abcam

Product datasheet

Anti-S6K1 antibody [E175] ab32359

敲除验证 重组 RabMAb

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概述

产品名称	Anti-S6K1抗体[E175]
描述	兔单克隆抗体[E175] to S6K1
宿主	Rabbit
特异性	This antibody may detect both phosphorylated and non-phosphorylated forms of p70 S6 Kinase.
经测试应用	**适用于:** WB, IHC-P **不适用于:** Flow Cyt or ICC/IF
种属反应性	**与反应:** Mouse, Human **预测可用于:** Rat, Cow
免疫原	Synthetic peptide within Human S6K1. The exact sequence is proprietary.
阳性对照	Jurkat cell lysate; human breast carcinoma WB: C-Myc/DDK tagged human S6K1 recombinant protein, C-Myc/DDK tagged human S6K2 recombinant protein.
常规说明	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

性能

形式	Liquid
存放说明	Shipped at 4°C. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.
存储溶液	pH: 7.20 Preservative: 0.05% Sodium azide Constituents: 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant
纯度	Protein A purified

克隆	单克隆
克隆编号	E175
同种型	IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab32359于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB	★★★★☆ (1)	1/1000 - 1/5000. Detects a band of approximately 70 kDa (predicted molecular weight: 59 kDa).
IHC-P	★★★☆☆ (2)	1/50. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

应用说明 Is unsuitable for Flow Cyt or ICC/IF.

靶标

功能 Acts to integrate nutrient and growth factor signals in regulation of protein synthesis, cell proliferation, cell growth, cell cycle progression and cell survival. Downstream effector of the mTOR signaling pathway. Phosphorylates specifically ribosomal protein S6 in response to insulin or several classes of mitogens. During translation initiation, the inactive form associatess with the eIF-3 complex under conditions of nutrient depletion. Mitogenic stimulation leads to phosphorylation and dissociation from the eIF-3 complex and the free activated form can phosphorylate other translational targets including EIF4B. Promotes protein synthesis by phosphorylating PDCD4 at 'Ser-67' and targeting it for degradation. Phosphorylates RICTOR leading to regulation of mammalian target of rapamycin complex 2 (mTORC2) signaling; probably phosphorylates RICTOR at 'Thr-1135'. Phosphorylates IRS1 at multiple serine residues coupled with insulin resistance; probably phosphorylates IRS1 at 'Ser-270'. Required for TNF-alpha induced IRS-1 degradation. Phosphorylates EEF2K in response to IGF1 and inhibits EEF2K activity. Phosphorylates BAD at 'Ser-99' in response to IGF1 leading to BAD inactivation and inhibition of BAD-induced apoptosis. Phosphorylates mitochondrial RMP leading to dissociation of a RMP:PPP1CC complex; probably phosphorylates RMP at 'Ser-99'. The free mitochondrial PPP1CC can dephosphorylate RPS6KB1 at Thr-412 which is proposed to be a negative feed back mechanism for the RPS6KB1 antiapoptotic function. Phosphorylates GSK3B at 'Ser-9' under conditions leading to loss of the TSC1-TSC2 complex. Phosphorylates POLDIP3.

组织特异性 Widely expressed.

序列相似性 Belongs to the protein kinase superfamily. AGC Ser/Thr protein kinase family. S6 kinase subfamily.
Contains 1 AGC-kinase C-terminal domain.
Contains 1 protein kinase domain.

结构域 The autoinhibitory domain is believed to block phosphorylation within the AGC-kinase C-terminal domain and the activation loop.
The TOS (TOR signaling) motif is essential for activation by mTORC1.

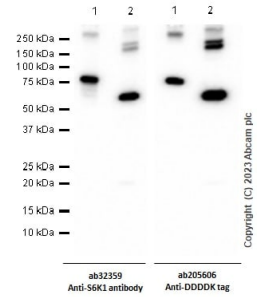
翻译后修饰 Phosphorylation at Thr-412 is regulated by mTORC1. The phosphorylation at this site is

maintained by an agonist-dependent autophosphorylation mechanism.

