

## Purified anti-mouse TNF- $\alpha$

**Catalog # / Size:** 506301 / 50  $\mu$ g  
506302 / 500  $\mu$ g

**Clone:** MP6-XT22

**Isotype:** Rat IgG1,  $\kappa$

**Immunogen:** *E. coli*-expressed, recombinant mouse TNF- $\alpha$

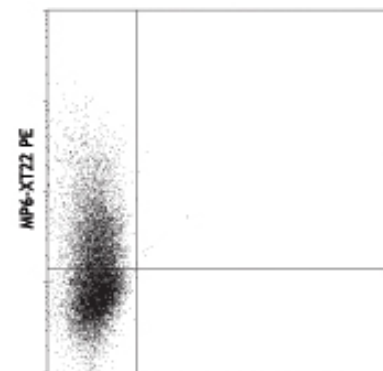
**Reactivity:** Mouse

**Preparation:** The antibody was purified by affinity chromatography.

**Formulation:** Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide.

**Concentration:** 0.5 mg/ml

**Storage:** The antibody solution should be stored undiluted at 4°C.



PMA/Ionomycin-stimulated BALB/c T cells were stained with MP6-XT22 PE

## Applications:

**Applications:** ELISA, ICFC - *Quality tested*  
CyTOF®, IF - *Validated*  
IHC, WB - *Reported in the literature*

**Recommended Usage:** Each lot of this antibody is quality control tested by ELISA assay. For ELISA capture applications, a concentration range of 2-6  $\mu$ g/ml is recommended. To obtain a linear standard curve, serial dilutions of mouse TNF- $\alpha$  recombinant protein ranging from 500 to 4 pg/ml are recommended for each ELISA plate. It is recommended that the reagent be titrated for optimal performance for each application.

**Application Notes:** **ELISA or ELISPOT Detection:** The biotinylated MP6-XT22 antibody is useful as a detection antibody for a sandwich ELISA or ELISPOT assay, when used in conjunction with purified 6B8 antibody (Cat. No. 510802/510804) as the capture antibody.

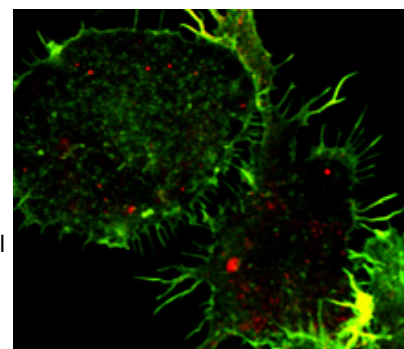
**Flow Cytometry<sup>6,11,12</sup>:** The fluorochrome-labeled MP6-XT22 antibody is useful for intracellular immunofluorescent staining and flow cytometric analysis to identify TNF- $\alpha$ -producing cells within mixed cell populations. To view the intracellular cytokine staining protocol, please visit [www.biolegend.com](http://www.biolegend.com) and click on the support section.

**Neutralization<sup>1,5,10</sup>:** The MP6-XT22 antibody can neutralize the bioactivity of natural or recombinant TNF- $\alpha$ . The LEAF™ purified antibody (Endotoxin <0.1 EU/ $\mu$ g, Azide-Free, 0.2  $\mu$ m filtered) is recommended for neutralization of mouse TNF- $\alpha$  bioactivity *in vivo* and *in vitro* (Cat. No. 506310). For *in vivo* studies or highly sensitive assays, we recommend Ultra-LEAF™ purified antibody (Cat. No. 506332) with a lower endotoxin limit than standard LEAF™ purified antibodies (Endotoxin <0.01 EU/ $\mu$ g).

**Additional reported applications (for the relevant formats) include:** Western blotting, immunohistochemical staining of paraformaldehyde-fixed, saponin-treated frozen tissue sections<sup>7-9</sup>, *in vivo* detection<sup>5</sup>, immunofluorescence, and immunocytochemistry.

**Note:** For testing mouse TNF- $\alpha$  in serum, plasma or supernatant, BioLegend's ELISA Max™ Sets (Cat. No. 430901 to 430906) are specially developed and recommended.

- Application References:**
1. Abrams J, *et al.* 1992. *Immunol. Rev.* 127:5. (Neut)
  2. Abrams J, *et al.* 1995. *Curr. Prot. Immunol.* John Wiley and Sons, New York. Unit 6.20
  3. Mo X, *et al.* 1995. *J. Virol.* 69:1288.
  4. Sarawar S, *et al.* 1994. *J. Immunol.* 153:1246.
  5. Via C, *et al.* 2001. *J. Immunol.* 167:6821. (Neut)
  6. Infante-Duarte C, *et al.* 2000 *J. Immunol.* 165:6107. (FC)
  7. Jacobs M, *et al.* 2000. *Immunology* 100:494. (IHC)
  8. Marinova-Mutachieva L, *et al.* 1997. *Clin. Exp. Immunol.* 107:507. (IHC)
  9. Williams RO, *et al.* 2000. *J. Immunol.* 165:7240. (IHC)
  10. Scanga CA, *et al.* 1999. *Infect. Immun.* 67:4531. (Neut)
  11. Akilov OE, *et al.* 2007. *J. Leukoc. Biol.* 2007;10.1189/jlb.0706439. (FC)
  12. Lawson BR, *et al.* 2007. *J. Immunol.* 178:5366. (FC)
  13. Patole PS, *et al.* 2005. *J. Am. Soc. Nephrol.* 16:3273. PubMed



Immortalized murine bone marrow-derived macrophages stimulated overnight with LPS were stained with Atto-488 phalloidin (green) and purified TNF- $\alpha$  (clone MP6-XT22), secondarily stained with Goat anti-Rat IgG Dylight 594 (red). Data provided by James Harris, Trinity College.



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14. Wu S, *et al.* 2005. *Neurosci Lett.* 394:158. PubMed  
 15. Carlson MJ, *et al.* 2009. *Blood* 113:1365. PubMed

**Description:** TNF- $\alpha$  is secreted by macrophages, monocytes, neutrophils, T-cells (principally CD4<sup>+</sup>), and NK-cells. Many transformed cell lines also secrete TNF- $\alpha$ . Monomeric mouse TNF- $\alpha$  is a 156 amino acid protein (N-glycosylated) with a reported molecular weight of 17.5 kD. TNF- $\alpha$  forms multimeric complexes; stable trimers are most common in solution. A 26 kD membrane form of TNF- $\alpha$  has also been described. TNF- $\alpha$  binding to surface receptors elicits a wide array of biologic activities including: cytolysis and cytostasis of many tumor cell lines *in vitro*, hemorrhagic necrosis of tumors *in vivo*, increased fibroblast proliferation, and enhanced chemotaxis and phagocytosis in neutrophils.

- Antigen References:** 1. Fitzgerald K, *et al.* Eds. 2001. The Cytokine FactsBook. Academic Press, San Diego.  
 2. Beutler B, *et al.* 1988. *Annu. Rev. Biochem.* 57:505.  
 3. Beutler B, *et al.* 1989. *Annu. Rev. Immunol.* 7:625.  
 4. Tracey K, *et al.* 1993. *Crit. Care Med.* 21:S415.

Related Products:	Product	Clone	Application
	Cell Staining Buffer		FC, ICC, ICFC
	Fixation Buffer		ICC, ICFC
	Permeabilization Wash Buffer (10X)		ICC, ICFC, IHC
	Brefeldin A Solution (1,000X)		ICFC
	Monensin Solution (1,000X)		ICFC
	Purified Rat IgG1, $\kappa$ Isotype Ctrl	RTK2071	FC, ICFC, ICC, IF, IHC, IP, WB



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