abcam

ab189380

Anti-CRISPR-Cas9 antibody [EPR18991]

Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380) is a rabbit monoclonal antibody detecting CRISPR-Cas9 in **Western Blot, Flow Cytometry (Intra), IHC-P, ICC/IF**.

- Biophysical QC for unrivalled batch-batch consistency

Recombinant | RabMAb | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR18991
Purification technique	Affinity purification Protein A
Concentration	0.55 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ICC

Tested

Species	Transfected cell line - Streptococcus pyogenes serotype M1
Dilution info	1/100

Notes	-

Predicted

Species	Streptococcus pyogenes, Streptococcus pyogenes serotype M1
Dilution info	-
Notes	-

WB

Tested

Species	Transfected cell line - Streptococcus pyogenes serotype M1
Dilution info	1/1000
Notes	ab189380 detects Cas9 and dCas9. This product has not been tested against SaCas9. Please contact Abcam Scientific Support for more information.

Predicted

Species	Streptococcus pyogenes, Streptococcus pyogenes serotype M1
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Transfected cell line - Streptococcus pyogenes serotype M1
Dilution info	1/500
Notes	-

Predicted

Species	Streptococcus pyogenes, Streptococcus pyogenes serotype M1
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Transfected cell line - Streptococcus pyogenes serotype M1
Dilution info	1/70
Notes	-

Predicted

Species	Streptococcus pyogenes, Streptococcus pyogenes serotype M1
Dilution info	-
Notes	-

IHC-P

Expected

Species	Transfected cell line - Streptococcus pyogenes serotype M1
Dilution info	1/100
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Predicted

Species	Streptococcus pyogenes, Streptococcus pyogenes serotype M1
Dilution info	-
Notes	-

Target data

See full target information cas9

Function	CRISPR (clustered regularly interspaced short palindromic repeat) is an adaptive immune system that provides protection against mobile genetic elements (viruses, transposable elements and conjugative plasmids) (PubMed:21455174). CRISPR clusters contain spacers, sequences complementary to antecedent mobile elements, and target invading nucleic acids. CRISPR clusters are transcribed and processed into CRISPR RNA (crRNA). In type II CRISPR systems correct processing

of pre-crRNA requires a trans-encoded small RNA (tracrRNA), endogenous ribonuclease 3 (rnc) and this protein. The tracrRNA serves as a guide for ribonuclease 3-aided processing of pre-crRNA; Cas9 only stabilizes the pre-crRNA:tracrRNA interaction and has no catalytic function in RNA processing (PubMed:24270795). Subsequently Cas9/crRNA/tracrRNA endonucleolytically cleaves linear or circular dsDNA target complementary to the spacer; Cas9 is inactive in the absence of the 2 guide RNAs (gRNA). The target strand not complementary to crRNA is first cut endonucleolytically, then trimmed 3'-5' exonucleolytically. DNA-binding requires protein and both gRNAs, as does nuclease activity. Cas9 recognizes the protospacer adjacent motif (PAM) in the CRISPR repeat sequences to help distinguish self versus nonself, as targets within the bacterial CRISPR locus do not have PAMs. DNA strand separation and heteroduplex formation starts at PAM sites; PAM recognition is required for catalytic activity (PubMed:24476820). Confers immunity against a plasmid with homology to the appropriate CRISPR spacer sequences (CRISPR interference) (PubMed:21455174).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

What is this antibody validated in?
Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF).

What is the molecular weight of CRISPR-Cas9?
Anti-CRISPR-Cas9 [EPR18991] (ab189380) specifically detects a band for CRISPR-Cas9 (UniProt: Q99ZW2) at a molecular weight of 158kDa.

Trusted by the scientific community
Anti-CRISPR-Cas9 [EPR18991] (ab189380) was first used in a scientific publication in 2016 and has been cited over 10 times in peer-reviewed journals.

