abcam

Product datasheet

Anti-S6K1 antibody [E343] - BSA and Azide free ab203558

敲除验证 重组 RabMAb

14 图像

概述

产品名称	Anti-S6K1抗体[E343] - BSA and Azide free
描述	兔单克隆抗体[E343] to S6K1 - BSA and Azide free
宿主	Rabbit
特异性	This antibody detects both alpha I and alpha II isoforms.
经测试应用	**适用于:** Flow Cyt (Intra), WB, IP, IHC-P, ICC/IF
种属反应性	**与反应:** Mouse, Rat, Human
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
常规说明	ab203558 is the carrier-free version of **ab32529**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. Use our **conjugation kits** for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc. 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. Store at +4°C. Do Not Freeze.
存储溶液	pH: 7.2 Constituent: PBS
无载体	**是**
纯度	Protein A purified
克隆	单克隆
克隆编号	E343
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab203558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
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab199376** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
WB		Use at an assay dependent concentration. Detects a band of approximately 70 kDa (predicted molecular weight: 59 kDa). For Rat and Mouse samples 1/500 dilution has only been tried. We have not tested if similarly to Human samples a lot higher dilutions can be used.
IP		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		Use at an assay dependent concentration.

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.

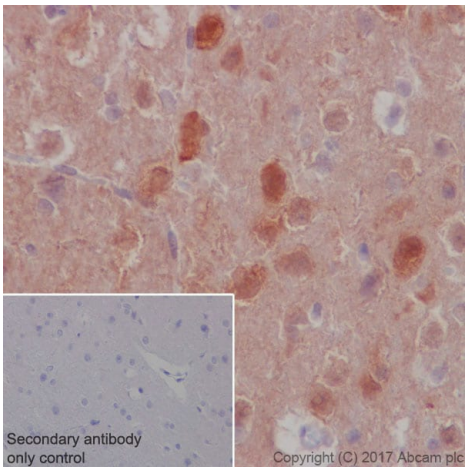
序列相似性 Belongs to the protein kinase superfamily. AGC Ser/Thr protein kinase family. S6 kinase subfamily.
Contains 1 AGC-kinase C-terminal domain.
Contains 1 protein kinase domain.

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.

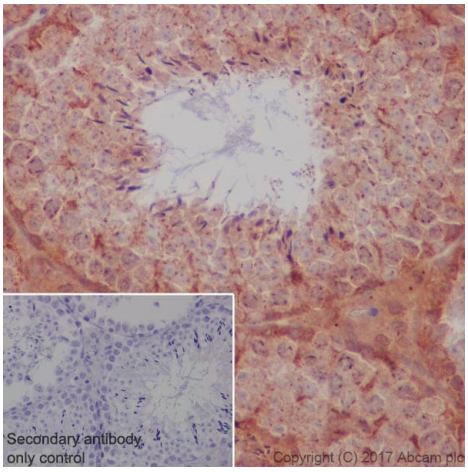
图片



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Immunohistochemical analysis of rat brain tissue labeling S6K1 with **ab32529** at 1/500 dilution (4.4 µg/mL). The secondary antibody used was ImmunoHistoProbe one step HRP Polymer (ready to use). Secondary antibody only control-PBS instead of the primary antibody. Antigen retrieval was heat mediated using **ab93684** (Tris/EDTA buffer, pH 9.0). The tissue was counterstained with Hematoxylin.

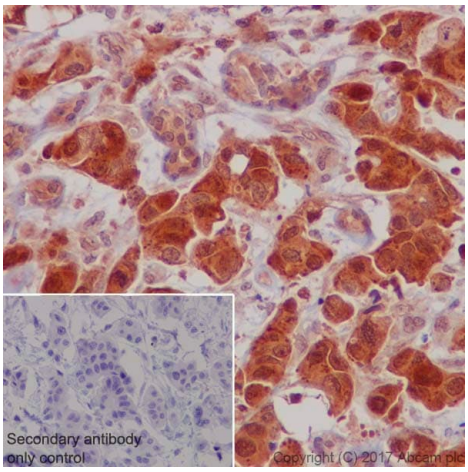
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Immunohistochemical analysis of mouse testis tissue labeling S6K1 with **ab32529** at 1/500 dilution (4.4 µg/mL). The secondary antibody used was ImmunoHistoProbe one step HRP Polymer (ready to use). Secondary antibody only control-PBS instead of the primary antibody. Antigen retrieval was heat mediated using **ab93684** (Tris/EDTA buffer, pH 9.0). The tissue was counterstained with Hematoxylin.

