

AbFlex® Cas9 antibody (rAb)

Catalog Nos: 91123, 91124

RRID: AB_2793783

Application(s): ELISA, WB

Reactivity: Human

Quantities: 100 µg, 10 µg

Purification: Ni-NTA

Host: Mouse

Isotype: IgG2a

Concentration: 1 µg/µl

Molecular Weight: 160 kDa

Background: AbFlex® antibodies are recombinant antibodies (rAbs) that have been generated using defined DNA sequences to produce highly specific, reproducible antibodies. Each AbFlex antibody contains a 6xHis Tag, a Biotinylation Tag for enzymatic biotin conjugation using the biotin ligase, BirA, and a sortase recognition motif (LPXTG) to attach a variety of labels directly to the antibody including fluorophores, enzymatic substrates (HRP, AP), peptides, drugs as well as solid supports.

AbFlex® Cas9 antibody was expressed as full-length IgG with mouse immunoglobulin heavy and light chains (IgG2a isotype) in mammalian 293 cells.

Cas9 is a nuclease from *Streptococcus pyogenes* that can be targeted to particular DNA sequences through a guide RNA that results in double-stranded breaks in DNA. Cas9 is part of the CRISPR/Cas9 gene-editing system that can create a DNA break at a specific location with the genome.

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.

Immunogen: This antibody was raised against a recombinant protein within the N-terminal region of *Streptococcus pyogenes* Cas9. This antibody recognizes Cas9 and dCas9.

Buffer: Purified IgG in 140 mM Hepes, pH 7.5, 70 mM NaCl, 32 mM NaOAc, 30% glycerol and 0.035% sodium azide. Sodium azide is highly toxic.

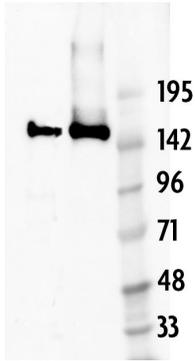
Application Notes:

Validated Applications:

Bead-based ELISA: 9 - 75 ng/ml

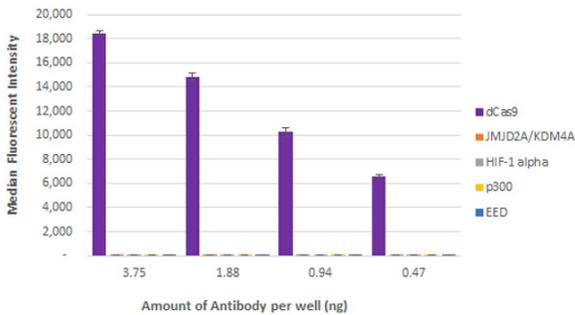
Storage and Guarantee: Some products may be shipped at room temperature. This will not affect their stability or performance. Upon receipt, unconjugated antibodies may be stored at -20°C for up to 2 years. Fluorophore- & enzyme-conjugated antibodies should be stored at 4°C. Fluorophore-conjugated antibodies should be protected from light. Keep reagents on ice when not in storage; to avoid repeated freeze/thaw cycles, we recommend aliquoting items that will be stored frozen into single-use fractions prior to freezing. This product is guaranteed for 6 months from date of receipt.

This product is for research use only and is not for use in diagnostic procedures.



AbFlex® Cas9 antibody (rAb) tested by Western blot.

50 ng recombinant Cas9 protein was probed with 0.5 µg/ml AbFlex® Cas9 antibody (Lane 1). HEK293T cells were transfected with a plasmid containing a mammalian expression construct for dCas9 (*S. pyogenes*). Chromatin was prepared 48 hours post-transfection, boiled and 15 µl (~200,000 cell equivalents) was run on a SDS-PAGE gel and probed with 2 µg/ml AbFlex® Cas9 antibody (Lane 2). A molecular weight marker was run in Lane 3.



AbFlex® Cas9 antibody (rAb) tested by bead-based specificity analysis.

Luminex bead-based specificity analysis was used to confirm the specificity of AbFlex® Cas9 antibody (rAb) antibody for Cas9. Various proteins were conjugated to MagPlex Luminex beads and incubated with various amounts of AbFlex® Cas9 antibody (rAb). Protein-bound antibody was detected with anti-mouse IgG-Phycoerythrin and read in a Luminex instrument.

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