# **Product Data Sheet**

## Biotin anti-human TNF-α

Catalog # / Size: 3114520 / 500 µg

3114515 / 50 μg

Clone: MAb11

Isotype: Mouse IgG1, κ

E. coli-expressed, recombinant human Immunogen:

TNF-α

Reactivity: Human

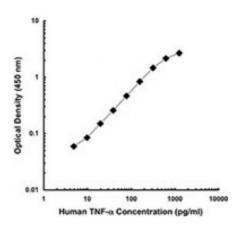
**Preparation:** The antibody was purified by affinity

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

Phosphate-buffered solution, pH 7.2, Formulation:

containing 0.09% sodium azide.

**Concentration:** 0.5



## **Applications:**

**Applications:** Other

Recommended

**Usage:** 

Each lot of this antibody is quality control tested by ELISA assay. For ELISA detection applications, a concentration range of 0.25-1.0 microg/ml is recommended. To obtain a linear standard curve, serial dilutions of TNF- $\alpha$ recombinant protein ranging from 500 to 4 pg/ml are recommended for each

ELISA plate. For use as an ELISPOT detection antibody, a concentration range of 0.5-2.0 microg/ml is recommended. 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 MAb11 antibody is useful as

the detection antibody in a sandwich ELISA or ELISPOT, when used in conjunction with the purified MAb1 antibody (Cat. No. 502802/502804) as

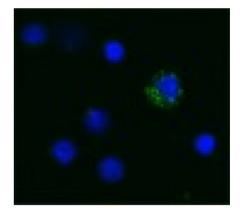
the capture antibody.

Flow Cytometry<sup>3,5,6,10</sup>: The

fluorochrome-labeled MAb11 antibody is useful for intracellular and membranebound immunofluorescent staining and flow cytometric analysis to identify TNFα-producing cells within mixed cell

populations.

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



Human PBMCs, stimulated with 1 microg/ml of LPS for 8 h and treated with Brefeldin A during the last 4 h, were prepared by cytospin, fixed and permeabilized on a slide and then treated with endogenous biotin blocking kit (Vector labs). Slides were stai

neutralization<sup>1,2</sup>, immunohistochemical staining of paraformaldehyde-fixed, saponin-treated frozen tissue sections4 and acetone-fixed frozen tissue sections<sup>8</sup>, immunocytochemistry<sup>7</sup>, and immunofluorescence<sup>9</sup>. The MAb11 antibody can neutralize the bioactivity of natural or recombinant TNF- $\alpha$ . **Note:** For testing human TNF- $\alpha$  in serum or plasma, BioLegend's ELISA Max<sup>™</sup> Sets (Cat. No. 430201 to 430206) are specially developed and recommended. The LEAF™ purified antibody (Endotoxin < 0.1 EU/µg, Azide-Free, 0.2 µm filtered) is recommended for neutralization of human TNF-α bioactivity (Cat. No. 502922).

The Purified MAb1 antibody is useful in neutralization2 and as the capture antibody in a sandwich ELISA or ELISPOT assay, when used in conjunction with the biotinylated MAb11 antibody (Cat. No. 502904/502914) as the detecting antibody.

# Application References:

- 1. Rathjen D, et al. 1991. Mol. Immunol. 28:79. (Neut)
- 2. Danis V, et al. 1991. Clin. Exp. Immunol. 85:143. (Neut)
- 3. Enr quez J, et al. 2002. Adv. Perit. Dial. 18:177. (ICFC)
- 4. Andersson U, et al. 1999. Detection and quantification of gene expression. New York:Springer-Verlag. (IHC)
- 5. Chen H, et al. 2005. J. Immunol. 175:591. (ICFC)
- 6. Iwamoto S, et al. 2007. J. Immunol. 179:1449. (ICFC) PubMed
- 7. Andersson U, et al. 2000. J. Exp. Med. 192:565. (ICC)
- 8. Moormann AM, et al. 1999. J. Infect. Dis. 180:1987. (IHC)
- 9. Zhao XJ, et al. 2003. J. Immunol. 170:2923. (IF)
- 10. Rieger R, et al. 2009. Cancer Gene Ther. 1:53-64. (FC)

#### **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 human TNF- $\alpha$  is a 157 amino acid protein (non-glycosylated) with a reported molecular weight of 17 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 biological activities including: cytolysis and cytostasis of many tumor cell lines *in vitro*, hemorraghic 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.