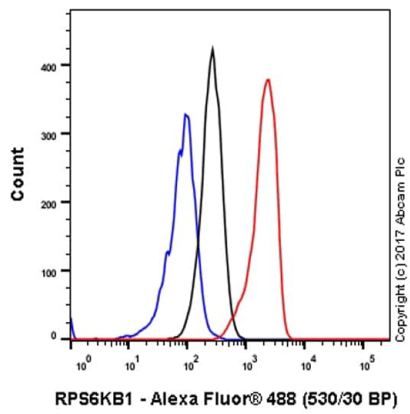
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Immunohistochemical analysis of Human breast cancer tissue labeling S6K1 with **ab32529** at 1/500 dilution (4.4 µg/mL). The secondary antibody used was ImmunoHistoProbe one step HRP Polymer (ready to use). Secondary antibody only control-PBS instead of the primary antibody. Antigen retrieval was heat mediated using **ab93684** (Tris/EDTA buffer, pH 9.0). The tissue was counterstained with Hematoxylin.

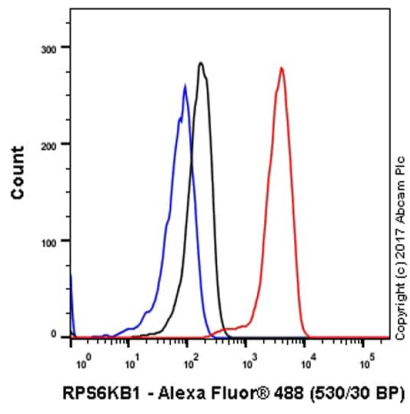
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Flow Cytometry (Intracellular) - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Intracellular Flow Cytometry analysis of C6 (Rat glial tumor glial cell) cells labelling with **ab32529** (purified) at 1/2200 dilution (1 µg/mL) (red). Cells were fixed with 4% paraformaldehyde . Goat anti rabbit IgG (Alexa Fluorr® 488, **ab150077**) was used as the secondary antibody at 1/2000 dilution. Isotype control - 90% methanol . Unlabeled control - Rabbit monoclonal IgG (**ab172730**) / Black.

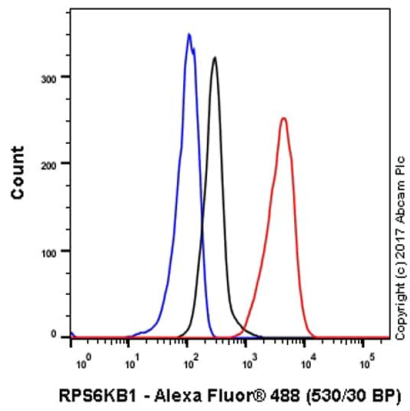
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Flow Cytometry (Intracellular) - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Intracellular Flow Cytometry analysis of 293T (Human embryonic kidney epithelial cell) cells labelling with **ab32529** (purified) at 1/2200 dilution (1 µg/mL) (red). Cells were fixed with 4% paraformaldehyde . Goat anti rabbit IgG (Alexa Fluorr® 488, **ab150077**) was used as the secondary antibody at 1/2000 dilution. Isotype control - 90% methanol . Unlabeled control - Rabbit monoclonal IgG (**ab172730**) / Black.

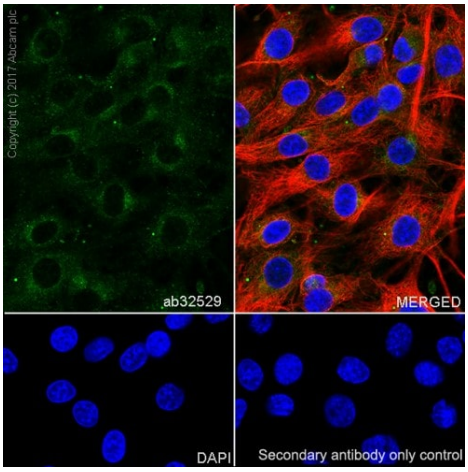
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Flow Cytometry (Intracellular) - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Intracellular Flow Cytometry analysis of Neuro-2a (Mouse neuroblastoma neuroblast) cells labelling with **ab32529** (purified) at 1/2200 dilution (1 µg/mL) (red). Cells were fixed with 4% paraformaldehyde . Goat anti rabbit IgG (Alexa Fluorr® 488, **ab150077**) was used as the secondary antibody at 1/2000 dilution. Isotype control - 90% methanol . Unlabeled control - Rabbit monoclonal IgG (**ab172730**) / Black.

