3050 Spruce Street
Saint Louis, Missouri 63103 USA
Telephone 800-325-5832 • (314) 771-5765
Fax (314) 286-7828
email: techserv@sial.com
sigma-aldrich.com

ProductInformation

ANTI-HUMAN CD40
Developed in Goat,
Affinity Isolated Antibody

Product Number **C9099**

Product Description

Anti-Human CD40 is developed in goat using a purified recombinant human CD40, extracellular domain, expressed in mouse NSO cells as immunogen. The antibody is purified using human CD40 affinity chromatography.

Anti-Human CD40 recognizes recombinant human CD40 by ELISA and immunoblotting. By immunoblotting and ELISA, the antibody does not cross react with recombinant mouse CD40, recombinant human soluble TNF RI, and recombinant human soluble TNF RII and recombinant human CD40 ligand.

Anti-Human CD40 may be used in ELISA and immunoblotting.

CD40 (48-50 kDa) is a transmembrane glycoprotein mainly expressed on the surface of B cells and is also expressed on monocytes, dendritic cells, endothelial cells, and epithelial cells.[1-4] CD40 is a member of the tumor necrosis factor (TNF)[1,2] receptor superfamily, which includes the low affinity nerve growth factor (NGF) receptor and CD95/Fas.[4,5] CD40 ligand (CD40L, CD154, gp39, TRAM) belongs to the TNF gene family[1-3] and is expressed more widely than activated CD4+ T cells. Following interaction with CD40L, CD40 mediates a number of major immunoregulatory functions, is central to the control of thymus-dependent humoral immunity and may be critical in the development of cell-mediated immune responses. Other biological actions include B cell homotypic adhesion, proliferation, immunoglobulin isotype switch, and secretion.[1-4] Activation of CD40 has also been shown to inhibit the growth of certain B cell lymphoma[6-9] and to induce the death of transformed cells of mesenchymal or epithelial origin.[10-12]

In their resting state up to 50% of B cells may express CD40L in their cytoplasm, but not on the surface and this cytoplasmic CD40L is readily released as a soluble receptor. The proportion of cells expressing and the amount of CD40L is increased by signaling through immunoglobulin (Ig) or CD38.[13]

The signal transduction pathways triggered through CD40 have not yet been fully delineated. Early biochemical events include activation of Ras,[14] of Lyn and Syk protein tyrosine kinases,[15-17] phosphorylation and activation of phospholipase C2 and phosphoinositide-3-kinase,[16] and induction of mitogen-activated protein kinases.[18-20] Activation processes seem to be initiated by association of the CD40 receptor with signaling molecules of the TRAF family.[1,2]

Reagents

Anti-Human CD40 is supplied lyophilized from a 0.2 µm filtered solution of phosphate buffered saline. Endotoxin level is < 10 ng per mg antibody as determined by the LAL method.

Preparation Instructions

To one vial of lyophilized powder, add 1 ml of 0.2 µm-filtered PBS to produce a 0.1 mg/ml stock solution of antibody. If aseptic technique is used, no further filtration should be needed for use in cell culture environments

Storage/Stability

Prior to reconstitution, store at –20 °C. Reconstituted product may be stored at 2-8 °C for at least one month. For prolonged storage, freeze in working aliquots at –20 °C. Avoid repeated freezing and thawing

Product Profile

For indirect ELISA, a working concentration of

0.5-1.0 µg/ml is determined to detect a limit of

0.03 ng/well of recombinant human CD40.

For indirect immunoblotting, a working concentration of 0.1-0.2 µg/ml is determined using human CD40 at 2 ng/lane and 25 ng/lane under non-reducing and reducing conditions, respectively.

Note: In order to obtain best results in different techniques and preparations we recommend determining optimal working dilutions by titration test.

References

1. van Kooten, C., and Banchereau, J. Curr. Opin. Immunol. **9**, 330-337 (1997).
2. van Kooten, C., and Banchereau, J., J. Leukoc. Biol., **67**, 2-17 (2000).
3. Foy, T. M., et al., Annu. Rev. Immunol. **14**, 591-617 (1996).
4. Grewal, I. S., and Flavell, R. A. Immunol. Today, **17**, 410-414 (1996).
5. Smith, C. A., et al., Cell, **76**, 959-962 (1994).
6. Stamenkovic, I., et al., EMBO J. **8**, 1403-1410 (1989)
7. Heath, A. W., et al. Cell Immunol. **152**, 468-480 (1993).
8. Funakoshi, S., et al., Blood, **83**, 2787-2794 (1994).
9. Baker, M. P., et al., Blood, **92**, 2830-2843 (1998).
10. Goldstein, M. D., and Watts, T. H. J. Immunol., **157**, 2837-2843 (1996).
11. Eliopoulos, A. G., et al., Oncogene, **13**, 2243-2254 (1996).
12. Hess, S., and Engelmann, H., J. Exp. Med., **183**, 159-167 (1996).
13. Bergamo, A., et al., Br. J. Haematol., **97**, 652-655 (1997).
14. Wykes M, et al., Eur. J. Immunol., **28**, 548-559 (1998).
15. Gulbins, E., et al., J. Immunol. **157**, 2844-2850 (1996).
16. Uckun, F. M., et al., J. Biol. Chem., **266**, 17478-17485 (1991).
17. Ren, C. L., et al., J. Exp. Med., **179**, 673-680 (1994).
18. Faris, M., et al., J. Exp. Med., **179**, 1923-1931 (1994).
19. Sakata, N., et al., J. Biol. Chem., **270**, 30823-30828 (1995).
20. Berberich, I., et al., EMBO J., **15**, 92-101 (1996). Sutherland, C. L., et al., J. Immunol., **157**, 3381-3390 (1996).

j.o/lpg 3/00