

CAD Antibody (Center)

Affinity Purified Rabbit Polyclonal Antibody (Pab) Catalog # AP11110c

Specification

CAD Antibody (Center) - Product Information

Application Primary Accession Other Accession Reactivity Predicted Host Clonality Isotype Calculated MW Antigen Region WB, IHC-P, FC,E <u>P27708</u> <u>B2ROC6</u>, <u>NP_004332.2</u> Human Mouse Rabbit Polyclonal Rabbit IgG 242984 780-809

CAD Antibody (Center) - Additional Information

Gene ID 790

Other Names CAD protein, Glutamine-dependent carbamoyl-phosphate synthase, Aspartate carbamoyltransferase, Dihydroorotase, CAD

Target/Specificity

This CAD antibody is generated from rabbits immunized with a KLH conjugated synthetic peptide between 780-809 amino acids from the Central region of human CAD.

Dilution WB~~1:2000 IHC-P~~1:25 FC~~1:25 E~~Use at an assay dependent concentration.

Format

Purified polyclonal antibody supplied in PBS with 0.09% (W/V) sodium azide. This antibody is purified through a protein A column, followed by peptide affinity purification.

Storage

Maintain refrigerated at 2-8°C for up to 2 weeks. For long term storage store at -20°C in small aliquots to prevent freeze-thaw cycles.

Precautions

CAD Antibody (Center) is for research use only and not for use in diagnostic or therapeutic procedures.

CAD Antibody (Center) - Protein Information



Name CAD (<u>HGNC:1424</u>)

Function Multifunctional protein that encodes the first 3 enzymatic activities of the de novo pyrimidine pathway: carbamoylphosphate synthetase (CPSase; EC 6.3.5.5), aspartate transcarbamylase (ATCase; EC 2.1.3.2) and dihydroorotase (DHOase; EC 3.5.2.3). The CPSase-function is accomplished in 2 steps, by a glutamine-dependent amidotransferase activity (GATase) that binds and cleaves glutamine to produce ammonia, followed by an ammonium-dependent carbamoyl phosphate synthetase, which reacts with the ammonia, hydrogencarbonate and ATP to form carbamoyl phosphate. The endogenously produced carbamoyl phosphate is sequestered and channeled to the ATCase active site. ATCase then catalyzes the formation of carbamoyl-L-aspartate from L-aspartate and carbamoyl phosphate. In the last step, DHOase catalyzes the cyclization of carbamoyl aspartate to dihydroorotate.

Cellular Location

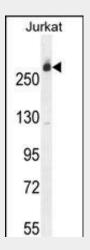
Cytoplasm. Nucleus. Note=Cytosolic and unphosphorylated in resting cells, translocates to the nucleus in response to EGF stimulation, nuclear import promotes optimal cell growth

CAD Antibody (Center) - Protocols

Provided below are standard protocols that you may find useful for product applications.

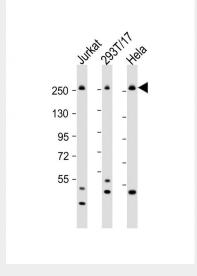
- <u>Western Blot</u>
- Blocking Peptides
- Dot Blot
- <u>Immunohistochemistry</u>
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

CAD Antibody (Center) - Images

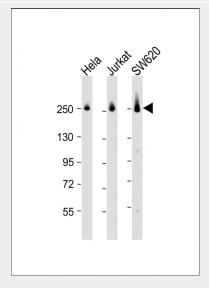


CAD Antibody (Center) (Cat. #AP11110c) western blot analysis in Jurkat cell line lysates (35ug/lane).This demonstrates the CAD antibody detected the CAD protein (arrow).



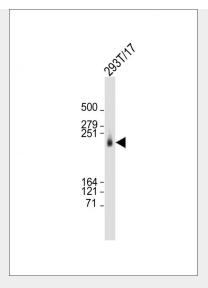


All lanes : Anti-CAD Antibody (Center) at 1:2000 dilution Lane 1: Jurkat whole cell lysate Lane 2: 293T/17 whole cell lysate Lane 3: Hela whole cell lysate Lysates/proteins at 20 μ g per lane. Secondary Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/10000 dilution. Predicted band size : 243 kDa Blocking/Dilution buffer: 5% NFDM/TBST.