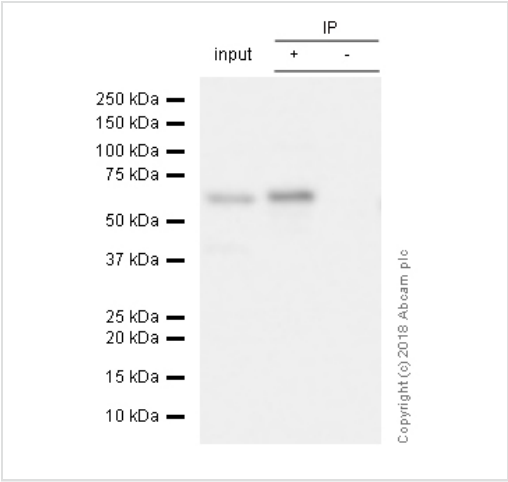
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Immunocytochemistry/ Immunofluorescence - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Immunocytochemistry/Immunofluorescence analysis of C6 cells (Rat glial tumor glial cell) labelling S6K1 with **ab32529** at a dilution of 1:200, 11.1 µg/ml. Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100. A 1:1000 dilution (2µg/ml) was used for the secondary antibody Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**). The cells were co-stained with 1:200, 2.5µg/ml with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594). Nuclei counterstained with DAPI (blue). Control: 1:1000 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



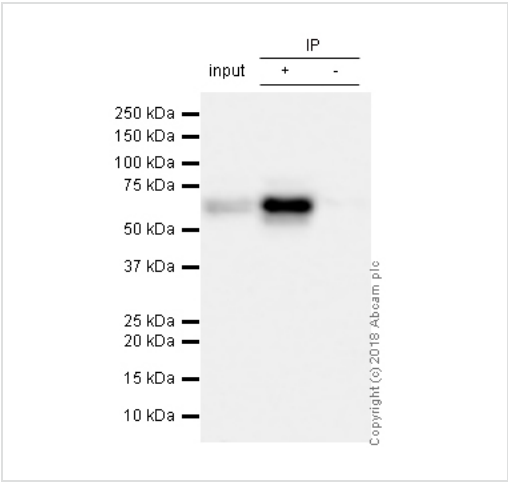
Immunoprecipitation - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Lane 1: Neuro2a (Mouse neuroblastoma neuroblast) whole cell lysate, 10µg
Lane 2: Neuro2a whole cell lysate 350µg and **ab32529**, 2µg
Lane 3: Neuro2a cell lysate, 350µg and rabbit IgG (**ab172730**), 2µg

Purified **ab32529** immunoprecipitating S6K1 in HEK293T cell lysates. Primary antibody was used at a 1:500 dilution (4.4 µg/ml). For western blotting, VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/1000 dilution.

Blocking and diluting buffer used: 5% NFDM/TBST.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Immunoprecipitation - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Lane 1: HEK293T (Human embryonic kidney epithelial cell) whole cell lysate, 10µg
Lane 2: HEK293T whole cell lysate, 10µg and **ab32529**, 2µg
Lane 3: HEK293T cell lysate, 350µg and rabbit IgG (**ab172730**) , 2µg

Purified **ab32529** immunoprecipitating S6K1 in HEK293T cell lysates. Primary antibody was used at a 1:500 dilution (4.4 µg/ml). For western blotting, VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/1000 dilution.

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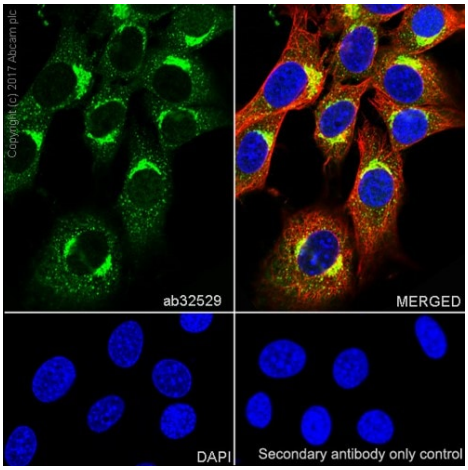
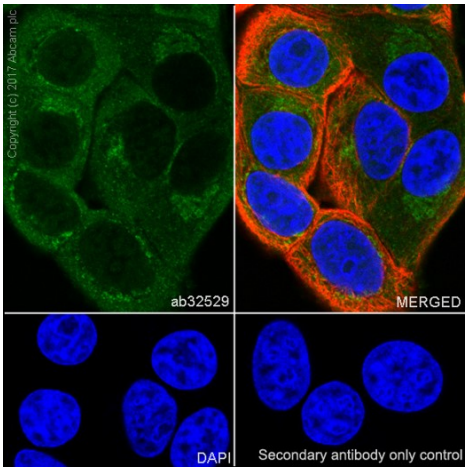


