APC-H7 Mouse Anti-Human CD40

Product Information

Material Number: 561211
Alternate Name: TNFRSF5; TNF receptor superfamily member 5; CD40L receptor; Bp50; p50
Size: 50 Tests
Vol. per Test: 5 µl
Clone: 5C3
Isotype: Mouse IgG1, κ
Reactivity: QC Testing: Human
Workshop: V CD40.4
Storage Buffer: Aqueous buffered solution containing BSA, protein stabilizer, and ≤0.09% sodium azide.

Description

This 5C3 monoclonal antibody specifically binds to CD40, a 45-48 kDa type I integral membrane glycoprotein. CD40 is expressed on B lymphocytes, but is not expressed on terminally differentiated B cells. CD40 is also expressed by endothelial cells, basal epithelial cells and some epithelial cell carcinomas, follicular dendritic cells, macrophages, fibroblasts, keratinocytes, and CD34+ hematopoietic progenitor cells. This antibody is useful for studying the roles played by CD40 in B-cell growth, proliferation, and differentiation including immunoglobulin isotype switching. Anti-CD40 antibodies have been reported to stimulate B-cell proliferation when costimulated with anti-µ, anti-CD20 antibodies or with phorbol esters. 5C3 is capable of inducing B-cell proliferation when presented with IL-4.

Clone 5C3 reacts with the human form of the 45-48 kDa type I integral membrane glycoprotein, CD40. This clone also cross-reacts with a subset of peripheral blood lymphocytes, but not monocytes nor granulocytes, of baboon and both rhesus and cynomolgus macaque monkeys. The distribution on lymphocytes is similar to that seen with normal human donor lymphocytes, with the reactivity being restricted to CD20+ lymphocytes.

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 APC-H7 under optimum conditions, and unconjugated antibody and APC-H7 were removed.

Application Notes

Application

| Flow cytometry | Routinely Tested |

Flow cytometric analysis of CD40 expression on human peripheral blood lymphocytes. Whole blood was stained with APC-H7 Mouse Anti-Human CD40 antibody (Cat. No. 561211; solid line histogram) or with a APC-H7 Mouse IgG1, κ Isotype Control (Cat. No. 560167; dashed line histogram). The erythrocytes were lysed with BD PharmLyse™ Lysing Buffer (Cat. No. 555899). The fluorescence histograms were derived from events with the forward and side light-scatter characteristics of viable lymphocytes. Flow cytometry was performed using a BD™ LSR II Flow Cytometer System.

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Suggested Companion Products

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<th>Catalog Number</th>
<th>Name</th>
<th>Size</th>
<th>Clone</th>
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<tr>
<td>560167</td>
<td>APC-H7 Mouse IgG1, κ Isotype Control</td>
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<td>MOPC-21</td>
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<tr>
<td>555899</td>
<td>Lysing Buffer</td>
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<td>554656</td>
<td>Stain Buffer (FBS)</td>
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<td>554657</td>
<td>Stain Buffer (BSA)</td>
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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 × 10⁶ 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. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
6. BD APC-H7 is a tandem conjugate and an analog of APC-Cy7 with the same spectral properties. It has decreased intensity but it is engineered for greater stability and less spillover in the APC channel and consequently offers better performance than APC-Cy7. It has an absorption maximum of approximately 650 nm. When excited by light from a red laser, the APC fluorochrome can transfer energy to the cyanine dye, which then emits at a longer wavelength. The resulting fluorescent emission maximum is approximately 767 nm. BD recommends that a 750-nm longpass filter be used along with a red-sensitive detector such as the Hamamatsu R3896 PMT. As with APC-Cy7 special filters are required when using APC-H7 in conjunction with APC.
7. Note: Although our APC-H7 products demonstrate higher lot-to-lot consistency than other APC tandem conjugate products, and every effort is made to minimize the lot-to-lot variation in residual emission from APC, it is strongly recommended that every lot be tested for differences in the amount of compensation required and that individual compensation controls are run for each APC-H7 conjugate.
8. Although BD APC-H7 is engineered to minimize spillover to the APC channel and is more stable and less affected by light, temperature, and formaldehyde-based fixatives, compared to other APC-cyanine tandem dyes, it is still good practice to minimize as much as possible, any light, temperature and fixative exposure when working with all fluorescent conjugates.
9. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.

References
Stamenkovic I, Clark EA, Seed B. A B lymphocyte activation molecule related to the nerve growth factor receptor and induced by cytokines in carcinomas. *EMBO J.* 1989; 8(5):1403-1410. (Biology)