细胞定位

Cytoplasm; Nucleus. Cytoplasm and Cell junction > synapse > synaptosome. Mitochondrion outer membrane.

图片



Western blot - Anti-S6K1 antibody [E175] (ab32359)

All lanes : Anti-S6K1 antibody [E175] (ab32359) at 1/1000 dilution

Lane 1 : C-Myc/DDK tagged human S6K1 recombinant protein 10ng

Lane 2 : C-Myc/DDK tagged human S6K2 recombinant protein 10ng

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 59 kDa

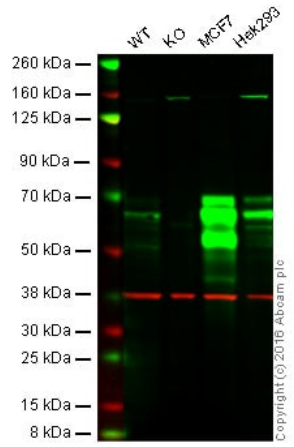
Observed band size: 60,75 kDa

Exposure time: 10 seconds

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

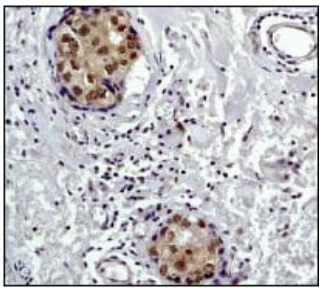
ab205606 was also used at 1/1000 dilution to detect C-Myc/DDK tagged human S6K1 and S6K2 recombinant proteins shown on the right panel.

ab32539 showed cross reactivity with S6K2 in recombinant protein testing. S6K1 and S6K2 have different observed band size in endogenous and recombinant protein tests.



Western blot - Anti-S6K1 antibody [E175] (ab32359)

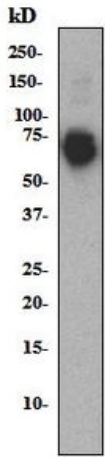
Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: S6K1 knockout HAP1 cell lysate (20 µg)
Lane 3: MCF7 cell lysate (20 µg)
Lane 4: HEK293 cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab32359 observed at 68 kDa. Red - loading control, **ab8245**, observed at 37 kDa.
ab32359 was shown to recognize S6K1 when S6K1 knockout samples were used, along with additional cross-reactive bands. Wild-type and S6K1 knockout samples were subjected to SDS-PAGE. ab32359 and **ab8245** (loading control to GAPDH) were diluted 1/1000 and 1/10000 respectively and incubated overnight at 4°C. Blots were developed with goat anti-rabbit IgG (H + L) and goat anti-mouse IgG (H + L) secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-S6K1 antibody [E175] (ab32359)

Paraffin-embedded human breast carcinoma
ab32359 at 1/50 dilution

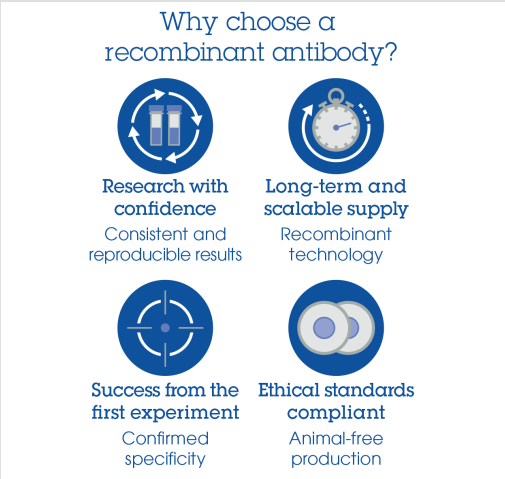
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-S6K1 antibody [E175] (ab32359)

Anti-S6K1 antibody [E175] (ab32359) + HEK-293T (Human epithelial cell line from embryonic kidney transformed with large T antigen) whole cell lysate

Predicted band size: 59 kDa



Anti-S6K1 antibody [E175] (ab32359)

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