

## anti-CD40 antibody, mouse monoclonal (5C3)

## 72-030 100 μg

CD40 is a 45-50-kDa glycoprotein belonging to the tumor necrosis factor (TNF) receptor superfamily. CD40 is specifically expressed on the surface of B cells and specialized antigen-presenting cells such as dendritic cells and macrophages. CD40 interacts with the CD40 ligand (CD154) which is found primarily on T cells, playing a role in both humoral and cell-mediated immune responses. Activation of CD40 on B cells by CD40 ligand causes B cell proliferation, differentiation, immunoglobulin isotype switching, germinal center formation, and stimulation of the humoral memory response. CD40 has been found to mediate a broad variety of immune and inflammatory responses. Within the cell, the CD40 molecule acts as a transmembrane signal transducer that leads to activation of intracellular kinases and transcription factors.

## Applications:

- 1. Flow cytometry
- 2. Western Blotting (1/1,000)
- 3. Immunofluorescence staining and Immunocytochemistry (1/100)
- 4. Immunohistochemistry (acetone or zinc-fixed or frozen section; indirect immuno-staining) (1/100)
- 5. . Stimulation of proliferation of B cell and dendritic cells  $\,$  (20  $\,\mu$  g/ml). Enhancement of proinflamentory cytokine production in human monocytes, such as TNF-a, IL-6 and IL-8

**Isotype:** Mouse IgG1κ

Immunogen: Recombinant extracellular domain of CD40

**Product;** Purified from serum-free medium by salting-out and ion-exchange chromatography under mild conditions.

Form: 1mg/ml in PBS, 50% glycerol, filter-sterilized. Azide- and carier-free

Specificity: Human

**Storage:** Shipped at  $4^{\circ}$ C or with ice-pack and stored at  $-20^{\circ}$ C

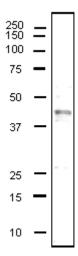
Data Link: Swiss-Prot P25942

**References:** This antibody is used in ref.2 and 3.

- Inui S et al (1990) "Identification of the intracytoplasmic region essential for signal transduction through a B cell activation molecule, CD40." Eur J Immunol 20: 1747-1753 PMID: 1698631
- 2. Yasui T *et al* (2002) "Dissection of B cell differentiation during primary immune responses in mice with altered CD40 signals." *Int Immunol* **14**: 319-329 PMID: 11867568 **FC**



3. Ishida I *et al* (2003) "Involvement of CD100, a lymphocyte semaphoring, in the activation of the human immune system via CD72: implications for the regulation of immune and inflammatory responses." *Int Immunol.* **15**: 1027-1034 PMID: <u>12882840</u> FC, Stimulation of B cell prolification.



## Fig.1 Detection of human CD40 in whole cell extract of MCF7 by western blot using anti-CD40 antibody (clone 5C3).

Proteins in the extract were separated by 12.5% SDS-PAGE and blotted by wet system. First antibody (5C3) was used at 1/1,000 dilution and  $2^{\rm nd}$  antibody, HRP conjugated goat anti-mouse IgG was used at 1/10,000 dilution.

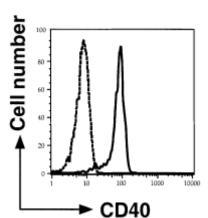


Fig.2 Expression of CD40 on the cell suface of monocyte derived dendric cells as analyzed by Flow Cytometry with anti-CD40 antibody (5C3).

Monocyte-derived dendric cells from healthy adult donor were stained with anti-hCD40 antibody (solid line) or with isotype control (broken line).

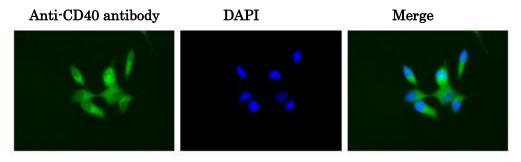


Fig.3 Immunofluorescent staining of CD40 in MCF7 cells with anti-CD40 antibody (5C3) Cells were fixed in 4% paraformaldehyde and permeabilized in 0.25% Triton X-100. Anti-CDCD40 was used at 1/100 dilution and as 2<sup>nd</sup> antibody, Alex 488 conjugated goat anti-mouse IgG was used at 1/1,000 dilution. DNA was stained with DAPI.



Related Products: #72-031 anti-CD40 antibody (5C3), Biotin-conjugated. #72-031 anti-CD40 antibody (5C3), FITC-conjugated.