

## DATA SHEET

Versions: 01

Revision date: 25/11/2023

### 1. Identification

Product name: ELISA kits for Mouse of Mouse TNF- $\alpha$ 

Reactions: 96,48 rxns

Cat. No.: PRA-M TNF- $\alpha$ -96, PRA-M TNF- $\alpha$ -48

### 2. Description

Tumor Necrosis Factor-alpha (TNF- $\alpha$ ), a pivotal inflammatory cytokine, is primarily synthesized by innate immune cells, particularly macrophages. Recognized for its multifaceted inflammatory properties, TNF- $\alpha$  plays a crucial role in combating bacterial, viral, and fungal infections. Simultaneously, it assumes a significant role in the development of diseases mediated by cellular immunity, making it a central factor in infectious shock disease. TNF- $\alpha$  is instrumental in initiating and amplifying inflammatory responses. Its well-defined role in combating diverse infections, including bacterial, viral, and fungal, underscores its importance in immune defense. TNF- $\alpha$ 's involvement in diseases mediated by cellular immunity establishes it as a key player in pathological processes. The cytokine is recognized as a primary contributor to infectious shock disease, emphasizing its potent impact on the immune system. As a crucial inflammatory indicator, TNF- $\alpha$  is extensively employed in laboratory studies. Its application facilitates the investigation of disease conditions and the assessment of the inflammatory or anti-inflammatory effects of various drugs. The current kit, meticulously designed and produced, utilizes mouse anti-TNF- $\alpha$  monoclonal antibodies. It is essential to note that this kit is specifically intended for measuring TNF- $\alpha$  levels in mice and may not be suitable for similar animal and human cases. This comprehensive overview highlights the central role of TNF- $\alpha$  in inflammation, immune response, and disease mediation, making it a focal point in both experimental research and clinical investigations.



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### 3. Kit Contents

Component	Cat. no	Quantity
Human anti-TNF- $\alpha$ pre-coated plate	PRA- CTNF- $\alpha$ P	96 vials
Standards	PRA-TNF- $\alpha$ N1-4	200 $\mu$ l
HRP-Avidin buffer	PRA-HA	5 ml
HRP	PRA-HAA	540 $\mu$ l
Substrate	PRA-SU	5 ml
Stopping	PRA-ST	7 ml
10X washing buffer	PRA-WB	40 ml
Detection Ab	PRA- TNF- $\alpha$ D	5 ml

### 4. Storage specifications

All components of the ELISA kits can be stored at 4°C temperature.

### 5. Applications

Detection of both inflammatory and anti-inflammatory factors through ELISA.

### 6. Assay Procedure

How to Prepare Solutions:

1. Washing Buffer:

- To prepare the washing solution, dilute it with distilled water at a 1:10 ratio.

2. HRP-Avidin:

- Spin the HRP vial using a microfuge device, then add all its contents to the HRP-Avidin buffer vial.

- For quantities less than 48 assays, mix 416  $\mu$ L of HRP-Avidin, 41  $\mu$ L of HRP per 8-well row.



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### Working with the Kit:

#### 1. Plate Preparation:

- Remove the plate from its package and let it reach room temperature in a dry environment.

- Add 50  $\mu$ L of standards #4 to #1 in the first to fourth wells.

#### 2. Sample Incubation:

- Add 50  $\mu$ l of the desired sample to the remaining wells.

- Incubate for 60 minutes on a 200 RPM shaker at room temperature.

#### 3. Plate Washing:

- After proper incubation, wash the plates three times using the washing solution (after adding the washing solution, incubate the plates for approximately 1 minute at room temperature and then drain).

#### 4. Conjugated Antibody Addition:

- Add 50  $\mu$ l of conjugated antibody (Detection ab) to all wells.

- Incubate for 60 minutes on a 200 RPM shaker at room temperature.

#### 5. Plate Washing (Again):

- After proper incubation, wash the plates three times using washing solution.

#### 6. HRP-Avidin Addition:

- Add 50  $\mu$ l of HRP-Avidin solution to all wells.

- Incubate for 30 minutes on a shaker (at least at RPM 200).

#### 7. Plate Washing (Again):

- After proper incubation, wash the plates five times using washing solution.

#### 8. Substrate Addition:

- Add 50  $\mu$ l of substrate to all wells and incubate for 15 minutes. Note that 15 minutes is enough for incubation, but if the amount of color produced is low, the time can be increased to 20 minutes.

#### 9. Stopping Reaction:

- Add 25  $\mu$ l of the stopping solution to all the wells.

#### 10. Measurement:

- Measure the absorbance of the samples in an ELISA reader at a wavelength of 450 nm.



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### 7. Safety

- The solutions used in the kit have oxidizing and acidic properties.
- Avoid direct contact with skin and eyes.
- In case of contact with the mentioned tissues, wash with plenty of water and go to the nearest medical center.

### 8. Quality Certifications

### 9. Further information

- This product is developed, designed, and sold exclusively only for research purposes use.
- The product was not tested for use in diagnostics or for drug development.
- It is not suitable for administration to humans or animals.

### 10. Other Kits

Other ELISA kits:

Human:

IL-1 $\beta$ , IL-2, IL-4, IL-6, IL-8, IL-10, IL-12, IL-13, IL-18, IL-23, IL-29, IL-17A, TGF- $\beta$ , VEGF, TNF- $\alpha$ , IFN- $\gamma$ , CCL2 (MCP-1), CCL3 (MIP-1-alpha), CXCL10 (IP-10), CXCL12 (SDF-1), CCL21

Mouse:

IL-1 $\beta$ , IL-2, IL-4, IL-6, IL-10, IL-13, IL-33, IL-18, TNF- $\alpha$ , TGF- $\beta$ , CCL3, IFN- $\gamma$ , Total IgG, Total IgE

Rat:

TNF- $\alpha$ , IL-1 $\beta$ , IL-6, IL-10, IL-17A



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### NOTE

All products have been manufactured by Karmania Pars Gene Company in the Iran.

