SONY

Product Data Sheet

Brilliant Violet 421™ anti-mouse TNF-α

Catalog # / Size: 3131640 / 50 µg
3131635 / 125 µl

Clone: MP6-XT22

Isotype: Rat IgG1, κ

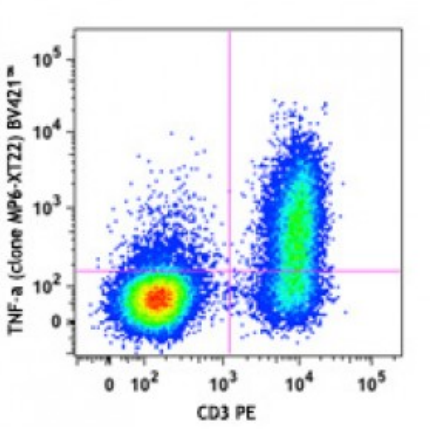
Immunogen: *E. coli*-expressed, recombinant mouse TNF-α

Reactivity: Mouse

Preparation: The antibody was purified by affinity chromatography and conjugated with Brilliant Violet 421™ under optimal conditions. The solution is free of unconjugated Brilliant Violet 421™ and unconjugated antibody.

Formulation: Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide and BSA (origin USA).

Concentration: microg sizes: 0.2 mg/ml
microL sizes: lot-specific

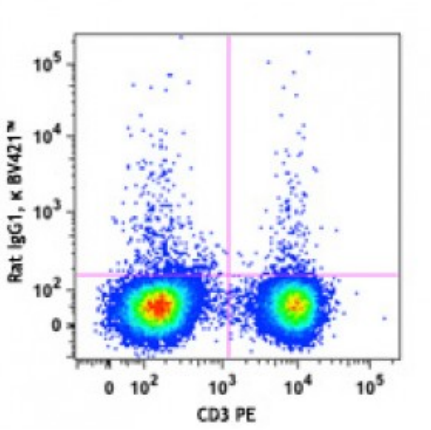


PMA + Ionomycin-stimulated C57BL/6 mouse splenocytes (in the presence of monensin) were stained with CD3 PE, fixed, permeabilized and then stained with TNF-α (clone MP6-XT22) Brilliant Violet 421™ (top) or rat IgG1, κ Brilliant Violet 42

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 using the microg size, the suggested use of this reagent is ≤0.25 microg per million cells in 100 microL volume. For flow cytometric staining using the microL size, the suggested use of this reagent is ≤5 microL per million cells or 5 microL per 100 microL of whole blood. It is recommended that the reagent be titrated for optimal performance for each application.



Brilliant Violet 421™ excites at 405 nm and emits at 421 nm. The standard bandpass filter 450/50 nm is recommended for detection. Brilliant Violet 421™ is a trademark of Sirigen Group Ltd.

This product is subject to proprietary rights of Sirigen Inc. and is made and sold under license from Sirigen Inc. The purchase of this product conveys to the buyer a non-transferable right to use the purchased product for research purposes only. This product may not be

resold or incorporated in any manner into another product for resale. Any use for therapeutics or diagnostics is strictly prohibited. This product is covered by U.S. Patent(s), pending patent applications and foreign equivalents.

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[6,11,12]: 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[1,5,10,16,17]: The MP6-XT22 antibody can neutralize the bioactivity of natural or recombinant TNF-α. The LEAF™ purified antibody (Endotoxin <0.1 EU/µg, Azide-Free, 0.2 µm filtered) is recommended for neutralization of mouse TNF-α 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/microg).

Additional reported applications (for the relevant formats) include: Western blotting, immunohistochemical staining of paraformaldehyde-fixed, saponin-treated frozen tissue sections[7-9], *in vivo* detection5, immunofluorescence, and immunocytochemistry.

Note: For testing mouse TNF-α 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
14. Wu S, *et al.* 2005. *Neurosci Lett.* 394:158. PubMed
15. Carlson MJ, *et al.* 2009. *Blood* 113:1365. PubMed

Description: TNF-α is secreted by macrophages, monocytes, neutrophils, T-cells (principally $CD4^+$), and NK-cells. Many transformed cell lines also secrete TNF-α. Monomeric mouse TNF-α is a 156 amino acid protein (N-glycosylated) with a reported molecular weight of 17.5 kD. TNF-α forms multimeric complexes; stable trimers are most common in solution. A 26 kD membrane form of TNF-α has also been described. TNF-α 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.