

ORDERING INFORMATION

Catalog Number: MAB4708

Clone: 488806

Lot Number: CBFX01

Size: 100 µg

Formulation: 0.2 µm filtered solution in PBS with 5% trehalose

Storage: -20° C

Reconstitution: sterile PBS

Specificity: human TRIM

Immunogen: *E. coli*-derived rhTRIM

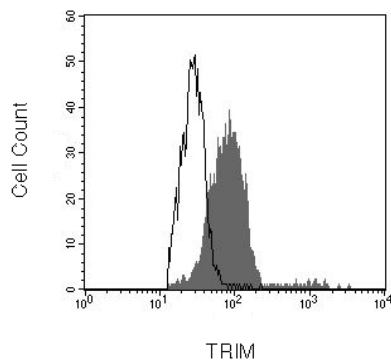
Ig class: mouse IgG₁

Recommended Applications:

Flow cytometry
Western blot

Other Application:

Direct ELISA



CD3⁺ blood-derived lymphocytes were stained with anti-TRIM (R&D Systems, Cat. # MAB4708, clone 488806, filled histogram) or isotype control (R&D Systems, Cat. # MAB002, open histogram) followed by APC-conjugated anti-mouse antibody (R&D Systems, Cat. # F0101B).

Background

Human TRIM (T cell receptor-interacting molecule) (also trat1, T cell receptor-associated transmembrane adaptor 1, and pp29/30) is a 30 kDa, type III transmembrane protein, that is a member of the transmembrane adaptor protein (TRAP) family. It contains a short, 8 aa extracellular region, a 19 aa transmembrane region, and a 159 aa cytosolic tail. Its cytoplasmic tail contains several tyrosine motifs with the potential to bind to Src-homology 2 (SH2) domains of signaling proteins. TRIM is present in T cells and NK cells. Human TRIM shares 66% aa sequence identity with mouse TRIM.

Preparation

This antibody was produced from a hybridoma resulting from the fusion of a mouse myeloma with B cells obtained from a mouse immunized with purified, *E. coli*-derived, recombinant human TRIM (rhTRIM; aa 50 - 186; Accession # Q6PIZ9). The IgG fraction of the tissue culture supernatant was purified by Protein G affinity chromatography.

Formulation

Lyophilized from a 0.2 µm filtered solution in phosphate-buffered saline (PBS) with 5% trehalose.

Reconstitution

Reconstitute with sterile PBS. If 0.2 mL of PBS is used, the antibody concentration will be 500 µg/mL.

Storage

Lyophilized samples are stable for twelve months from date of receipt when stored at -20° C to -70° C. Upon reconstitution, the antibody can be stored at 2° - 8° C for 1 month without detectable loss of activity. Reconstituted antibody can also be aliquotted and stored frozen at -20° C to -70° C in a manual defrost freezer for six months without detectable loss of activity. **Avoid repeated freeze-thaw cycles.**

Specificity

This antibody detects rhTRIM in direct ELISAs and Western blots.

Applications

Intracellular Flow cytometry - This antibody was tested for flow cytometry using CD3⁺ blood-derived lymphocytes. For intracellular staining to detect TRIM, cells must first be fixed and permeabilized using 4% paraformaldehyde and 0.1% saponin in PBS. Dilute this antibody to 25 µg/mL and add 10 µL of the diluted solution to 1 - 5 x 10⁵ cells in a total reaction volume not exceeding 200 µL. The binding of unlabeled monoclonal antibodies may be visualized by adding a secondary developing reagent such as anti-mouse IgG conjugated to a fluorochrome.

Western blot - This antibody can be used at 1 - 2 µg/mL with the appropriate secondary reagents to detect human TRIM. Using a colorimetric detection system, the detection limit for rhTRIM is approximately 50 ng/lane under non-reducing and reducing conditions. Chemiluminescent detection will increase sensitivity by 5 to 50 fold.

Direct ELISA - This antibody can be used at 0.5 - 1.0 µg/mL with the appropriate secondary reagents to detect human TRIM. The detection limit for rhTRIM is approximately 1 ng/well.

Optimal dilutions should be determined by each laboratory for each application.