Immunocytochemistry/ Immunofluorescence - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Immunocytochemistry/Immunofluorescence analysis of NIH/3T3 (Mouse embryonic fibroblast) labelling with **ab32529** at a dilution of 1:200, 11.1 µg/ml. Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100. A 1:1000 dilution (2µg/ml) was used for the secondary antibody Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**). The cells were co-stained at 1:200 dilution, 2.5µg/ml with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594). Nuclei counterstained with DAPI (blue). Control: 1:1000 dilution.

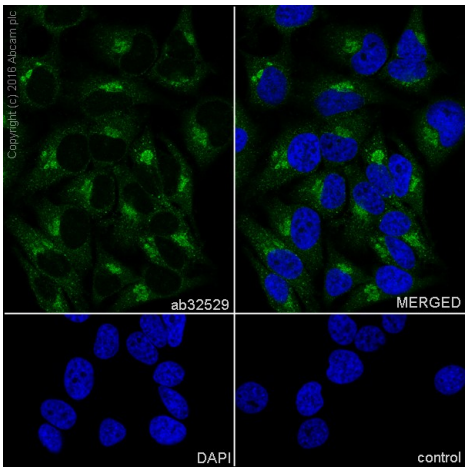
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Immunocytochemistry/ Immunofluorescence - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Immunocytochemistry/Immunofluorescence analysis of MCF 7 (Human breast adenocarcinoma epithelial cell) labeling S6K1 with **ab32529** at a dilution of 1:200, 11.1 ug/ml. Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100. A dilution of 1/1000 (2µg/ml) was used for the secondary antibodyGoat anti rabbit IgG (Alexa Fluor® 488, **ab150077**). The cells were co-stained at 1:200 dilution, 2.5µg/ml with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) . Nuclei counterstained with DAPI (blue). Control: 1:1000 dilution.

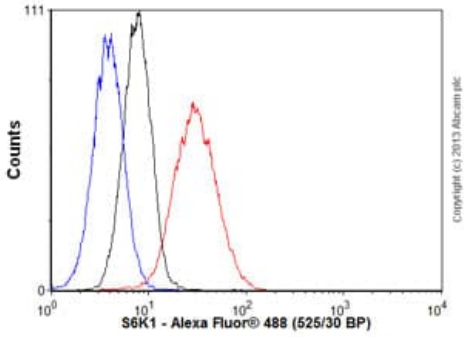
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Immunocytochemistry/ Immunofluorescence - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

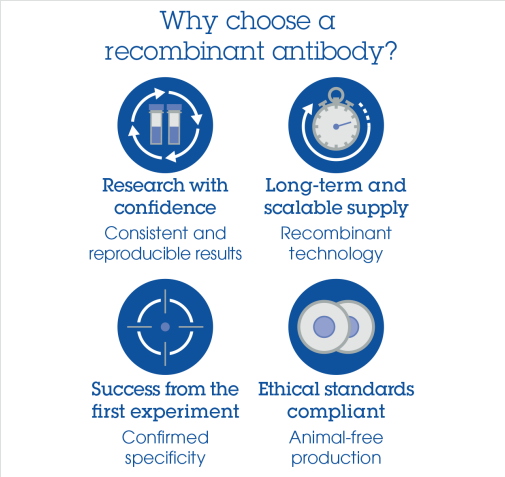
Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human epithelial cell line from cervix adenocarcinoma) labeling S6K1 with **ab32529** at a dilution of 1/500. Cells were fixed with 4% Paraformaldehyde and permeabilised with 0.1% tritonX-100. **ab150077** at 1/1000 was used as the secondary antibody. Nuclei were counterstained with DAPI (blue).

Confocal image showing cytoplamic staining on HeLa cell line. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Flow Cytometry (Intracellular) - Anti-S6K1 antibody [E343] - BSA and Azide free (ab203558)

Overlay histogram showing HeLa cells stained with **ab32529** (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (**ab32529**, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) (**ab150077**) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (0.1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab32529**).



Anti-S6K1 antibody [E343] - BSA and Azide free

(ab203558)

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