

Datasheet for 600-401-GK0S**Cas 9 Antibody****Overview**

Description:	Anti-Cas9 (RABBIT) Antibody - 600-401-GK0S
Item No.:	600-401-GK0S
Size:	25 µL
Applications:	ELISA, IF, Multiplex, WB
Reactivity:	<i>S. pyogenes</i> serotype M1
Host Species:	Rabbit

Product Details

Background:	The Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) and CRISPR Associated (Cas) system was discovered in bacteria as a defense against foreign DNA, even being believed to act as a sort of prokaryotic immune system. The Cas9 protein is becoming a useful tool in the field of genomic editing for its ability to induce site-directed double strand breaks in DNA. It can cleave almost any sequence complementary to the guide RNA and has recently been used for modifying the genome of human embryos for the first time.
Synonyms:	rabbit anti-Cas9 antibody, Cas-9, Cas 9, CRISPR, CRISPR-associated endonuclease Cas9/Csn1, SpCas9, SpyCas9, csn1, Cas-9 Antibody
Host Species:	Rabbit
Clonality:	Polyclonal
Format:	IgG

Target Details

Gene Name:	cas9
Reactivity:	<i>S. pyogenes</i> serotype M1
Immunogen Type:	Conjugated Peptide
Immunogen:	Cas-9 affinity purified antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to a region near the C-terminal of <i>Streptococcus pyogenes</i> Cas-9 protein.

Purity/Specificity:	Anti-Cas9 antibody is directed against <i>Streptococcus pyogenes</i> Cas-9. The product was affinity purified from monospecific antiserum by immunoaffinity chromatography.
Relevant Links:	• GeneID - 901176 • NCBI - NP_269215.1 • UniProtKB - Q99ZW2

Application Details

Tested Applications:	ELISA, IF, Multiplex, WB
Application Note:	This affinity purified antibody has been tested for use in ELISA, Immunofluorescence, and Western blotting. Specific conditions for reactivity should be optimized by the end user. Expect a band ~158 kDa in size corresponding to Cas-9 protein by western blotting in the appropriate cell lysate or extract.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:25,000-1:30,000
IF:	1µg/mL
WB:	1µg/mL
Other:	User Optimized

Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	1.0 mg/mL by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	None

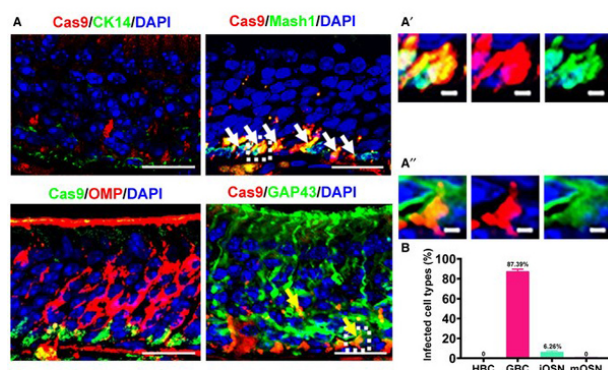
Shipping & Handling

Shipping Condition:	Dry Ice
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Storage Condition: Store vial at -20° C or below prior to opening. This vial contains a relatively low volume of reagent (25 µL). To minimize loss of volume dilute 1:10 by adding 225 µL of the buffer stated above directly to the vial. Recap, mix thoroughly and briefly centrifuge to collect the volume at the bottom of the vial. Use this intermediate dilution when calculating final dilutions as recommended below. Store the vial at -20°C or below after dilution. Avoid cycles of freezing and thawing.

Expiration: Expiration date is one (1) year from date of receipt.

