

SCAP Antibody (Center)

Affinity Purified Rabbit Polyclonal Antibody (Pab) Catalog # AP8568c

Specification

SCAP Antibody (Center) - Product Information

Application WB, IF, IHC-P, FC,E

Primary Accession <u>Q12770</u>

Other Accession <u>A2RRU4, Q5MNU5, Q6GOT6, A6QM06</u>

Reactivity Human, Mouse, Rat

Predicted Bovine, Pig
Host Rabbit
Clonality Polyclonal
Isotype Rabbit IgG
Antigen Region 604-632

SCAP Antibody (Center) - Additional Information

Gene ID 22937

Other Names

Sterol regulatory element-binding protein cleavage-activating protein, SCAP, SREBP cleavage-activating protein, SCAP, KIAA0199

Target/Specificity

This SCAP antibody is generated from rabbits immunized with a KLH conjugated synthetic peptide between 604-632 amino acids from the Central region of human SCAP.

Dilution

WB~~1:2000 IF~~1:100 IHC-P~~1:50~100 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

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

SCAP Antibody (Center) - Protein Information



Name SCAP {ECO:0000303|PubMed:10570913, ECO:0000312|HGNC:HGNC:30634}

Function Escort protein required for cholesterol as well as lipid homeostasis (By similarity). Regulates export of the SCAP-SREBP complex from the endoplasmic reticulum to the Golgi upon low cholesterol, thereby regulating the processing of sterol regulatory element-binding proteins (SREBPs) SREBF1/SREBP1 and SREBF2/SREBP2 (PubMed:26311497). At high sterol concentrations, formation of a ternary complex with INSIG (INSIG1 or INSIG2) leads to mask the ER export signal in SCAP, promoting retention of the complex in the endoplasmic reticulum (By similarity). Low sterol concentrations trigger release of INSIG, a conformational change in the SSD domain of SCAP, unmasking of the ER export signal, promoting recruitment into COPII-coated vesicles and transport of the SCAP-SREBP to the Golgi: in the Golgi, SREBPs are then processed, releasing the transcription factor fragment of SREBPs from the membrane, its import into the nucleus and up-regulation of LDLR, INSIG1 and the mevalonate pathway (PubMed:26311497). Binds cholesterol via its SSD domain (By similarity).

Cellular Location

Endoplasmic reticulum membrane; Multi-pass membrane protein. Golgi apparatus membrane; Multi-pass membrane protein. Cytoplasmic vesicle, COPII-coated vesicle membrane {ECO:0000250|UniProtKB:P97260}; Multi-pass membrane protein. Note=Moves from the endoplasmic reticulum to the Golgi in the absence of sterols (PubMed:26311497). Requires the presence of SPRING1 for proper localization to endoplasmic reticulum (PubMed:32111832). {ECO:0000250|UniProtKB:P97260, ECO:0000269|PubMed:26311497, ECO:0000269|PubMed:32111832}

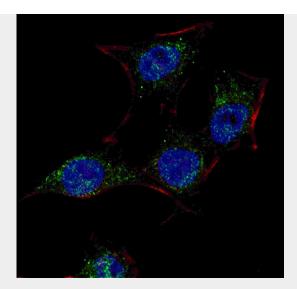
SCAP Antibody (Center) - Protocols

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

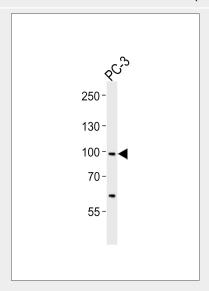
- Western Blot
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture

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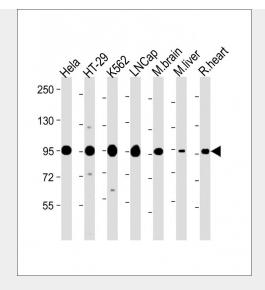


Fluorescent confocal image of HeLa cells stained with SCAP (Center) antibody. HeLa cells were fixed with 4% PFA (20 min), permeabilized with Triton X-100 (0.2%, 30 min). Cells were then incubated with AP8568c SCAP (Center) primary antibody (1:100, 2 h at room temperature). For secondary antibody, Alexa Fluor® 488 conjugated donkey anti-rabbit antibody (green) was used (1:1000, 1h). Nuclei were counterstained with Hoechst 33342 (blue) (10 µg/ml, 5 min).

