Technical Data Sheet

Purified Rat Anti-Mouse CD40

Product Information

Material Number: 553788
Size: 0.5 mg
Concentration: 0.5 mg/ml
Clone: 3/23
Immunogen: Mouse CD40 Recombinant Protein
Isotype: Rat (LOU) IgG2a, κ
Reactivity: QC Testing: Mouse
Storage Buffer: Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The 3/23 clone reacts with CD40, a 40-50 kDa glycoprotein expressed on B lymphocytes and other antigen-presenting cells. CD40 has been reported to be transiently expressed on activated CD4+ and CD8+ T cells and in some mouse strains, the 3/23 mAb has been reported to react with 5-10% of T lymphocytes in adult mouse, but not neonatal, spleen. CD40 plays a key role in B-cell growth and differentiation where interactions of CD40 with its ligand, CD154, are involved in the initiation, effector, and memory stages of cell-mediated immune responses. In addition, CD40 has been reported to be involved in the triggering of NK cells and NK-T cells. Ligation of CD40 with the 3/23 antibody has been reported to induce splenic B cells to express the costimulatory molecule CD86 (B7-2). In addition, although the 3/23 antibody by itself is a weak B-cell mitogen, it has been reported to synergize markedly with mitogenic anti-IgM, anti-IgD mAb or IL-4 to promote B-cell proliferation.

This antibody is routinely tested by flow cytometric analysis. Other applications were tested at BD Biosciences Pharmingen during antibody development only or reported in the literature.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. Store undiluted at 4°C.

Two-color analysis of the expression of CD40 on mouse spleen cells. BALB/c splenocytes were simultaneously stained with PE-conjugated anti-mouse CD3e mAb 145-2C11 (Cat. No. 553063) and purified mAb 3/23 (right panel), followed by FITC-conjugated anti-rat Ig, κ light chain mAb MRK-1 (Cat. No. 553872). Please note that staining of a T-cell subset by mAb 3/23 has not been consistently observed. Flow cytometry was performed on a BD FACScan™ flow cytometry system.
**Application Notes**

**Application**

<table>
<thead>
<tr>
<th>Method</th>
<th>Status</th>
</tr>
</thead>
<tbody>
<tr>
<td>Flow cytometry</td>
<td>Routinely Tested</td>
</tr>
<tr>
<td>(Co)-stimulation</td>
<td>Reported</td>
</tr>
<tr>
<td>Immunohistochemistry-frozen</td>
<td>Reported</td>
</tr>
<tr>
<td>Immunohistochemistry-formalin (antigen retrieval required)</td>
<td>Not Recommended</td>
</tr>
</tbody>
</table>

**Recommended Assay Procedure:**

**Caution:** Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE™ (No Azide/Low Endotoxin) antibody format for in vitro and in vivo use.

**Suggested Companion Products**

<table>
<thead>
<tr>
<th>Catalog Number</th>
<th>Name</th>
<th>Size</th>
<th>Clone</th>
</tr>
</thead>
<tbody>
<tr>
<td>553872</td>
<td>FITC Mouse Anti-Rat Ig κ</td>
<td>0.5 mg</td>
<td>MRK-1</td>
</tr>
<tr>
<td>553063</td>
<td>PE Hamster Anti-Mouse CD3ε</td>
<td>0.1 mg</td>
<td>145-2C11</td>
</tr>
<tr>
<td>553927</td>
<td>Purified Rat IgG2a κ Isotype Control</td>
<td>0.5 mg</td>
<td>R35-95</td>
</tr>
<tr>
<td>553787</td>
<td>Purified NA/LE Rat Anti-Mouse CD40</td>
<td>0.5 mg</td>
<td>3/23</td>
</tr>
<tr>
<td>550285</td>
<td>Purified Rat Anti-Mouse CD40</td>
<td>1.0 ml</td>
<td>3/23</td>
</tr>
</tbody>
</table>

**Product Notices**

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
3. 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.

**References**


Parry SJ, Hasbold J, Holman MJ, Klaus GG. Hypercross-linking surface IgM or IgD receptors on mature B cells induces apoptosis that is reversed by costimulation with IL-4 and anti-CD40. *J Immunol*. 1994; 152(6):2821-2829. (Biology)