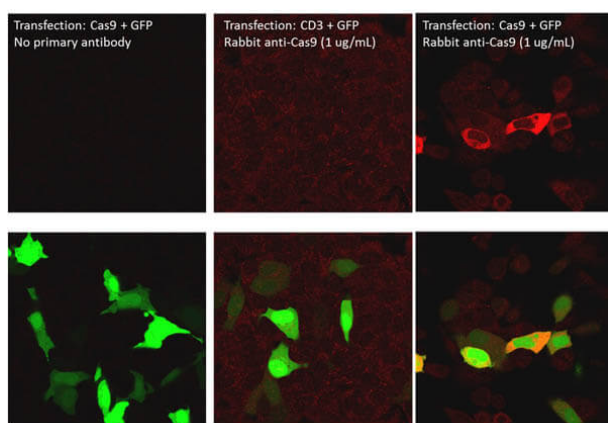
Images



Immunofluorescence Microscopy

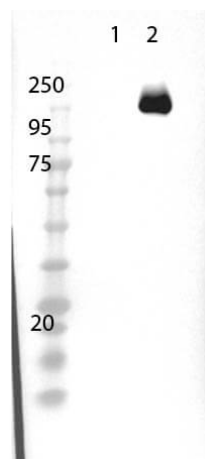
A) Representative IF costaining with Cas9 antibody and antibodies for cell markers (CK14, Mash1, GAP43 and OMP) in the MOE of the mice injected with AAV for 8 weeks. The white arrows indicate the cells positive for both Cas9 and Mash1, and the yellow arrows indicate the cells positive for both Cas9 and GAP43. (A') and (A'') are shown at higher magnification of the boxed region, as three-channel merged images (left image) and the separated channel (middle and right images). (A): Scale bars, 20 µm; (A') and (A''): Scale bars, 2 µm.

B) The ratio of Cas9+ cells of the cells of (CK14+ + Mash1+ + GAP43+ + OMP+) cells in the MOE of the mice injected with AAV for 8 weeks (n = 4 mice; data represent the mean ± SEM; Student's t-test). Figure EV1. PMID: 32677323



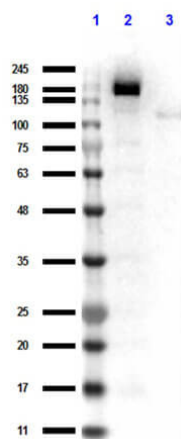
Immunofluorescence Microscopy

Immunofluorescence of Rabbit Anti-Cas9 Antibody. Cells: HeLa cells transfected with GFP+Cas9 or GFP+CD-3. Fixation: Formaldehyde fixed, permeabilized Triton X-100. Primary: Anti-Cas9 used at 1µg/mL. Secondary: Donkey anti-Rabbit at 1µg/mL, staining seen in red. Bottom images show GFP stain overlay.



Western Blot

Western Blot of Rabbit Anti-Cas9 Antibody. Lane 1: 293T non transfected cell lysate. Lane 2: 293T Cas9 over expressed cell lysate. Load: 15µg per lane. Primary Antibody cas9 used at 1µg/mL using MB-070 overnight at 4°C. Secondary Antibody: goat anti-rabbit HRP at 1:40,000 for 30 min at room temp. Expect: 158kDa.



Western Blot

Western Blot results of Rabbit Anti-Cas 9 Antibody. Lane 1: Opal Prestained Molecular Weight Ladder (p/n MB-210-0500). Lane 2: HEK/293 Cas9 Over Expressing WCL. Lane 3: HEK/293 non Transfected WCL. Load: 10ul. Primary Antibody: Rabbit Anti-Cas 9 Antibody at 1µg/mL overnight at 4°C. Secondary Antibody: Goat anti-Rabbit HRP (p/n 611-103-122) at 1:70,000 for 30min at RT. Blocking: BlockOut (p/n MB-073) for 30 min at RT.

References

- Alonso-Lerma B et al. Evolution of CRISPR-associated endonucleases as inferred from resurrected proteins. *Nat Microbiol.* (2023)
- Yang D et al. The microRNA/TET3/REST axis is required for olfactory globose basal cell proliferation and male behavior. *EMBO Rep.* (2020)

Disclaimer

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