

DESCRIPTION

Species Reactivity	Porcine
Specificity	Detects porcine TNF- α in direct ELISAs and Western blots. In Western blots, no cross-reactivity with recombinant human (rh) APRIL, rhFasL, rhGITRL, rhLIGHT, recombinant mouse (rm) TRANCE, rhBAFF, rhTNF- α , rmTNF- α , recombinant rat TNF- α , or rhTNF- β is observed.
Source	Monoclonal Mouse IgG ₁ Clone # 103302
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	<i>E. coli</i> -derived recombinant porcine TNF- α Arg78-Leu232 Accession # P23563
Endotoxin Level	<0.10 EU per 1 μ g of the antibody by the LAL method.
Formulation	Lyophilized from a 0.2 μ m filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied as a 0.2 μ m filtered solution in PBS.

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Western Blot	1 μ g/mL	Recombinant Porcine TNF- α (Catalog # 690-PT)
Immunocytochemistry	8-25 μ g/mL	See Below
Neutralization	Measured by its ability to neutralize TNF- α -induced cytotoxicity in the PK-15 porcine kidney epithelial cell line. Matthews, N. and M. L. Neale (1987) in <i>Lymphokines and Interferons, A Practical Approach</i> . Clemens, M. J. <i>et al.</i> (eds): IRL Press. 221; Bertoni <i>et al.</i> (1993) <i>J. Immunol. Meth.</i> 160 :267. The Neutralization Dose (ND ₅₀) is typically 0.005-0.02 μ g/mL in the presence of 0.05 ng/mL Recombinant Porcine TNF- α and 1 μ g/mL actinomycin D.	

DATA

<p>Neutralization</p> <p>Cytotoxicity Induced by TNF-α and Neutralization by Porcine TNF-α Antibody. Recombinant Porcine TNF-α (Catalog # 690-PT) induces cytotoxicity in the PK-15 porcine kidney epithelial cell line in a dose-dependent manner (orange line). Cytotoxicity elicited by Recombinant Porcine TNF-α (0.05 ng/mL) is neutralized (green line) by increasing concentrations of Mouse Anti-Porcine TNF-α Monoclonal Antibody (Catalog # MAB6903). The ND₅₀ is typically 0.005-0.02 μg/mL in the presence of the metabolic inhibitor actinomycin D (1 μg/mL).</p>	<p>Immunocytochemistry</p> <p>TNF-α in Porcine PBMCs. TNF-α was detected in immersion fixed porcine peripheral blood mononuclear cells treated with calcium ionomycin and PMA using Mouse Anti-Porcine TNF-α Monoclonal Antibody (Catalog # MAB6903) at 25 μg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cytoplasm. View our protocol for Fluorescent ICC Staining of Non-adherent Cells.</p>
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PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

Tumor necrosis factor alpha (TNF- α), also known as cachectin and TNFSF2, is the prototypic ligand of the TNF superfamily. It is a pleiotropic molecule that plays a central role in inflammation, apoptosis, and immune system development. TNF- α is produced by a wide variety of immune and epithelial cell types (1, 2). Porcine TNF- α consists of a 35 amino acid (aa) cytoplasmic domain, a 21 aa transmembrane segment, and a 176 aa extracellular domain (ECD) (3). Within the ECD, porcine TNF- α shares 69%-86% aa sequence identity with bovine, canine, cotton rat, equine, feline, human, mouse, rat, and rhesus TNF- α . The 26 kDa type 2 transmembrane protein is assembled intracellularly to form a noncovalently linked homotrimer (4). Ligation of this complex induces reverse signaling that promotes lymphocyte costimulation but diminishes monocyte responsiveness (5). Cleavage of membrane bound TNF- α by TACE/ADAM17 releases a 55 kDa soluble trimeric form of TNF- α (6, 7). TNF- α trimers bind the ubiquitous TNF RI and the hematopoietic cell-restricted TNF RII, both of which are also expressed as homotrimers (1, 8). TNF- α regulates lymphoid tissue development through control of apoptosis (2). It also promotes inflammatory responses by inducing the activation of vascular endothelial cells and macrophages (2). TNF- α is a key cytokine in the development of several inflammatory disorders (9). It contributes to the development of type 2 diabetes through its effects on insulin resistance and fatty acid metabolism (10, 11).

References:

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