abcam

Product datasheet

Anti-CRISPR-Cas9 antibody [KANI345B] ab271293

Recombinant

★★★★★ 1 Abreviews 8 Images

Overview

Product name Anti-CRISPR-Cas9 antibody [KANI345B]

Description Rat monoclonal [KANI345B] to CRISPR-Cas9

Host species Rat

Tested applications Suitable for: WB, IHC-P, ICC

Species reactivity

Predicted to work with: Streptococcus pyogenes

A

Immunogen Recombinant full length protein. This information is considered to be commercially sensitive.

Positive control WB: HEK-293 transfected with CRISPR-Cas9 (Q99ZW2, Streptococcus pyogenes serotype M1)

with GFP-Myc tag, whole cell lysate. IHC-P: HEK-293T transfected with a GFP-Myc-tagged CRISPR-associated endonuclease Cas9/Csn1 construct. ICC: HEK-293T cells transfected with

GFP-tagged Cas9 expression vector.

General notes

This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or

conjugation for your experiments, please contact orders@abcam.com.

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.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer Preservative: 0.01% Sodium azide

Constituents: 59.04% PBS, 0.05% BSA, 40% Glycerol (glycerin, glycerine)

Purity Protein A purified

Clonality Monoclonal

1

Clone number

KANI345B

Isotype

lgG2a

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab271293 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

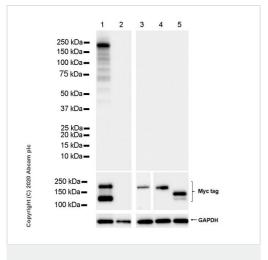
Application	Abreviews	Notes
WB		1/5000.
IHC-P	*****(1)	1/200. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC		1/50.

Target

Relevance

[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). 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) (Probable). 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. Subsequently Cas9/crRNA/tracrRNA endonucleolytically cleaves linear or circular dsDNA target complementary to the spacer. The target strand not complementary to crRNA is first cut endonucleolytically, then trimmed by 3'-5' exonucleolytically. DNA-binding requires protein and both RNA species. Cas9 probably recognizes a short motif in the CRISPR repeat sequences (the PAM or protospacer adjacent motif) to help distinguish self versus nonself.

Images



Western blot - Anti-CRISPR-Cas9 antibody [KANI345B] (ab271293)

All lanes : Anti-CRISPR-Cas9 antibody [KANI345B] (ab271293) at 1/5000 dilution

Lane 1: HEK-293 (human embryonic kidney epithelial cell) transfected with CRISPR-Cas9 (Q99ZW2, Streptococcus pyogenes serotype M1) with GFP-Myc tag, whole cell lysate

Lane 2: HEK-293T transfected with an empty vector (vector control), containing a myc-His-tag®, whole cell lysate

Lane 3: HEK-293 transfected with CRISPR-Cas9 (G3ECR1, Streptococcus thermophilus) with Myc-His tag, whole cell lysate

Lane 4: HEK-293 transfected with CRISPR-Cas9 (Q03Jl6, Streptococcus thermophilus) with Myc-His tag, whole cell lysate

Lane 5: HEK-293 transfected with CRISPR-Cas9 (J7RUA5, Staphylococcus aureus subsp. aureus) with Myc-His tag, whole cell lysate

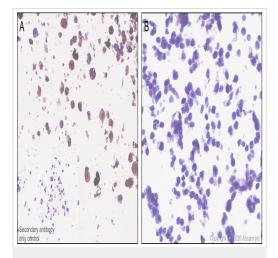
Lysates/proteins at 5 µg per lane.

Secondary

All lanes : Peroxidase-Conjugated Goat anti-Mouse IgG (H+L) at 1/10000 dilution

Exposure time: 1 second

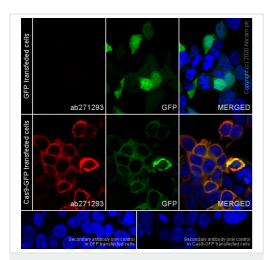
Blocking/Dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CRISPR-Cas9 antibody [KANI345B] (ab271293)

Immunohistochemical analysis of paraffin-embedded (Panel A) HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) transfected with a GFP-Myctagged CRISPR-associated endonuclease Cas9/Csn1 construct and (Panel B) HEK-293T (human epithelial cell line from embryonic kidney transformed with large T antigen) transfected with empty plasmid, labeling CRISPR-Cas9 with ab271293 at 1/200 (4.58 ug/ml) dilution followed by a ready to use Goat Anti-Rat lgG H&L (HRP polymer) (ab214882). Counterstained with Hematoxylin. Positive staining on (A) HEK-293T transfected with a GFP-Myctagged CRISPR-associated endonuclease Cas9/Csn1 construct, no staining on (B) HEK-293T transfected with empty plasmid.

Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rat lgG H&L (HRP polymer) (ab214882).

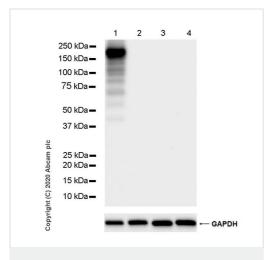


Immunocytochemistry - Anti-CRISPR-Cas9 antibody [KANI345B] (ab271293)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized 293T+OE-94 cells labelling CRISPR-Cas9 with ab271293 at 1/50 dilution (18.32 ug/ml), followed by ab150160 Goat Anti-Rat lgG H&L (Alexa Fluor® 594) antibody at 1/1000 dilution (2 ug/ml) (Red). The nuclear counterstain was DAPI (Blue).

Confocal image showing cytoplasmic staining in 293T cells transfected with GFP-tagged Cas9 expression vector.

Secondary antibody only control: Secondary antibody is ab150160
Goat Anti-Rat lgG H&L (Alexa Fluor® 594) antibody at 1/1000 dilution (2 ug/ml).



Western blot - Anti-CRISPR-Cas9 antibody [KANI345B] (ab271293)

All lanes : Anti-CRISPR-Cas9 antibody [KANI345B] (ab271293) at 1/5000 dilution

Lane 1: HEK-293 (human embryonic kidney epithelial cell) transfected with CRISPR-Cas9 (Q99ZW2, Streptococcus pyogenes serotype M1) with GFP-Myc tag, whole cell lysate

Lane 2 : HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 3: NIH/3T3 (mouse embryonic fibroblast) whole cell lysate

Lane 4: C6 (rat glial tumor glial cell), whole cell lysate

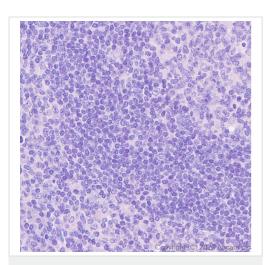
Lysates/proteins at 5 µg per lane.

Secondary

All lanes : Peroxidase-Conjugated Goat anti-Mouse IgG (H+L) at 1/10000 dilution

Exposure time: 1 second

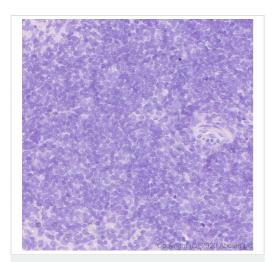
Blocking/Dilution buffer: 5% NFDM/TBST.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CRISPR-Cas9 antibody [KANI345B] (ab271293)

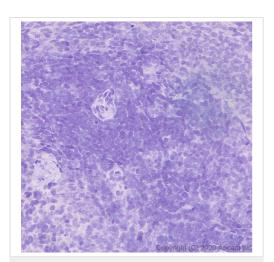
Immunohistochemical analysis of paraffin-embedded human tonsil tissue labeling CRISPR-Cas9 with ab271293 at 1/200 (4.58 ug/ml) dilution followed by a ready to use Goat Anti-Rat lgG H&L (HRP polymer) (ab214882). Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rat lgG H&L (HRP polymer) (ab214882). Heat mediated antigen retrieval using ab93678 (citrate buffer, pH 6.0).

Negative control: No staining on the human tonsil.



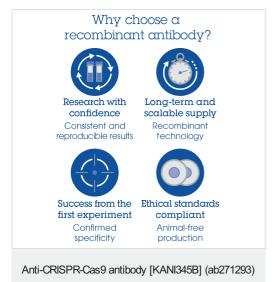
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CRISPR-Cas9 antibody [KANI345B] (ab271293)

Immunohistochemical analysis of paraffin-embedded mouse spleen tissue labeling CRISPR-Cas9 with ab271293 at 1/200 (4.58 ug/ml) dilution followed by a ready to use Goat Anti-Rat lgG H&L (HRP polymer) (ab214882). Counterstained with Hematoxylin. Heat mediated antigen retrieval using ab93678 (citrate buffer, pH 6.0). Negative control: No staining on the mouse spleen.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-CRISPR-Cas9 antibody [KANI345B] (ab271293)

Immunohistochemical analysis of paraffin-embedded rat spleen tissue labeling CRISPR-Cas9 with ab271293 at 1/200 (4.58 ug/ml) dilution followed by a ready to use Goat Anti-Rat lgG H&L (HRP polymer) (ab214882). Counterstained with Hematoxylin. Heat mediated antigen retrieval using ab93678 (citrate buffer, pH 6.0). Negative control: No staining on the rat spleen.



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