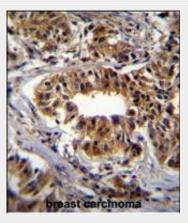


All lanes : Anti-CAD Antibody (Center) at 1:2000 dilution Lane 1: Hela whole cell lysate Lane 2: Jurkat whole cell lysate Lane 3: SW620 whole cell lysate Lysates/proteins at 20 μ g per lane. Secondary Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/10000 dilution. Predicted band size : 243 kDa Blocking/Dilution buffer: 5% NFDM/TBST.

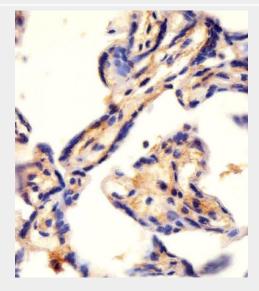




Anti-CAD Antibody (Center) at 1:2000 dilution + 293T/17 whole cell lysate Lysates/proteins at 20 μ g per lane. Secondary Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/10000 dilution. Predicted band size : 243 kDa Blocking/Dilution buffer: 5% NFDM/TBST.

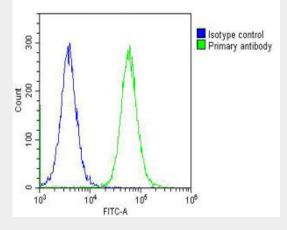


CAD Antibody (Center) (Cat. #AP11110c)immunohistochemistry analysis in formalin fixed and paraffin embedded human breast carcinoma followed by peroxidase conjugation of the secondary antibody and DAB staining. This data demonstrates the use of CAD Antibody (Center) for immunohistochemistry. Clinical relevance has not been evaluated.





AP11110c staining CAD in human placenta tissue sections by Immunohistochemistry (IHC-P - paraformaldehyde-fixed, paraffin-embedded sections). Tissue was fixed with formaldehyde and blocked with 3% BSA for 0. 5 hour at room temperature; antigen retrieval was by heat mediation with a citrate buffer (pH6). Samples were incubated with primary antibody (1/25) for 1 hours at 37°C. A undiluted biotinylated goat polyvalent antibody was used as the secondary antibody.



Overlay histogram showing Hela cells stained with AP11110c (green line). The cells were fixed with 2% paraformaldehyde (10 min) and then permeabilized with 90% methanol for 10 min. The cells were then icubated in 2% bovine serum albumin to block non-specific protein-protein interactions followed by the antibody (AP11110c, 1:25 dilution) for 60 min at 37°C. The secondary Goat-Anti-Rabbit antibody used was lgG, **DyLight**® 488 Conjugated Highly Cross-Adsorbed(OH191631) at 1/200 dilution for 40 min at 37°C. Isotype control antibody (blue line) was rabbit IgG $(1\mu g/1 \times 10^{6} \text{ cells})$ used under the same conditions. Acquisition of >10, 000 events was performed.

CAD Antibody (Center) - Background

The de novo synthesis of pyrimidine nucleotides is required for mammalian cells to proliferate. This gene encodes a trifunctional protein which is associated with the enzymatic activities of the first 3 enzymes in the 6-step pathway of pyrimidine biosynthesis: carbamoylphosphate synthetase (CPS II), aspartate transcarbamoylase, and dihydroorotase. This protein is regulated by the mitogen-activated protein kinase (MAPK) cascade, which indicates a direct link between activation of the MAPK cascade and de novo biosynthesis of pyrimidine nucleotides.

CAD Antibody (Center) - References

Jia, P., et al. Schizophr. Res. 122 (1-3), 38-42 (2010) : Rose, J.E., et al. Mol. Med. 16 (7-8), 247-253 (2010) : Ahuja, V., et al. J. Inherit. Metab. Dis. 31(4):481-491(2008) Sugiyama, N., et al. Mol. Cell Proteomics 6(6):1103-1109(2007) Olsen, J.V., et al. Cell 127(3):635-648(2006) **CAD Antibody (Center) - Citations** • Oncogenic HSP90 Facilitates Metabolic Alterations in Aggressive B-cell Lymphomas