Other related products
We have a range of other formats of antibody clone [EPR18991] also available for your convenience: ab189380, Alexa Fluor® 488 - ab215239, Alexa Fluor® 647 - ab215240, HRP - ab215345, Carrier free - ab232379

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	CRISPR-Cas9 also known simply as Cas9 is a protein that acts as a molecular scissor in gene editing. It has a molecular weight of approximately 160 kDa. Cas9 is a part of the CRISPR-Cas system originally discovered in bacteria where it serves as an adaptive immune system. In bacterial cells Cas9 targets and cleaves specific DNA sequences allowing for targeted gene modifications. While the expression of the CRISPR-Cas system occurs naturally in prokaryotes scientists now harness it in various organisms for genetic manipulation.
Biological function summary	The Cas9 protein functions as an integral part of the CRISPR-Cas9 complex which includes a guide RNA to direct the protein to specific DNA sequences. This complex enables precise cuts at targeted locations within the genome. The Cas9 protein size allows it to fit effectively within cells facilitating genome editing in areas such as research agriculture and therapeutics. The complexity of CRISPR-Cas9 also includes the interaction with other cellular components that assist in DNA repair post-cleavage.
Pathways	CRISPR-Cas9 plays a role in DNA repair pathways particularly non-homologous end joining and homologous recombination. After Cas9-induced DNA breaks these pathways become active to repair the cleaved DNA. The gene editing process facilitated by CRISPR-Cas9 often involves interaction with DNA repair proteins like ku70/80 and Rad51 in response to induced breaks. This allows for either the incorporation of new genetic material or the modification of existing genes.
Associated diseases and disorders	CRISPR-Cas9 has potential in treating genetic disorders such as cystic fibrosis and Duchenne muscular dystrophy. By correcting mutated genes scientists aim to restore normal gene function offering potential therapeutic benefits. Additionally

CRISPR-Cas9 relates indirectly to p53 a protein known for its tumor suppressor functions because gene-editing processes can sometimes activate p53-dependent pathways leading to cell cycle arrest or apoptosis. This highlights the importance of understanding the broader implications of CRISPR-Cas9 in therapeutic applications.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

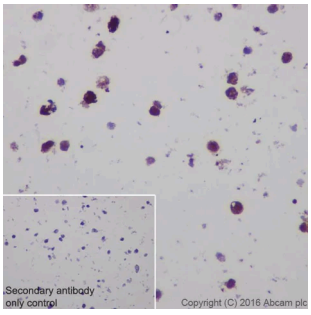
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

10 product images



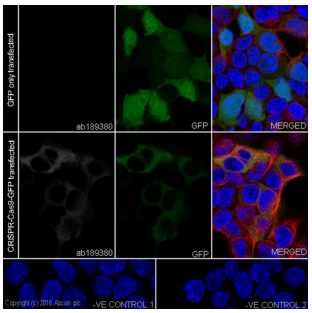
Immunocytochemistry - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380)

CRISPR-Cas9 Immunocytochemistry staining using rabbit Anti-CRISPR-Cas9 antibody

Immunocytochemical analysis of agarose-embedded sections of 293T (Human epithelial cell line from embryonic kidney) cells transfected with *Streptococcus pyogenes serotype M1* Cas9 (pcDNA3.1(+)-GFP-Myc) labeling CRISPR-Cas9 with ab189380 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.
Positive staining on 293T cells transfected with *Streptococcus pyogenes serotype M1* Cas9 (pcDNA3.1(+)-GFP-Myc) is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380)

CRISPR-Cas9 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-CRISPR-Cas9 antibody

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized 293T (Human epithelial cell line from embryonic kidney) cells transfected with CRISPR-Cas9-GFP or GFP only, labeling CRISPR-Cas9 with ab189380 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 647) (ab150079) secondary antibody at 1/1000 dilution.
Confocal image showing positive staining on 293T cells transfected with CRISPR-Cas9 with GFP-tag.

