/ml

Vial 3 (white top)

Streptavidin-Alkaline Phosphatase (250 μ l)

Vial 4

Recombinant porcine TNF- α 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.

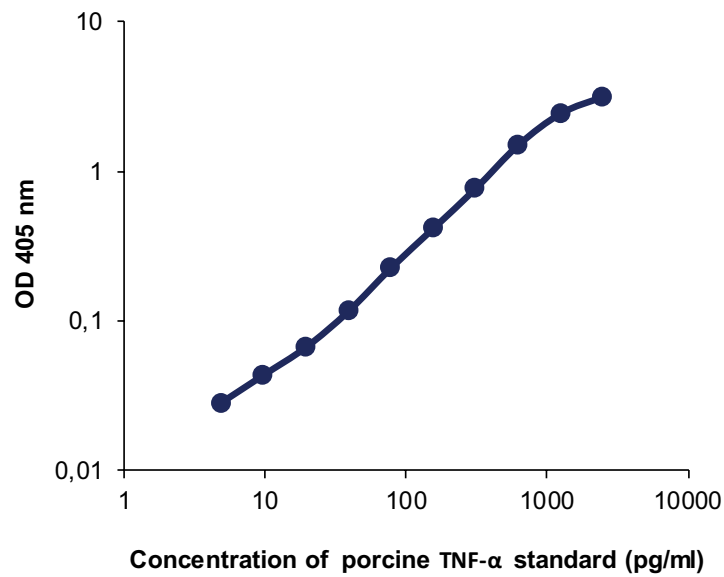
General

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

Reagents: Antibodies are supplied in sterile-filtered (0.2 μ m) PBS with sodium azide (0.02%). Streptavidin-ALP is supplied in 0.1 M Tris buffer with 0.002% Kathon CG.

Standard range: 10-1000 pg/ml

Standard calibration: No international standard exists for calibration.



Guidelines for Porcine TNF- α ELISA

- Day 1**
1. Coat a high protein binding ELISA plate with mAb MT1P14, diluted to 2 $\mu\text{g}/\text{ml}$ 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% Tween20
 5. Prepare porcine TNF- α standard by reconstituting contents of vial 4 in 1 ml PBS to give a concentration of 2.5 $\mu\text{g}/\text{ml}$. Leave at room temperature for 15 minutes, then vortex the tube and spin down. 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 $\mu\text{l}/\text{well}$ 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 MT859-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.



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