### **Product Data Sheet**

#### PE anti-mouse TNF-α

**Catalog # / Size:**  $3131525 / 25 \mu g$ 

3131530 / 100 μg

Clone: MP6-XT22 Isotype: Rat IgG1, κ

**Immunogen:** E. coli-expressed, recombinant mouse

 $\mathsf{TNF-}\alpha$ 

Reactivity: Mouse

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

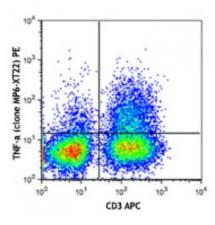
chromatography, and conjugated with PE under optimal conditions. The solution is free of unconjugated PE and

unconjugated antibody.

Formulation: Phosphate-buffered solution, pH 7.2,

containing 0.09% sodium azide.

**Concentration:** 0.2



PMA + Ionomycin-stimulated C57BL/6 mouse splenocytes (in the presence of monensin) were stained with CD3 APC, fixed, permeabilized and then stained with TNF- $\alpha$  (clone MP6-XT22) PE (top) or rat IgG1,  $\kappa$  PE isotype control (bottom).

#### **Applications:**

**Applications:** Flow Cytometry

Recommended

**Usage:** 

Each lot of this antibody is quality control tested by intracellular

immunofluorescent staining with flow cytometric analysis. For flow cytometric staining, the suggested use of this reagent is  $\leq 0.25$  microg per  $10^6$  cells in 100 microL volume. It is recommended that the reagent be titrated for optimal performance for each application.

Application Notes:

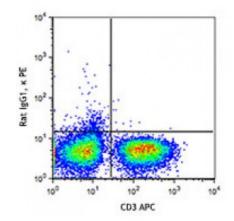
**ELISA or ELISPOT Detection:** The biotinylated MP6-XT22 antibody is

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-α-producing cells within mixed cell populations.

**Neutralization**<sup>1,5,10,16,17</sup>: The MP6-XT22 antibody can neutralize the bioactivity of natural or recombinant TNF-α. The LEAF<sup>™</sup> 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<sup>TM</sup> purified antibody (Cat. No. 506332) with a lower endotoxin limit than standard LEAF<sup>TM</sup> purified antibodies (Endotoxin <0.01 EU/microg).

# Additional reported applications (for the relevant formats) include:

Western blotting, immunohistochemical staining of paraformaldehyde-fixed, saponin-treated frozen tissue sections<sup>7</sup>-

<sup>9</sup>, *in vivo* detection5, immunofluorescence, and immunocytochemistry.

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

### Application References:

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- 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
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- 15. Carlson MJ, et al. 2009. Blood 113:1365. PubMed
- 16. Asrat S, 2014. PLoS Pathog. 10:1004229. PubMed
- 17. Lu X, et al. 2015. J Immunol. 194:2011. PubMed
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#### **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.