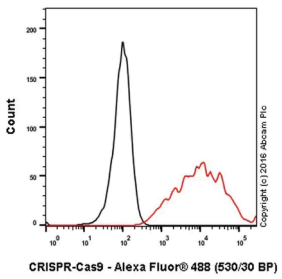
The nuclear counterstain is DAPI (blue).

Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (Alexa Fluor® 594 Goat anti-Mouse secondary) at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab189380 at 1/500 dilution followed by ab150120 (Alexa Fluor® 594 Goat anti-Mouse secondary) at 1/1000 dilution.

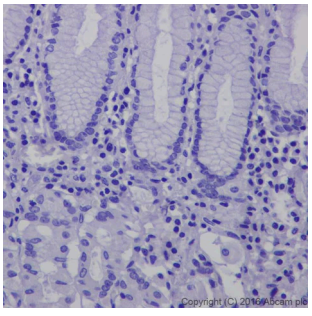
-ve control 2: ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution followed by ab150079 (Alexa Fluor® 647 Goat Anti-Rabbit IgG H&L) at 1/1000 dilution.



Flow Cytometry (Intracellular) - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380)

CRISPR-Cas9 Flow Cytometry (Intracellular) staining using rabbit Anti-CRISPR-Cas9 antibody

Intracellular Flow Cytometry analysis of HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate (transfected with CRISPR-Cas9 (Q99ZW2, Streptococcus pyogenes serotype M1) with GFP-Myc tag) labelling CRISPR-Cas9 (red) with ab189380 at dilution of 1/70. The secondary antibody used was Alexa Fluorr® 488 goat-anti-rabbit IgG (1/2000). Cells were fixed with 4% paraformaldehyde. Isotype control antibody was (ab172730) Rabbit monoclonal IgG (black).



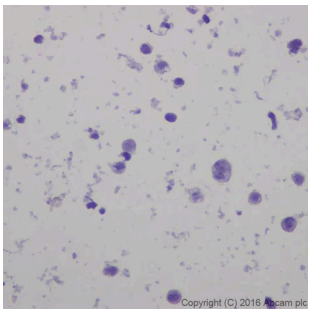
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380)

CRISPR-Cas9 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CRISPR-Cas9 antibody

Immunohistochemical analysis of paraffin-embedded Human stomach tissue labeling CRISPR-Cas9 with ab189380 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.
No staining on Human stomach is observed.

Counter stained with Hematoxylin.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

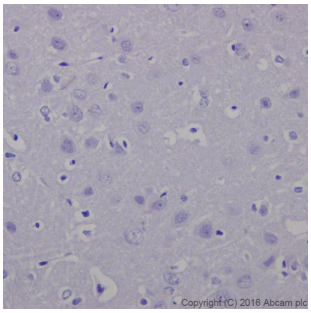


Immunocytochemistry - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380)

CRISPR-Cas9 Immunocytochemistry staining using rabbit Anti-CRISPR-Cas9 antibody

Immunocytochemical analysis of agarose-embedded sections of 293T (Human epithelial cell line from embryonic kidney) cells transfected with blank pcDNA3.1(+)-GFP-Myc vector labeling CRISPR-Cas9 with ab189380 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.
Negative on 293T cells transfected with blank pcDNA3.1(+)-GFP-Myc vector.

Counter stained with Hematoxylin.



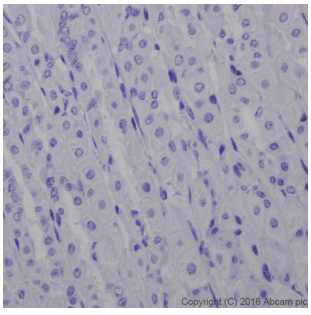
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380)

CRISPR-Cas9 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CRISPR-Cas9 antibody

Immunohistochemical analysis of paraffin-embedded Rat cerebrum tissue labeling CRISPR-Cas9 with ab189380 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.
No staining on rat cerebrum is observed.

