BV421 Mouse Anti-Human CD154

Product Information

Material Number: 566268
Alternate Name: CD40LG; CD40 ligand; CD40L; TNFSF5; T-BAM; TRAP
Size: 25 Tests
Vol. per Test: 5 µl
Clone: TRAP1 (also known as TRAP-1)
Immunogen: Human TRAP1 Transfected Cell Line
Isotype: Mouse (BALB/c) IgG1, κ
Reactivity: QC Testing: Human
Tested in Development: Rhesus, Cynomolgus, Baboon
Workshop: VI 6T-068
Storage Buffer: Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The TRAP1 monoclonal antibody specifically binds to CD154. CD154 is a 39 kDa type II membrane glycoprotein that is a member of the tumor necrosis factor superfamily. Tumor necrosis factor ligand superfamily member 5 (TNFSF5). CD154 is expressed on a variety of cell types including activated CD4+ T cells and some CD8+ T cells, NK cells, mast cells and basophils. CD154 is also known as CD40 ligand (CD40L); it serves as a ligand for CD40 that is expressed on B cells, macrophages, and dendritic cells. The expression of CD154 by activated T-helper cells costimulates B-cell activation and proliferation through binding to CD40 expressed on B cells. In response to T-dependent antigens, the CD154 and CD40 interaction is required for B-lymphocyte differentiation, including immunoglobulin production and isotype switching and memory B cell generation. The TRAP1 antibody can partially block T cell-B cell interaction and inhibit the subsequent proliferation, differentiation, and memory formation of B cells. It has been reported that patients with X-linked hyper-IgM syndrome have defective expression of functional CD154 due to mutations in the CD40LG gene that encodes CD154.

The antibody was conjugated to BD Horizon BV421 which is part of the BD Horizon Brilliant™ Violet family of dyes. With an Ex Max of 407-nm and Em Max at 421-nm, BD Horizon BV421 can be excited by the violet laser and detected in the standard Pacific Blue™ filter set (eg, 450/50-nm filter). BD Horizon BV421 conjugates are very bright, often exhibiting a 10 fold improvement in brightness compared to Pacific Blue conjugates.

Flow cytometric analysis of CD154 expression on stimulated human peripheral blood lymphocytes. Human peripheral blood mononuclear cells were stimulated for 4 hours with 20 ng/mL Phorbol 12-Myristate 13-Acetate (PMA; Sigma-Aldrich Cat. No. P-8139) and 250 ng/mL Calcium Ionophore A23187 (Sigma-Aldrich Cat. No. C-9278). The cells were then stained with either BD Horizon™ BV421 Mouse IgG1, κ Isotype Control (Cat. No. 562438; dashed line histogram) or BD Horizon BV421 Mouse Anti-Human CD154 antibody (Cat. No. 563886/566268; solid line histogram). The flow cytometric fluorescence histograms were derived from gated events with the forward and side light-scatter characteristics of viable lymphocytes. Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer System.

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated with BD Horizon™ BV421 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV421 were removed.

Application Notes

Application

Flow cytometry Routinely Tested
Recommended Assay Procedure:
For optimal and reproducible results, BD Horizon Brilliant Stain Buffer should be used anytime two or more BD Horizon Brilliant dyes are used in the same experiment. Fluorescent dye interactions may cause staining artifacts which may affect data interpretation. The BD Horizon Brilliant Stain Buffer was designed to minimize these interactions. More information can be found in the Technical Data Sheet of the BD Horizon Brilliant Stain Buffer (Cat. No. 563794).

Suggested Companion Products

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<tr>
<th>Catalog Number</th>
<th>Name</th>
<th>Size</th>
<th>Clone</th>
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<tbody>
<tr>
<td>554656</td>
<td>Stain Buffer (FBS)</td>
<td>500 mL</td>
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<tr>
<td>554657</td>
<td>Stain Buffer (BSA)</td>
<td>500 mL</td>
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<tr>
<td>563794</td>
<td>Brilliant Stain Buffer</td>
<td>100 Tests</td>
<td>(none)</td>
</tr>
<tr>
<td>563886</td>
<td>BV421 Mouse Anti-Human CD154</td>
<td>100 Tests</td>
<td>TRAP1</td>
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<tr>
<td>562438</td>
<td>BV421 Mouse IgG1, k Isotype Control</td>
<td>50 µg</td>
<td>X40</td>
</tr>
</tbody>
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Product Notices
1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10^6 cells in a 100-µl experimental sample (a test).
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
4. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
5. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
6. Pacific Blue™ is a trademark of Molecular Probes, Inc., Eugene, OR.
7. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.
8. BD Horizon Brilliant Stain Buffer is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,575,303; 8,354,239.
9. BD Horizon Brilliant Violet 421 is covered by one or more of the following US patents: 8,158,444; 8,362,193; 8,575,303; 8,354,239.

References