

CD40 (EA-5): sc-65264

BACKGROUND

Resting B cells can be activated and clonally expanded into antibody-producing cells in response to a combination of cell contact and soluble signals provided by primed helper T (Th) cells. While cytokines IL-4 and IL-13 alone are inadequate for B cell activation, contact with Th cells seems to be sufficient for delivery of proliferative signals. A receptor ligand pair central to the transmission of this signal is CD40, expressed on the surface of B cells, together with CD40L, expressed on activated T cells. In the presence of such stimulus, IL-4 and IL-13 are capable of triggering immunoglobulin class switching and secretion of IgE. B cells are sensitive to these cytokines only subsequent to CD40/CD40L-driven DNA synthesis. A downstream mediator of the CD40 signaling pathway, designated CRAF, is a member of an expanding family of proteins that contain a conserved cysteine- and histidine-rich RING finger motif. Other members of the family include TRAF1 and TRAF2. The latter proteins have been shown to regulate TNF-R2 as well as CD40 signaling through activation of the NFκB family of transcription factors.

REFERENCES

1. Kehry, M.R., et al. 1994. B cell activation by helper T cell membranes. *Crit. Rev. Immunol.* 14: 221-238.
2. Hu, H.M., et al. 1994. A novel RING finger protein interacts with the cytoplasmic domain of CD40. *J. Biol. Chem.* 269: 30069-30072.
3. Rothe, M., et al. 1994. A novel family of putative signal transducers associated with the cytoplasmic domain of the 75 kDa tumor necrosis factor receptor. *Cell* 78: 681-692.

CHROMOSOMAL LOCATION

Genetic locus: CD40 (human) mapping to 20q13.12.

SOURCE

CD40 (EA-5) is a mouse monoclonal antibody raised against L cells transfectants expressing CD40 of human origin.

PRODUCT

Each vial contains 200 µg IgG₁ kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

APPLICATIONS

CD40 (EA-5) is recommended for detection of CD40 of human origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and flow cytometry (1 µg per 1 x 10⁶ cells).

Suitable for use as control antibody for CD40 siRNA (h): sc-29250, CD40 shRNA Plasmid (h): sc-29250-SH and CD40 shRNA (h) Lentiviral Particles: sc-29250-V.

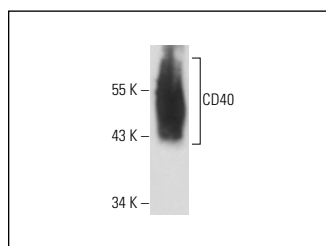
Molecular Weight of CD40: 43 kDa.

Positive Controls: NAMALWA cell lysate: sc-2234, BJAB whole cell lysate: sc-2207 or L428 cell lysate: sc-24728.

RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

DATA



CD40 (EA-5): sc-65264. Western blot analysis of CD40 expression in L428 whole cell lysate.

SELECT PRODUCT CITATIONS

1. Ahearne, M.J., et al. 2013. Enhancement of CD154/IL4 proliferation by the T follicular helper (Tfh) cytokine, IL21 and increased numbers of circulating cells resembling Tfh cells in chronic lymphocytic leukaemia. *Br. J. Haematol.* 162: 360-370.
2. Rosa, S., et al. 2019. A high-throughput screening method to identify compounds displaying human vascular embryonic toxicity. *Curr. Protoc. Stem Cell Biol.* 50: e93.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.



See **CD40 (H-10): sc-13128** for CD40 antibody conjugates, including AC, HRP, FITC, PE, and Alexa Fluor® 488, 546, 594, 647, 680 and 790.