Counter stained with Hematoxylin.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



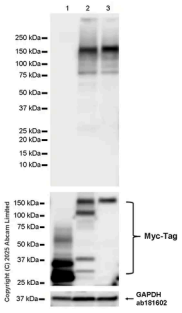
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380)

CRISPR-Cas9 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-CRISPR-Cas9 antibody

Immunohistochemical analysis of paraffin-embedded Mouse stomach tissue labeling CRISPR-Cas9 with ab189380 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.
No staining on mouse stomach is observed.

Counter stained with Hematoxylin.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Western blot - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380)

CRISPR-Cas9 Western blot staining using rabbit Anti-CRISPR-Cas9 antibody

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

All lanes:
Western blot - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380) at 1/1000 dilution

Lane 1:
HEK-293 (Human epithelial cell line from embryonic kidney) cells transfected with an empty vector containing a Myc-Tag, whole cell lysate at 20 µg

Lane 2:
HEK-293 cells transfected with CRISPR-Cas9 (Q99ZW2, Streptococcus pyogenes serotype M1) containing a Myc-Tag, whole cell lysate at 20 µg

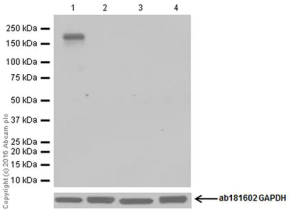
Lane 3:
HEK-293 cells transfected with Cas9/Csn1 point mutation (D10A and H840A) [dCas9] containing a Myc-Tag, whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 158 kDa

Exposure time: 4s



Western blot - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380)

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380) at 1/20000 dilution

Lane 1:
HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate transfected with CRISPR-Cas9 (Q99ZW2, Streptococcus pyogenes serotype M1) with GFP-Myc tag at 20 µg

Lane 2:
HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 20 µg

Lane 3:
NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell lysate at 20 µg

Lane 4:
Rat embryo lysate at 20 µg

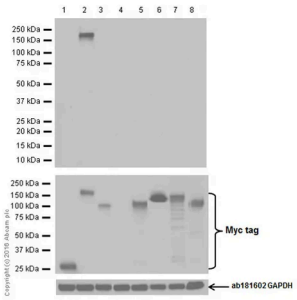
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 158 kDa

Observed band size: 184 kDa

Exposure time: 10s



Western blot - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380)

CRISPR-Cas9 Western blot staining using rabbit Anti-CRISPR-Cas9 antibody

Blocking/Dilution buffer: 5% NFDM/TBST.
All lanes:
Western blot - Anti-CRISPR-Cas9 antibody [EPR18991] (ab189380) at 1/20000 dilution

Lane 1:
Empty vector with GFP-Myc tag (vector control) transfected HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate at 20 µg

Lane 2:
HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate transfected with CRISPR-Cas9 (Q99ZW2, Streptococcus pyogenes serotype M1) with GFP-Myc tag at 20 µg

Lane 3:
HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate transfected with CRISPR-Cas9 (G3ECR1, Streptococcus thermophilus, N–terminal aa1-800) with GFP-Myc tag at 20 µg

Lane 4:
Empty vector with Myc-His tag (vector control) transfected HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate at 20 µg

Lane 5:
HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate transfected with CRISPR-Cas9 (A1IQ68, Neisseria meningitidis serogroup A / serotype 4A (strain Z2491)) with Myc-His tag at 20 µg

Lane 6:
HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate transfected with CRISPR-Cas9 (G3ECR1, Streptococcus thermophilus) with Myc-His tag at 20 µg

Lane 7:
HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate transfected with CRISPR-Cas9 (Q03JI6, Streptococcus thermophilus (strain ATCC BAA-491 / LMD-9)) with Myc-His tag at 20 µg

Lane 8:
HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate transfected with CRISPR-Cas9 (J7RUA5, Staphylococcus aureus subsp. aureus) with Myc-His tag at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 158 kDa

Observed band size: 184 kDa

Exposure time: 3s

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.