

Product Specification Sheet

Human CD40 Antibody

<input type="checkbox"/> Cat# CD4011-A	Rabbit anti-Mouse CD40 IgG, affinity purified	SIZE: 100 ug
<input type="checkbox"/> Cat# CD4011-P	Control/Blocking peptide for CD40 antibody	SIZE: 250 ug
<input type="checkbox"/> Cat# CD4011-S	Rabbit anti-Mouse CD40 antiserum	SIZE: 100 ul

Cluster of differentiation 40, CD40 is a costimulatory protein found on antigen presenting cells and is required for their activation. The binding of CD154 (CD40L) on TH cells to CD40 activates antigen presenting cells and induces a variety of downstream effects. CD40 McAb recognizes a 48 KD type I single chain transmembrane glycoprotein expressed on normal and neoplastic B cells, but not on terminally differentiated plasma cells. CD40 antigen is also present on Hodgkin and Reed-sterberg cells, follicular dendritic cells, some macrophages, basal epithelial cells and endothelial cells. The ligand for CD40 is CD154 (CD40L). The function of CD40 is involved in growth, differentiation, and isotype-switching on B cells and also involved to rescue B cells from apoptosis.

Source of Antigen or Antibodies

Uniprot: P27512

Host: Rabbit

Form: Polyclonal, antibody was purified through ammonium sulfate followed by affinity purification.

Immunogen: 20 amino acid peptide derived from Mouse CD40 cytoplasmic region conjugated to KLH

Species Reactivity: Mouse

Cross reactivity: The immunogen of this antibody shows 100% homology in Human, non-human primate, horse, rat, cat, and dog CD40. 90% in Goat, Sheep, and Bovine. Species reactivity has not been confirmed for other species.

Subcellular Location: Plasma membrane

Alternative names: CD40

Tissue specificity: B cells and primary carcinomas.

Isoforms: CD40 has 5 isoforms. This antibody will not recognize Isoforms II, III, or IV.

Isoform II: Missing 204-289. Mass: 22.5 kDa

Isoform III: Missing 235-289. Mass: 25.7 kDa

Isoform IV: Missing 223-289. Mass: 24.5 kDa

Recommended Secondary Antibody: Goat anti-Rabbit IgG-HRP

Negative Control: Non immune Rabbit IgG (**ADI cat# 20009-1**). Blocking peptide (**ADI cat# CD4011-P**), use 5-10 ug of control peptide per 1 ug of affinity pure antibody or 1 ul antiserum to confirm specificity.

Form & Storage

Affinity pure IgG

_____ ul solution; Concentration: _____
Supplied in PBS+0.1% sodium azide

lyophilized powder

Reconstitute powder in 200 ul PBS to 0.5 mg/ml

Control/blocking peptide

100 ug/100 ul solution

lyophilized powder

Reconstitute powder in PBS at 1 mg/ml.

Storage

Short-term: 4°C for 1 month

Long-term: at -20°C or below in suitable aliquots after reconstitution for 1 year. Do not expose to multiple freeze/thaw cycles or store working, diluted solutions.

Stability: 6-12 months at -20°C or below.

Recommended Usage

ELISA: Assay dependent concentration. Typically 0.1-2 ug/ml for capture/detection antibodies.

Western Blotting: 1-3 ug/ml using affinity pure. 1:1k-1:5k for antiserum.

Theoretical band size: 32 kDa for the mature protein

Band is observed at ~44 kDa. This may be due to multiple glycosylation or other post-translational modifications.

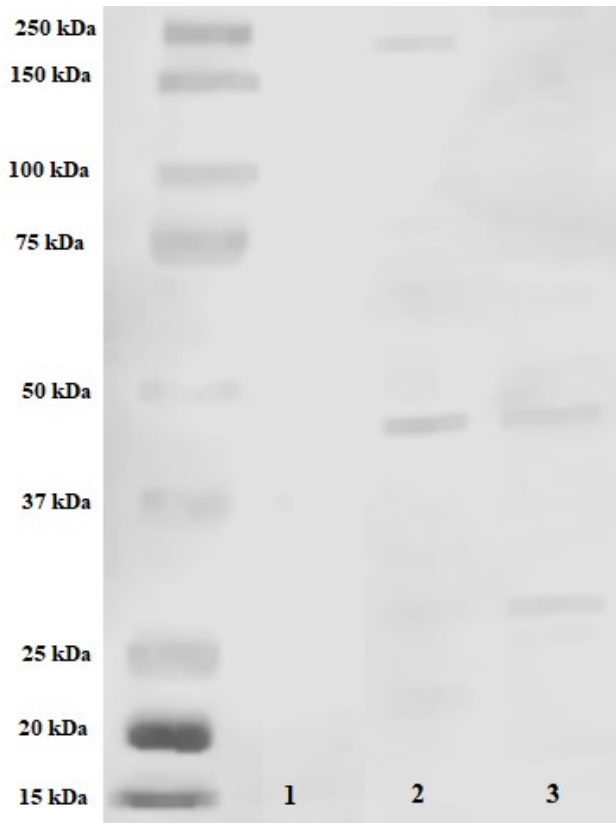
IHC: 1-4 ug/ml.

ICC: Not tested in-house. We suggest using similar concentrations as IHC. Permeabilization step with Tween-20 may be required.

*Above concentrations are a suggestion and user's must optimize assay based on their conditions. Antibody may work in other applications not listed. These methods have not been tested by ADI.

General References: Grewal IS et al (1998) Annu Rev Immunol 16: 111-35.

*This product is for In vitro research use only.



25 ug of HeLa (Lane#1), Jurkat (Lane#2) and Mouse Lung (Lane#3) were heated for 5 minutes at 95°C then loaded onto a 10% SDS page gel (Biorad). Gels were run for ~40 minutes at 200V and transferred to a 0.2 um nitrocellulose membrane using the mixed MW settings on a Transblot Turbo (Biorad). Ladders were annotated with ADI antigen-antibody pen and blocked for 1 hour at room temperature with 1% Fish plasma (Aquablock, EastCoastBio). CD4011-A was diluted with TBST+3% BSA to 1 ug/ml and incubated overnight at 4°C. Blot was washed with TBS-T 3 times for 5 minutes each. Goat anti-rabbit IgG HRP (ADI cat#20320) was added at 1:5,000 dilution for 1 hour at room temperature. Blot was washed 3 times with TBS-T for 5 minutes each. ADI Femto ECLsubstrate (ADI cat#80210) was added for 5 minutes and the blot was imaged on a LI-COR C-digit at high sensitivity settings. Specific band is observed at ~44 kDa