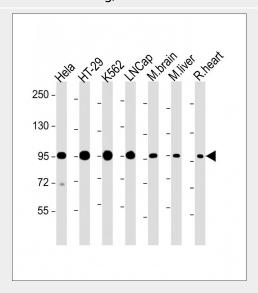


Western blot analysis of lysate from PC-3 cell line, using SCAP Antibody (Center)(Cat. #AP8568c). AP8568c was diluted at 1:1000. A goat anti-rabbit IgG H&L(HRP) at 1:10000 dilution was used as the secondary antibody. Lysate at 35ug.



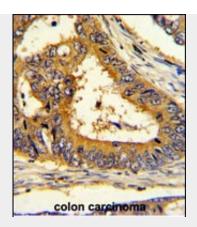


All lanes: Anti-SCAP Antibody (Center) at 1:2000 dilution Lane 1: Hela whole cell lysate Lane 2: HT-29 whole cell lysate Lane 3: K562 whole cell lysate Lane 4: LNCap whole cell lysate Lane 5: Mouse brain lysate Lane 6: Mouse liver lysate Lane 7: Rat heart lysate Lysates/proteins at 20 µg per lane. Secondary Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/10000 dilution. Predicted band size: 140, 98, 96 kDa Blocking/Dilution buffer: 5% NFDM/TBST.

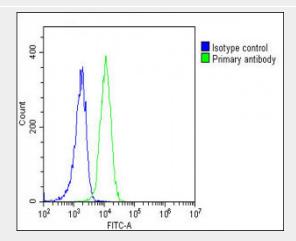


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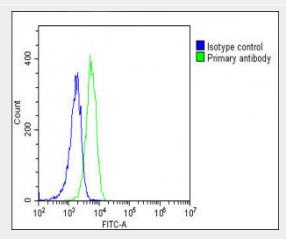




Formalin-fixed and paraffin-embedded human colon carcinoma with SCAP Antibody (Center), which was peroxidase-conjugated to the secondary antibody, followed by DAB staining. This data demonstrates the use of this antibody for immunohistochemistry; clinical relevance has not been evaluated.



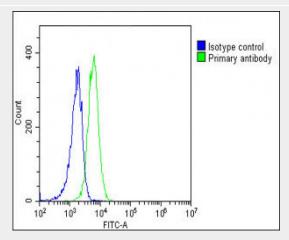
Overlay histogram showing K562 cells stained with AP8568c(green line). The cells were fixed with 2% paraformaldehyde (10 min) and then permeabilized with 90% methanol for 10 min. The cells were then icubated in 2% bovine serum albumin to block non-specific protein-protein interactions followed by the antibody (AP8568c, 1:25 dilution) for 60 min at 37 $^{\circ}$ C. The secondary antibody used was Goat-Anti-Rabbit IgG, DyLight® 488 Conjugated Highly Cross-Adsorbed(OH191631) at 1/200 dilution for 40 min at 37 $^{\circ}$ C. Isotype control antibody (blue line) was rabbit IgG1 (1 μ g/1x10 $^{\circ}$ 6 cells) used under the same conditions. Acquisition of >10, 000 events was performed.



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SCAP Antibody (Center) - Background

SCAP is a protein with a sterol sensing domain(SSD) and seven WD domains. In the presence of cholesterol, this protein binds to sterol regulatory element binding proteins (SREBPs) and mediates their transport from the ER to the Golgi. The SREBPs are then proteolytically cleaved and regulate sterol biosynthesis.

SCAP Antibody (Center) - References

Lu,Y., et.al., J. Lipid Res. 49 (12), 2582-2589 (2008)

SCAP Antibody (Center) - Citations

• <u>COPI-mediated retrieval of SCAP is crucial for regulating lipogenesis under basal and</u> sterol-deficient conditions.