Human CD40/TNFRSF5 Antibody

Monoclonal Mouse IgG_{2B} Clone # 82102

Catalog Number: MAB6322

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human CD40 in direct ELISAs and Western blots. In direct ELISAs and Western blots, no cross-reactivity with recombinant human (rh) 4-1BB, rhBAFF R, rhCD27, rhCD30, rhDR3, rhDR6, rhEDAR, rhFas, rhGITR, rhHVEM, rhNGF R, rhOPG, rhRANK, rhTAJ, rhTNF RI, or rhTNF RII is observed.
Source	Monoclonal Mouse IgG_{2B} Clone # 82102
Purification	Protein A or G purified from ascites
Immunogen	Mouse myeloma cell line NS0-derived recombinant human CD40/TNFRSF5 Glu21-Arg193 Accession # P25942
Endotoxin Level	<0.10 EU per 1 µg of the antibody by the LAL method.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

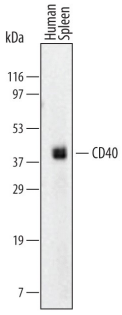
APPLICATIONS

Please Note: *Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.*

	Recommended Concentration	Sample
Western Blot	2 µg/mL	See Below
Neutralization	Measured by its ability to neutralize CD40 Ligand/TNFSF5-induced proliferation in human B cell enriched peripheral blood mononuclear cells (PBMC). At 5 µg/mL, this anti-hCD40 antibody will neutralize approximately 80% of 10 µg/mL Recombinant Human CD40 Ligand/TNFSF5 -induced proliferation of human B-cell enriched PBMCs.	

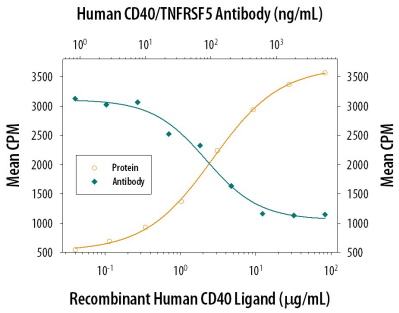
DATA

Western Blot



Detection of CD40/TNFRSF5 Mouse by Western Blot. Western blot shows lysates of human spleen tissue. PVDF membrane was probed with 2 µg/mL of Mouse Anti-Human CD40/TNFRSF5 Monoclonal Antibody (Catalog # MAB6322) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF007). A specific band was detected for CD40/TNFRSF5 at approximately 40 kDa (as indicated). This experiment was conducted under non-reducing conditions and using Immunoblot Buffer Group 1.

Neutralization



Cell Proliferation Induced by CD40 Ligand/TNFSF5 and Neutralization by Human CD40/TNFRSF5 Antibody. In the presence of Recombinant Human IL-4 (Catalog # 204-IL), Recombinant Human CD40 Ligand/TNFSF5 (Catalog # 6245-CL) stimulates proliferation in human B cell enriched peripheral blood mononuclear cells (PBMC) in a dose-dependent manner (orange line). Proliferation elicited by Recombinant Human CD40 Ligand/TNFSF5 (10 µg/mL) is neutralized (green line) by increasing concentrations of Human CD40/TNFRSF5 Monoclonal Antibody (Catalog # MAB6322). At 5 µg/mL, this anti-hCD40 antibody will neutralize approximately 80% of Recombinant Human CD40 Ligand/TNFSF5 -induced proliferation in the presence of Recombinant Human IL-4 (20 ng/mL).

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	**Use a manual defrost freezer and avoid repeated freeze-thaw cycles.** ● 12 months from date of receipt, -20 to -70 °C as supplied. ● 1 month, 2 to 8 °C under sterile conditions after reconstitution. ● 6 months, -20 to -70 °C under sterile conditions after reconstitution.

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Human CD40/TNFRSF5 Antibody

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Catalog Number: MAB6322

BACKGROUND

CD40 is a type I transmembrane glycoprotein belonging to the TNF receptor superfamily. Mature human CD40 consists of a 172 amino acid (aa) extracellular domain, a 22 aa transmembrane region and a 62 aa cytoplasmic domain (1). Human and mouse CD40 share 62% aa identity. CD40 is expressed in B cells, follicular dendritic cells, dendritic cells, activated monocytes, macrophages, endothelial cells, vascular smooth muscle cells, and several tumor cell lines (2). The extracellular domain has the cysteine-rich repeat regions, which are characteristic for many of the receptors of the TNF superfamily. Interaction of CD40 with its ligand, CD40L, leads to aggregation of CD40 molecules, which in turn interact with cytoplasmic components to initiate signaling pathways. Early studies on the CD40-CD40L system revealed its role in humoral immunity. Interaction between CD40L on T cells and CD40 on B cells stimulated B cell proliferation and provided the signal for immunoglobulin isotype switching (3). Mutations in the CD40L gene, which resulted in a CD40L molecule unable to interact with CD40, are responsible for the hyper-IgM syndrome (4). Cross-linking of CD40 with antibodies or by CD40 binding to CD40L produces cell type-specific responses which include costimulation and induction of proliferation, induction of cytokine production, rescue from apoptosis, and upregulation of adhesion molecules (5). Some of the early events of intracellular signaling by the CD40-CD40L system include the association of the CD40 with TRAFs and the activation of various kinases (6-8).

References:

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